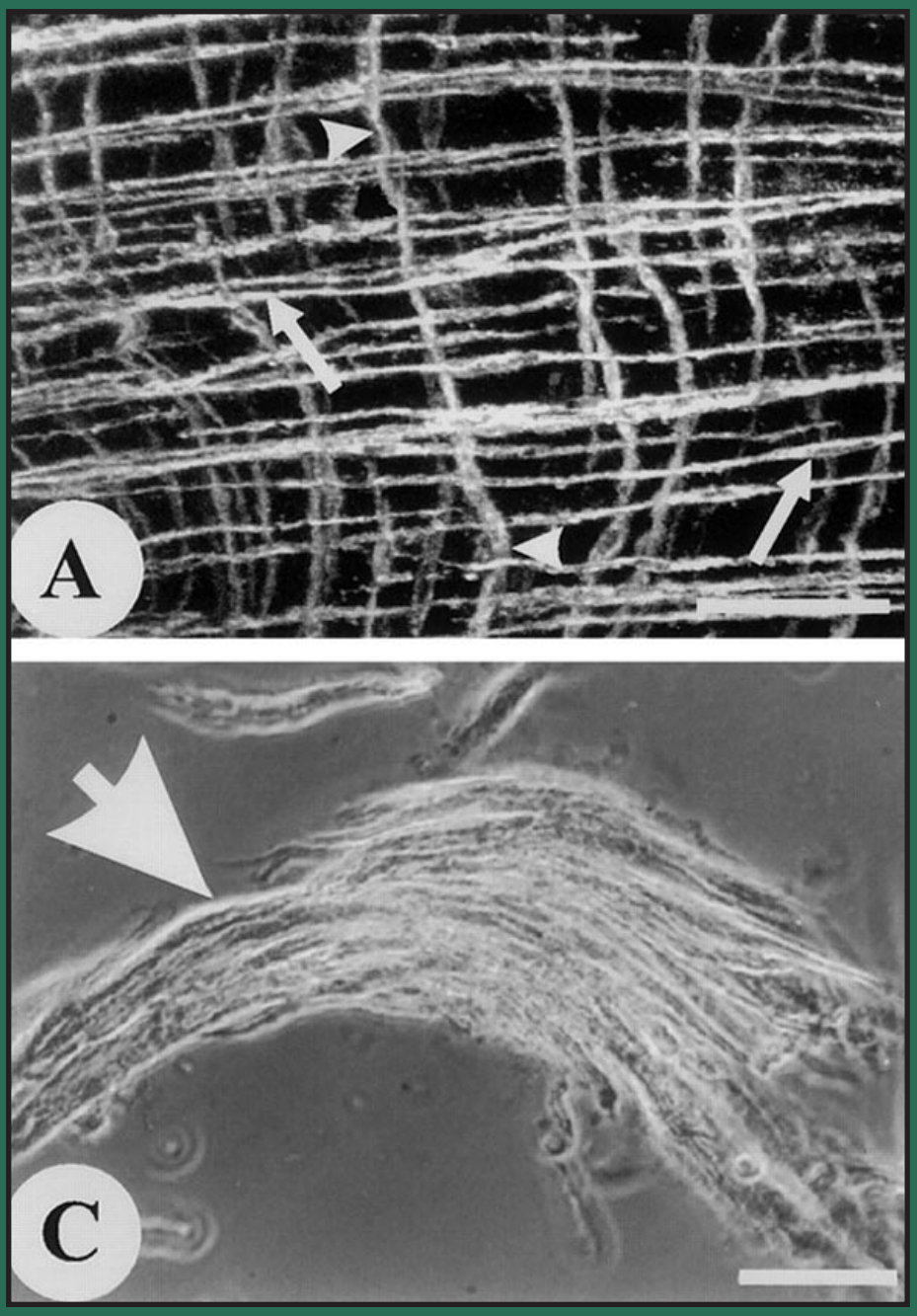

Fluorescent and phase-contrast images of mouse stomach indicating the presence of a protein called Kit, which is found in a particular cell type involved in gastrointestinal motility known as interstitial cells of Cajal.

Image courtesy of Dr. Kenton Sanders. Used with permission of the American Physiological Society. Am J Physiol Cell Physiol *279:C529-C539, 2000.*

Functional Gastrointestinal Disorders and Motility Disorders

SUMMARY OF RESEARCH GOALS

Functional gastrointestinal (GI) disorders and motility disorders, such as irritable bowel syndrome (IBS), functional dyspepsia, and gastroesophageal reflux disease (GERD), take a significant toll on the health and well-being of many Americans. The Commission offers several research goals designed to improve our understanding of normal motility and secretory activities of the GI tract, discover the physiologic changes that lead to disease, and develop more effective therapies to prevent, treat, or reverse these disorders. Research efforts are needed on the numerous systems and processes that may be impaired in functional GI and motility disorders, including brain-gut interactions, the enteric nervous system, interstitial cells of Cajal and smooth muscle cells, pain and sensory mechanisms, the gut mucosa and musculature, the intestinal microflora, and immune and inflammatory responses. It is important to define how factors such as genetic differences, age, sex, and gender influence a person's susceptibility to these disorders. Many individuals with diabetes develop GI motility disorders, such as gastroparesis and constipation. As the rate of diabetes continues to rise in the U.S., research on how diabetes affects the GI tract is increasingly important. Ultimately, research to discover the basic mechanisms of disease must be translated into new technologies, pharmacological therapies, and behavioral strategies to effectively treat all patients afflicted with functional GI and motility disorders.

INTRODUCTION AND BACKGROUND

Functional GI and motility disorders represent common conditions seen by both primary care specialists and gastroenterologists. A functional GI disorder is characterized clinically by a constellation of symptoms that may include physiologic dysfunction such as altered GI motility and secretion, visceral hypersensitivity, and brain-gut dysregulation. A motility disorder is defined by observable disturbances in neuromuscular functioning of the enteric nervous system (ENS) and the muscularis. There is significant overlap between what have been traditionally categorized as functional disorders or motility disorders. In order to develop a more visionary research effort, some investigators believe that a new paradigm should be established that moves away from the traditional compartmentalization of functional and motility disorders and develops a more encompassing and biomechanistic framework for these diseases. The traditional delineation of "functional" versus "organic" in this area of clinical medicine is likely to be unsustainable as the pathophysiology of these diseases is discovered.

These conditions have significant impact on the American public because of their high prevalence and negative effects on quality of life, high direct and indirect costs, and the devastating consequences of the rare forms of these diseases. Irrespective of the definition or criteria applied to study their prevalence, both IBS and functional dyspepsia show prevalence rates ranging from 6-30 percent of the U.S. population. GERD, which results from disordered competence of the motor functions of the lower esophageal sphincter, affects 14-29 percent of people at least weekly and results in medication costs of $8 billion or more annually. Some patients require surgery which may be unsuccessful in at least 10 percent of patients. Estimates for prevalence of constipation range from 3-19 percent of the U.S. population with a slightly lower prevalence of diarrhea. In a burden of illness study published in 2002, GERD, IBS, and chronic diarrhea had the highest prevalence rates and each had a very significant economic burden. With respect to claims data, abdominal pain is the most frequent GI symptom reported and results in significant healthcare utilization. Thus, GI motility and sensory abnormalities result in disorders of high prevalence and constitute frequent reasons for physician consultation.

Additionally, there is a series of motility disorders whose prevalence is lower, but the impact of these disorders can be deleterious, devastating, or life-threatening. These conditions include gastroparesis (slow emptying of the stomach), chronic intestinal pseudoobstruction, Hirschsprung's disease, megacolon, and fecal incontinence. The first four appear to result from significant neuromuscular impairment, leading to impaired ability to maintain hydration, nutrition, appropriate digestion, and excretion from the gut. Fecal incontinence constitutes a significant social burden that may result in stigma, social isolation, and impaired quality of life, particularly for women.

Pathogenesis and natural history: Functional GI and motility disorders are heterogeneous and defined largely by symptoms or pathophysiologic changes. The etiologies of these disorders are not well understood. Research into the pathologic mechanisms and etiology of functional GI and motility disorders is expected to lead to major changes in the current classifications of these disorders. Dependent upon the disorder, presentation may range from altered GI motor activity, including abnormal sphincter function and/or transit (resulting in nausea, vomiting, loss of appetite, constipation, or diarrhea), to a state of heightened visceral nociception (the perception of pain, discomfort, or bloating in the gut), or a combination of both motor and

sensory symptoms. Factors that predispose patients to develop hypersensitivity may be both peripherally and centrally mediated. In animal models and human diseases, gut inflammation, gut injury, altered gut mucosal immunity, psychiatric conditions, or psychosocial factors all may modulate the reciprocal pathways between the brain and gut, inducing states of hypersensitivity. Factors leading to abnormal motor function may include reduced inhibitory neuronal activity, heightened cholinergic neural activity, loss of pacemaker cells, or abnormal responses to endocrine mediators. The specific alteration in mediators is known for only a few disorders.

Current means of control, cure, and/or prevention: The range of therapeutic agents available for the treatment of functional GI disorders and motility disorders remains limited, with no agents that can cure or prevent the disorders. The lack of agents that can cure or prevent these disorders reflects the very limited understanding of their pathologic mechanisms and etiology and, perhaps, an inappropriate grouping of patients for testing of therapies. Agents that generically speed or slow motility, or tighten or loosen sphincter tone, are used across a variety of disease states. As a class, dopamine antagonists and 5HT4 agonists function as gastroprokinetic-type agents (i.e., drugs that stimulate stomach emptying), while anticholinergics and calcium channel blockers are antispasmodics (i.e., drugs that suppress smooth muscle activity). Erythromycin has been found to have only limited use as a gastroprokinetic, even though it has a degree of motilin-like binding activity. Alosetron is a 5HT3 receptor antagonist indicated for the treatment of severe, diarrhea-predominant, female IBS patients; tegaserod, a 5HT4 receptor agonist, is indicated for female, constipation-predominant IBS patients. However, these agents are only indicated for female patients with severe disease unresponsive to first line symptomatic remedies, and both have significant labeled safety warnings. Some data suggest a role for anti-depressants in the treatment of IBS, although confirmatory, contemporary efficacy studies have either not been performed or did not demonstrate efficacy in the intent-to-treat cohort. Antibiotics and probiotics may relieve bloating, but have not been fully established as treatments for other IBS symptoms.

Whether antibiotic prophylaxis will be a successful strategy to prevent post-infectious IBS remains to be determined. Several classes of agents are in development for various functional and motility-related disorders aimed at addressing either altered states of motility or visceral hypersensitivity: corticotropin-releasing factor (CRF) antagonists, novel motilin agonists, NK antagonists, atypical benzodiazepines, kappa-opiate agonists, mu-opiate antagonists/agonists, chloride channel openers/closers, beta agonists, guanylate cyclase C agonists, other agents that act on the serotonin system, cannabinoids, and mast cell stabilizers. The range of medications being tested reflects the complexity of the control mechanisms, the redundancy of the neurohormonal mediators, and the need to more clearly understand the pivotal mechanisms underpinning the development of these syndromes.

To fully understand these diseases and develop safe and effective therapies, a major research effort is underway to address the basic mechanisms underlying the neuromuscular control, including studies of extrinsic and intrinsic nerves that control the gut, pacemaker cells and muscular mechanisms, the development of the neuromuscular apparatus, the molecular and genetic disorders associated with human disease, the afferent mechanisms that convey sensations of pain and nausea to the conscious brain, and the reflex pathways that peripherally modulate those sensations to protect the conscious brain. In addition, the social, psychological, and behavioral aspects of these diseases

are being explored, and researchers are looking for new ways to enhance disease management by improving the delivery of health care, optimizing the doctor-patient relationship, and developing medications and devices that can restore normal function or provide means to support use of the gut for nutrition, normal bowel function, and continence.

RECENT RESEARCH ADVANCES

The brain-gut axis and neurohormonal control of motor and sensory functions of the GI tract

Specific advances include an understanding that serotonin activates intrinsic and extrinsic primary afferent neurons to promote peristaltic and secretory reflexes and to modulate sensory signaling in the brain-gut axis. Identification of different serotonin receptor subtypes in the GI tract has allowed development of drugs to modulate GI motility, secretion, and sensation. Polymorphisms in the promoter region of the gene encoding the serotonin reuptake transporter (SERT) may have a role in patients with diarrhea-predominant IBS. This new understanding of GI functional mechanisms has facilitated the development of novel pharmacologic treatments for GI motility disorders, functional GI disorders, and obesity. Treatments and potential new targets for intervention include: 5HT3 receptor antagonists, 5HT4 receptor agonists, CRF antagonists, ghrelin, neurokinins, nitric oxide and other gas neurotransmitters, mu-opioid receptor modulation, cannabinoids, and mast cell stabilizers.

Role of the immune system and inflammation in GI diseases

A growing understanding of the nature and complexity of the interactions between the central nervous system (CNS), the ENS, and the immune system has led to the recognition that the arbitrary division of gut disorders as inflammatory disorders or functional disorders may be misleading. Immune activation appears to be common in GI motility disorders, although a lack of common terminology and methodology has limited comparisons between sub-groups. Immune methodology, including genetic studies, is now widely available and can be applied to the field of GI motility disorders. Abnormal physiology may be triggered by an insult or perturbation, such as an infection, and result in long-term alterations in ENS or CNS responses to subsequent stimuli. Prospective evaluations of post-infectious IBS have shown that up to 20 percent of individuals with bacterial gastroenteritis may develop symptoms of IBS or dyspepsia. Infection has been shown in epidemiological studies to confer an increased risk for development of IBS. Predictive factors show two important features relative to the pathogenesis of this disorder: (a) association of mucosal inflammation and altered mucosal immunity; and (b) association of psychosocial disturbance at time of infection. Mast cells and enterochromaffin cells have been found to be markers of mucosal immune activation in IBS in humans and in animal models. Mast cell hyperplasia is a common finding in mucosal biopsies from the large intestine of IBS patients. Changes in the mucosal immune system may also increase the excitability of neurons involved in local reflexes and central pathways in response to food antigens and chemical stimuli. Such insults to the intestine may increase mucosal permeability, rendering the mucosa more susceptible to luminal antigens and aggravating the inflammatory state.

Altered bacterial flora in functional GI disorders (including post-infectious IBS)

Luminal bacterial microflora can affect mucosal inflammation and immune function leading to neural sensitization. A rapidly growing body

of evidence suggests that important signaling pathways exist between the microflora and the gut (including signaling to enterochromaffin, immune, and nerve cells), which may contribute to normal homeostasis and may be altered in disease states such as inflammatory bowel diseases (IBD), IBS, and possibly obesity. Research on the effects of "good" and "bad" bacteria on mucosal immune function suggests the possible benefits of probiotic bacteria to either prevent or treat these conditions. Further studies are needed to determine the patient subset that might be most responsive to this type of treatment.

Interaction of stress circuitry and gut function

Researchers have shown that dysregulation of stress circuitry can affect gut function. Evidence for this link includes: (a) increased corticotropin-releasing hormone (CRH) and adrenocorticotropic hormone (ACTH) reactivity to stress and increased motor and pain responses to CRH, which can be blocked by CRH antagonists; (b) increased mucosal immune activation; (c) stress-associated disruption of intestinal mucosal barrier function (i.e., increased membrane permeability, mast cell activation, and mucosal inflammation, which may lead to visceral sensitization); and (d) altered limbic system (i.e., anterior cingulate cortex) reactivity to visceral signals leading to increased pain response in functional GI disorders. The latter is enhanced by stress.

Immune response to the enteric nervous system

Autoimmune responses targeted to neuronal elements of the ENS can underlie a variety of conditions from IBS-like symptoms to chronic pseudoobstruction. Enteric inflammatory neuropathy disrupts the integrative functions of the brain-in-the-gut, including reduction in the population of inhibitory motor neurons to the musculature. Extreme loss of inhibitory motor neurons or disconnection of motor neurons with post-junctional cells manifests as disinhibitory motor disease, such as achalasia or pylorospasm in the smooth muscle sphincters, and hyperactive, disorganized contractile behavior of intestinal muscle cells, which results in pseudoobstruction. Detection of anti-enteric neuronal antibodies in the serum of patients with early symptoms of a functional GI or motility disorder may be a useful diagnostic test for inflammatory enteric neuropathy, including paraneoplastic disease associated with small cell lung cancer. Other diseases, including diabetes, cause an autoimmune-mediated visceral neuropathy probably affecting extrinsic and intrinsic nerves.

Neural stem cells (neural crest stem cells) in the gut

Researchers have discovered that neural crest stem cells persist in the gut after birth. Stem cells from other sources also show potential for giving rise to functional neurons in the GI tract. The field of directed stem cell therapy is developing and holds some promise for future clinical applications.

Development of the ENS

The molecular genetics of multiple endocrine neoplasia type 2B and Hirschsprung's disease—prototypic disorders characterized by gross and/or microscopic pathology of the ENS and associated dysmotility—have been identified. Intercellular signaling pathways involved in enteric neurodevelopment have been characterized. Mutations associated with abnormalities of neuroenteric development are known in animal models and in a spectrum of human disease processes, including:

aganglionosis, absence of specific neuronal cell populations, apoptosis of neurons, absence of interstitial cells of Cajal (ICC), and dysfunctional intestinal smooth muscle.

Role of the human brain in the perception and modulation of visceral afferent signals from the GI tract

Neuroimaging techniques have enabled research on the neurobiological mechanisms within the human brain that are involved in perception of pain. Brain regions and networks involved in the processing and in the modulation of visceral afferent signals have been determined. Cortico-limbic interactions in symptom modulation may link the large body of psychosocial information gathered in epidemiological studies with neurobiological substrates. With novel imaging techniques targeting neurochemistry as well as neuroanatomical substrates, it may be possible to monitor and guide drug development first in animal models and then in humans and to understand the ramifications of genetics on differential responses to anxiety and emotional triggers.

Role of the peripheral nervous system (autonomic and enteric nerves) in functional GI and motility disorders

Research has demonstrated novel findings about the organization and function of the enteric and autonomic nervous systems and the complex integration of information between structurally discrete groups of neurons. Integration of sensory information and the mechanisms of chemical and mechanosensitivity in the GI tract have been uncovered. It has long been appreciated that patients with IBS exhibit generalized digestive tract hypersensitivity and some have somatic hypersensitivity in organs such as the bladder. This could arise from peripheral and/or central mechanisms. Because various visceral afferent inputs converge onto the same spinal cord neurons, it has been assumed that cross-sensitization arises via a central mechanism. However, recent findings suggest that a single visceral sensory neuron can give rise to axons that innervate different organs. The specialized dorsal column pain pathway requires further elucidation in the context of functional GI disorders.

Role of interstitial cells of Cajal (ICC) in GI motility and disease

ICC are pacemaker cells, conduits for propagation and coordination of electrical activity, regulators of the basal level of excitability of the musculature, mediators of inputs from motor neurons, and stretch receptors. Other actions, such as a modulatory role of sensory nerve terminals, have been proposed but not yet fully documented. Loss of these cells in a variety of functional GI and motility disorders provides exciting new hypotheses for the etiology of disorders such as gastroparesis, pseudoobstruction, sphincter dysfunction, and colonic inertia.

Use of animal models of functional GI and motility disorders

Animal models developed to study functional GI and motility disorders have provided new insights into the pathologic mechanisms of these disorders. Several genetic and laboratory-induced animal models of diabetes have allowed evaluation of diabetes-specific changes in nerves, smooth muscle cells, and ICC that are associated with functional GI and motility disorders in type 1 and type 2 diabetes. Other effective animal models include: genetic models of developmental defects in the ENS in Hirschsprung's disease; surgical and inflammatory models of the development of post-operative ileus and motility defects in response to a variety of inflammatory conditions; models of allodynia and hyperalgesia that have provided insights into the factors and mechanisms leading to visceral hypersensitivity;

and genetic knock-out models that have pointed to the molecular mechanisms responsible for GI neuromuscular function and disease.

Improved imaging or diagnostic techniques

Better, more quantitative, and, in many cases, noninvasive techniques have been developed to understand normal and abnormal esophago-gastric motility (e.g., high-resolution manometry, impedance manometry and impedance pH, SPECT, and MRI), to assess GI motility (e.g., stable isotope breath tests, SmartPill), and to evaluate involvement of the CNS (e.g., PET, MRI, magnetoencephalography) in functional GI disorders.

Mechanisms of obstructed defecation and fecal incontinence

Obstructed defecation and fecal incontinence present a significant burden for patients. New techniques, including the use of biofeedback, have been developed to treat these disorders with evidence from randomized, controlled trials that these treatments are effective for constipation and fecal incontinence.

Evaluation of drugs with the potential for therapy in functional bowel disorders

Many different methods are available to evaluate potential new medicines to determine whether they provide benefit in the treatment of IBS. Great strides have been made in establishing a consistent design to study new potential medicines for the treatment of IBS, including: how to collect data, how long to study treatments, and what parameters to monitor to demonstrate efficacy based on valid psychometrics and construct validity. By having a strong, consistent way to test new medicines, it is more likely that the results of studies, whether positive or negative, are accurate and, if positive, will be regarded as acceptable by regulatory agencies.

GOALS FOR RESEARCH [8]

Research Goal 2.1: Understand the molecular and cellular events that yield normal motility, sensory behavior, and integration between motility and secretory activity in the GI tract and the pathophysiology of functional GI disorders and motility disorders.

In order to identify and properly repair defects associated with GI disease, it is critical to develop a detailed understanding of the normal functioning of the gut and the brain-gut axis. Research to elucidate major signaling pathways that perform the sensory and neuromuscular functions of the GI organs or the excitability and contractile mechanisms in these organs is warranted. Studies of the physiology of ICC, which serve as pacemakers and neural mediators in the gut, may lead to new insights for methods to prevent loss of these cells or compensate for their function in functional GI disorders.

Objectives:

- Develop cell-specific markers for each cellular component involved in gut neuromuscular function and visceral sensation.
- Characterize differences in gene expression in specific cell populations in health and disease.
- Develop biomarkers that can be used to assess the health and function of specific cellular components.

[8] *Research Goals are numbered for ease of reference only; the numbers do not indicate prioritization of scientific topics.*

- Determine why ICC are lost in many motility disorders and how to restore this population of cells.

Research Goal 2.2: Understand the development of the GI tract and brain-gut interactions and determine how the aging process and differences in sex and gender affect gut development and function and brain-gut interactions. (See also Goal 9.7.)

The burden of many functional GI disorders varies by age and sex/gender. Studying the effects of genetics, sex, hormonal status, and aging on the development and function of cells and integrated behaviors of cells in the GI tract will provide a more complete understanding of normal GI function throughout the lifespan. This information combined with the identification of developmental or gender-specific factors that predispose the gut to motor or functional pathologies will help researchers design targeted therapies to prevent, treat, or reverse these disorders in vulnerable populations.

Objectives:

- Develop a comprehensive picture of how genetics and environmental factors affect development and maintenance of normal function of the GI tract and the brain-gut axis.
- Determine why women are disproportionately affected by functional GI and motility disorders.
- Determine how hormonal status affects the neuromuscular apparatus of the GI tract and how it might predispose the gut to abnormal motility or abnormal sensation.
- Learn the mechanisms of fecal incontinence and develop means of prevention and treatment.

Research Goal 2.3: Understand the components and functional interactions of the peripheral (autonomic and enteric) and central nervous systems in normal physiology and in functional GI and motility disorders.

The nervous system of the gut is highly complex and involves multiple cellular components, such as efferent and afferent neurons, interneurons, and glia. A primary goal for research on functional GI disorders is to distinguish the role of these nervous system cells in health and disease. Interactions between the intrinsic and extrinsic nervous systems and alterations in neural targets and effectors in functional GI and motility disorders are also incompletely understood.

Objectives:

- Define the role of the vagal homeostatic system (e.g., vagal anti-nociception, vagal anti-inflammatory reflex).
- Determine the role of the sympathetic nervous system in regulating gut and sphincter function.
- Define how neural integration is accomplished in peripheral neurons and how GI sensation integrates with central pain pathways.
- Understand the degree to which cognitive and emotional processes participate in the generation and/or symptoms of chronic functional GI disorders.

Research Goal 2.4: Understand the immune functions of the muscularis, integration between mucosal and muscle immune responses, and how inflammatory processes contribute to the pathogenesis and maintenance of functional GI and motility disorders.

The GI tract is a key point of interaction between the immune system and the environment. Many GI motility disorders are associated with immune activation. Progress in this field requires a deeper understanding of the immune function of the gut and how it relates to the development of functional GI and motility disorders. Understanding the role of inflammatory responses of resident and infiltrating immune cells (innate and adaptive immune responses) and characterizing specific populations of immune cells that either reside in or infiltrate gut tissue are important research goals. Interactions

of the immune system with intrinsic and extrinsic neurons and other cells might also influence the onset of disease. Clinical testing is warranted to evaluate the effectiveness of mast cell stabilizers and mediators on GI symptoms. With increased knowledge about immune function in the gut, researchers will be able to design new therapies to blunt the effects of inflammation-induced motility disorders and their systemic consequences (e.g., systemic inflammatory response syndrome, barrier function failure).

Objectives:

- Define mucosal functions (immune, barrier, sensing/taste) and the role of inflammation.
- Determine the interactions of pro-inflammatory immune cells and neural cells in diseases of the ENS and hypersensitivity.
- Characterize the inflammatory responses to various insults to the GI tract and develop the means to disrupt or reverse these responses.
- Characterize the effects of probiotics on mucosal-immune interactions.
- Determine the efficacy of antibiotics in prevention and treatment of post-infectious and idiopathic IBS.

Research Goal 2.5: Understand peripheral and central pain and sensory pathways and how these pathways are affected in functional GI and motility disorders.

Pain is one of the most debilitating symptoms of functional GI and motility disorders. In order to develop better treatments to combat pain, it is important to identify the ion channels expressed in nociceptive nerves and to search for specific blockers of these channels that might relieve chronic visceral pain. Understanding how generator potentials develop in nociceptive neurons and how to block or reverse the processes that lead to hypersensitivity of afferent neurons will also help researchers design new therapies. Another key issue is to determine how sensory input from the gut integrates with information from other viscera and with activity of the CNS. Progress could be made by defining how the CNS senses or processes sensory information differently in animal models and in patients with functional GI and motility disorders.

Objectives:

- Clarify enterochromaffin cell-afferent nerve terminal interactions. What are the mechanisms of afferent nerve activation and sensitization in the GI tract?
- Characterize the integration between visceral nociceptive pathways and motor pathways.
- Determine the mechanisms for GI sensation and perception of sensation and how the gain of visceral sensory pathways increases in functional GI disorders.
- Develop new agents to reduce nociception and undesirable visceral sensation.
- Characterize the sites and mechanisms of central processing of inappropriate sensation in the GI tract in normal individuals and patients with functional GI disorders.

Research Goal 2.6: Understand the noxious visceral signaling causing nausea and vomiting related to gastric neuro-electrical and/or motor dysfunction and the bi-directional brain-gut interactions.

Gastroparesis provides an archetypal disease for investigative inquiry. Chronic vomiting, a debilitating and socially isolating digestive symptom, creates potentially life-threatening disruptions in fluid and electrolyte homeostasis and compromises nutritional status. Chronic nausea remains a significant hidden disability. Nausea and vomiting usually occur in tandem and overlay with other GI symptoms as well as presenting in numerous digestive diseases. More effective treatments for nausea and vomiting would improve quality of life and physical functioning in a vast array of illnesses. A paucity of research exists

for defining peripheral noxious signaling of nausea and vomiting related to primary GI motor/sensory disturbances. Understanding normal electrophysiologic motor function would assist in unraveling the perturbed sensory signaling in postprandial nausea and vomiting, as well as the nausea and vomiting of severe gastric motor derangements as are found in gastroparesis. More research related to the autonomic nervous system and circadian patterns may provide interesting insights and lead to the development of interventions that impact the brain-gut-humoral axis.

Objectives:

- Clarify electrophysiologic normality and abnormalities, especially tachyarrhythmias, and their contribution to noxious signaling of nausea/vomiting.
- Characterize function and dysfunction of gastric and small bowel ICC and the presence, absence, and relative functional phenotypes of ICC in full thickness tissue biopsies and other GI tissue samples to identify abnormalities in slow wave generation, frequency regulation, propagation, and neural regulation.
- Characterize the impact of diet, anxiety, and sleep disruption on modulation of nausea and vomiting.
- Use animal models to understand the role of cytokines and to increase understanding of afferent signaling in nausea and vomiting.
- Determine the modulating effect of therapeutic interventions on CNS function using evidence from functional MRI (fMRI) studies.
- Understand the role of esophageal, gastric, and small bowel sensory and mechanoreceptors (in nerves, ICC, and smooth muscle cells) in the generation of nausea and vomiting.

Research Goal 2.7: Understand the role of the microflora in functional GI disorders and motility disorders.

The human microflora include the diverse collection of bacteria and other microbes inhabiting the digestive tract; little is known about the role of the microflora in health and disease. An important research focus is to understand how luminal factors such as the gut microflora, inflammation, diet, or infection influence the relative types, density, and functions of enteroendocrine cells—hormone-producing cells found throughout the lining of the digestive tract. The impact of the intestinal microflora on bi-directional brain-gut communication and the interactions between a patient's genotype and that of the microflora in their gut are key issues. Understanding these processes could provide new insights into the triggers of symptoms and symptom differences (e.g., constipation, diarrhea, or bloating) in functional GI and motility disorders.

Objectives:

- Characterize the impact of the intestinal microflora on bi-directional brain-gut communication.
- Characterize the effects of stress, diet, and infections on gut microflora.
- Determine the relationship between a patient's genotype and the microbiome.
- Determine the impact of alterations in the content of the microflora on GI function and sensation.

Research Goal 2.8: Use information from studies of animal models and cellular physiology to understand the integrated function of the musculature and the intrinsic and extrinsic nervous systems.

Animal models that mimic human disease or that recapitulate specific pathways, structures, or behaviors contributing to human disease are critical tools for understanding the relationships between cellular defects and organ- or system-level symptoms in functional GI and motility disorders. These models allow researchers to

address important questions that cannot be studied in human patients, such as how cellular elements intersect to yield tissue- or organ-level behaviors or how nerves regulate the barrier function of the mucosa. Research is also warranted to define changes in the ENS that cause inappropriate motor patterns in the gut and the contribution of the CNS to altered function in the ENS. With animal models, researchers can study how the ENS stores motor patterns and potentially translate that knowledge into new strategies to selectively change an inappropriate motor pattern to one that would be beneficial to patients with functional GI and motility disorders.

Objectives:

- Develop conceptual and/or quantitative models to demonstrate how various pathophysiologic inputs—such as stress, altered gut microflora, or neuroendocrine dysfunction—might influence motor patterns, integration between motility and secretion, and visceral sensation.
- Use animal models to determine the role of infection, metabolic disease, stress, and sex on GI phenotype, neural integration, afferent nerve sensitivity, neuro-immune responses, neuro-muscular transmission, pacemaking, and integration of information between the peripheral and central nervous systems.
- Use selective genetic models to determine the role of specific signaling pathways, neurotransmitter systems, and immune responses in normal and pathophysiologic states.
- Develop noninvasive, *in vivo* research diagnostic tools suitable for small animal research. These tools (e.g., small animal MRI, ultrasound, PET, SQUID, breath tests) could be used to screen for GI phenotypes in genetic mutants and also to test pharmacological agents.
- Develop and utilize better data mining tools for the screening and comparison of large-scale expression data in animal disease models.
- Use animal models to develop new biomarkers of gut health and function.

Research Goal 2.9: Characterize the factors in diabetes that lead to the development of functional GI and motility diseases.

Patients with long-standing diabetes are prone to development of GI motility disorders at all levels of the GI tract from the esophagus to the anorectum. These problems are increasing as obesity and cases of type 2 diabetes increase. GI complications of diabetes may be complex, severe, and substantially decrease quality of life. Common complaints include: dysphasia, premature satiety, esophageal reflux, constipation, pain, nausea, vomiting, and diarrhea. GI motility defects have traditionally been attributed to the development of autonomic neuropathy; however, recent findings suggest that additional cellular defects may also contribute to GI complications. Although few studies have addressed the changes in visceral sensory function that occur in diabetes, abnormalities in pain perception thresholds, vagal activity, and evoked brain potentials suggest that diabetes-related neural changes may be both peripheral and within the CNS.

Objectives:

- Characterize changes in neural chemical coding in the ENS and functional consequences of identifiable neuropathies associated with long-standing diabetes.
- Determine non-neuronal cells that are functionally impaired by loss or suboptimal insulin/IGF-1 signaling.
- Determine whether dysfunction or loss of neuronal and non-neuronal cells in diabetes results from poor glycemic control, loss of or defective insulin/IGF-1 signaling, or other unidentified defects inherent to the complications of diabetes.
- Develop novel biomarkers to evaluate damage to specific cellular compartments or sub-classes of enteric neurons in diabetes.
- Characterize specific motility and functional defects in animal models of both type 1 and type 2 diabetes.

Research Goal 2.10: Determine how genotype contributes to or predisposes patients to the development of functional GI and motility disorders.

A primary goal in digestive diseases research is to understand the role of genetics in the etiology, manifestations, prognosis, and therapeutic responses of patients with functional GI and motility disorders. These diseases are multifactorial and likely to be polygenic in nature. Interactions between genes and the environment also contribute to the complexities of these disorders in unknown ways. By identifying specific genetic polymorphisms or gene-environment interactions that predict disease or correlate with intermediate phenotypes (e.g., sensitivity to pain, GI transit, autonomic parameters, activity and sensory processing in the CNS), it might be possible to design early intervention strategies to prevent or blunt the development of full clinical syndromes of functional GI and motility disorders in genetically at-risk people. This may also allow targeting of therapeutics to the population most likely to receive benefit.

Objectives:

- Develop genetic epidemiological studies to discover common genetic factors that predispose patients to develop functional GI or motility disorders.
- Develop and validate endophenotypes (intermediate hereditary characteristics between the disease and the genotype) as a means of clarifying links between the genotype and the complicated phenotypes of functional GI and motility disorders.
- Utilize validated endophenotypes to clarify classification and diagnosis of functional GI and motility disorders and to foster the development of animal models.
- Utilize a pharmacogenetic approach to predict which patients might respond to specific therapies.

Research Goal 2.11: Determine the role of food in the development of functional GI and motility disorders. (See also Goal 8.8)

Growing evidence suggests that food plays an important role in the pathogenesis of GI symptoms. It is possible that specific food constituents have primary effects on gut function and sensation, rather than an intermediary effect through gut microflora. Abnormal immunologic responses to food might be linked to the development of defects in gut function and sensation and other symptoms of functional GI and motility disorders.

Objectives:

- Utilize animal models to study the influence of specific food components on gut function and gut sensation.
- Develop animal models of food allergies.
- Determine the effects of specific food components and food allergies on enteroendocrine cell populations and on the function of afferent nerve function.
- Develop patient tests to identify food allergies or hypersensitivity to food components.
- Identify and critically evaluate dietary treatments for functional GI disorders.
- Determine the role of gut taste receptors on GI function.

Research Goal 2.12: Develop new technologies and therapeutic approaches to effectively treat patients with functional GI and motility disorders.

Progress in alleviating functional GI and motility disorders would be spurred by the development and routine application of innovative, state-of-the-art technologies for diagnosis and treatment. Imaging approaches in both animal models and human subjects could accelerate early clinical testing and evaluation of new therapeutics and help researchers identify the most promising candidates to test in clinical trials. Improvements in noninvasive electrical recording of the GI tract for diagnostic purposes would facilitate diagnosis of patients with functional GI disorders. The development of simple, noninvasive GI motility testing modalities that can be applied on a large scale would improve

our understanding of the prevalence of these disorders in the general population. Research on the causes and mechanisms of neural hypersensitivity in relation to other functional pain disorders could make it possible to apply more general pain management strategies to functional GI disorders. Finally, a major advance in treating functional GI disorders might come from improving small bowel transplantation so that this procedure becomes as routine as kidney transplantation.

Research into the biology of stem cells will facilitate the development of stem cell-based treatments of gut disorders where smooth muscle cells, ICC, or enteric neurons are reduced, absent, or malfunctioning as in fecal incontinence, pseudoobstruction, gastroparesis, and constipation. By using stem cell biology technology and tissue engineering approaches, it may be possible to repair or replace damaged cellular or tissue components.

Objectives:

- Develop and evaluate new therapeutic pharmaceutical agents for treatment of functional GI and motility disorders.
- Develop new devices or applications of novel stimulus regimes to target vagal nerve function.
- Develop new devices, surgical techniques, or tissue replacement approaches to enhance fecal continence.
- Develop and validate standard measures for health outcomes research (e.g., primary treatment endpoints, health-related quality of life, psychosocial assessment, health behaviors, such as healthcare utilization and decreased daily function, and costs).
- Standardize behavioral treatments to make them generalizable to a broader population.
- Identify GI tract-specific stem cell populations and develop techniques to regenerate specific cell populations or tissues.

Research Goal 2.13: Evaluate therapeutic outcomes and the impact of doctor/patient interactions to determine effective treatments for functional GI and motility disorders.

Functional GI and motility disorders result from a complex and poorly understood combination of physiologic and psychological factors that can give rise to a variety of symptoms. Patient-oriented research could be pursued to define the underlying causes of the diverse symptoms of GI motility disorders. Clinical trials can be designed to compare the relative clinical effectiveness and cost-effectiveness of standard treatment approaches to innovative disease management approaches that involve novel pharmacological compounds and non-pharmacological strategies (e.g., disease education, cognitive behavioral therapy, physician education, web-based telemedicine approaches). The impact of standardized physician training programs on GI patient outcomes and cost is another important research question.

Objectives:

- Develop innovative ways to optimize healthcare delivery systems for GI disorders to enhance outcomes and reduce costs.
- Determine the elements of the healthcare provider-patient relationship that will improve healthcare outcomes, such as interview techniques, relationship-centered care, emotion management, and placebo administration.
- Determine the effect of healthcare provider education and training to enhance the provider-patient relationship on clinical outcomes, including patient satisfaction, adherence to treatment, improved symptoms, quality of life, and healthcare costs.
- Conduct randomized, controlled trials of stress management and relaxation methods, hypnotherapy, and cognitive behavioral therapy in pediatric and adult populations.

MAJOR CHALLENGES AND STEPS TO ACHIEVE THE RESEARCH GOALS

Methods to study the diversity of GI cells: Functional GI and motility disorders are complex and result from functional defects in a wide variety of cell types. At present, few well-defined animal models have been developed, and human tissues and cells are infrequently available, except from biopsies. A better understanding is required of the many cell types that produce normal GI motility and appropriate levels of GI sensation, including muscle cells, intrinsic and extrinsic neurons, glial cells, ICC, and a variety of immune cells (both resident and recruited). The development of innovative techniques to isolate and identify specific types of cells from healthy and diseased gut samples would promote progress across the field. Such techniques would allow determination of specific changes in cellular phenotype that might occur during development of a disease process and facilitate the use of state-of-the-art technologies, such as genomic and proteomic analyses.

Moreover, phenotypes of GI cells involved in motility and neural responses are not conserved adequately in cell cultures because the gut microenvironment is important to establish and maintain specific cellular phenotypes. Smooth muscle cells, for example, change radically when removed from the natural microenvironment and can no longer be considered to represent a smooth muscle phenotype within a short period in culture. Higher standards for verification of phenotypic fidelity of cultured cells need to be established. A shortage of culture models makes large-scale genomic and cell biology studies very difficult. The development of new technologies is required in order to manipulate genetic expression of GI cell types while the cells are still in their native environment. For example, enhanced spatial and temporal targeting of transgenic methodologies would enable researchers to create better, cell-specific knock-outs in GI cells. Techniques to effectively turn on and off expression of specific genes in adult animals would also facilitate research in this area.

Technologies to study the influence of the nervous system: Visceral pain is among the most debilitating symptoms of many GI disorders. Better understanding of nociception, inappropriate sensation, and sensitization of afferent neurons and pathways would enhance the development of new therapies to combat pain. Development of techniques to discriminate between different classes of afferent nerve terminals and to clearly identify nociceptive nerve terminals in living tissues should be encouraged. Dynamic imaging techniques would provide a means to study the behavior of afferent nerve terminals in the lamina propria and muscularis layers of the GI tract. A more detailed understanding of the mechanisms of generator potentials in nociceptive neurons would strengthen the field.

A better understanding of the integration between the brain-gut axis in pathophysiologic conditions and pathways of communication between intrinsic and extrinsic neurons is required. Research in this area could be enhanced by the design of: better recording methods to measure activity of the autonomic nervous system in conscious experimental animals and to correlate neural activity with motility; techniques to record from the CNS in conscious experimental animals (e.g., fMRI or PET) that correlate activity with peripheral neural activity and motility; and methods to probe the links between disorders of GI function (e.g., constipation and diarrhea) and pain and discomfort in the gut.

Systems biology approaches: The development of models or a conceptual framework that unifies disciplines related to morphology and cell biology, including anatomy, histology, histochemistry, molecular biology, and pathway

analysis, with topics relevant to physiology, such as functions of cells and integration of cellular function in tissues and organs, would enhance research to understand how cellular and sub-cellular events summate to produce whole system physiology and how cellular dysfunction contributes to pathophysiologic behaviors. Assembling and comparing databases from gene and protein expression studies of specific GI cell types in normal and pathophysiologic states might begin to reveal the many micro-defects that can lead to whole-organ or system dysfunction. Understanding specific changes in gene and protein expression that are common to specific functional GI and motility disorders might provide insights into biomarkers that can characterize these diseases more definitively. More sophisticated dynamic imaging techniques would facilitate understanding of interactions between GI cells and how discrete cellular and sub-cellular defects in genetically altered animal models contribute to function and dysfunction of tissues and organs.

Integrative biology approaches: Studies of how environmental factors and life experiences affect gut function (e.g., how stress, psychosocial co-morbidities, and impairment of CNS-ENS regulatory systems affect the gut) will help to elucidate their impact on the morphology, function, and integration of GI cells and tissues. Many integrative studies are difficult or unethical to perform on human patients. Thus, improved animal models for functional GI and motility disorders would enable researchers to explore the cause-and-effect relationship between suspected pathologic factors and the development of symptoms and disease. While it would be extremely useful to have animal models that closely mimic human diseases, much can also be learned from animals that manifest partial phenotypes (i.e., that capture specific morphologic, pathophysiologic, or behavioral aspects of human disease). A clearer understanding of what can and cannot be extrapolated from animal models to humans will help refine animal models. Complementary studies on human GI tissues and cells will enable researchers to determine whether cellular and molecular components and pathways identified in animal models are relevant to human physiology and pathophysiology. The development of better techniques for monitoring GI function, autonomic function, and brain function in genetically altered animals would increase efficiency for phenotyping animal models. In addition, genetic screens to specifically identify defective GI motility, autonomic and CNS phenotypes, and the inactivation of genes linked to defective motility phenotypes in animals would promote progress.

New model systems are required to determine the functional effects of variations in candidate genes that are reputed to alter motor and sensory functions, as well as somatization, resulting in GI tract symptoms or diseases (i.e., testing of functional genomics). Further research is also required to develop techniques that evaluate GI motility in model organisms, such as zebrafish or mice, in order to screen genes that are linked to altered bowel function. Testing human polymorphisms in vertebrate models would shed light on the consequences of genetic variation, in terms of motor and/or sensory function.

Standardized molecular definitions, biomarkers, and clinical treatments: Progress in understanding and treating GI disorders would be accelerated by the development of consensus clinical descriptions of functional GI disorders and motility disorders. Such descriptions would, in turn, enable the establishment of multicenter patient registries and tissue/reagent banks. These resources would facilitate research on the physiologic responses of human GI tissues from comprehensively phenotyped and genotyped individuals with functional GI

and motility disorders to help determine the pathophysiology and mechanisms of these disorders. Moreover, a systems biology approach would allow patient biosamples to be analyzed for genetic similarities and differences and to be used to develop biomarkers that might be used to further categorize and diagnose functional GI disorders and motility disorders.

Similarly, standardization of clinical assessments and treatments of functional GI disorders and motility disorders across patient populations and clinical research centers would be an important step toward enabling researchers to directly compare data and outcomes. Standardization of techniques and methods of analysis of brain imaging, assays for mucosal cytokines and neuropeptides, and reporting of clinical outcomes in treatment trials would all strengthen the research field as a whole.

A consortium approach would be beneficial because it would provide the large numbers of samples and controls that are required for informative studies of these disorders.

Innovative transplantation techniques: Better transplantation therapies are required, possibly based on stem cell therapies and tissue engineering. To address this challenge, more information is needed about the stem cell populations in the GI tract and how to manipulate the phenotypes of stem cell derivatives. Improvements in transplantation techniques would allow GI transplantation to become as common as with other organ systems. The development of innovative techniques to restore function within regions of GI organs through tissue engineering could also be encouraged.