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REVIEWS IN BASIC AND CLINICAL GASTROENTEROLOGY AND HEPATOLOGY

Chronic Visceral Pain: New Peripheral Mechanistic Insights and Resulting Treatments

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Chronic visceral pain is one of the most common reasons for patients with gastrointestinal disorders, such as inflammatory bowel disease or disorders of brain-gut interaction, to seek medical attention. It represents a substantial burden to patients and is associated with anxiety, depression, reductions in quality of life, and impaired social functioning, as well as increased direct and indirect health care costs to society. Unfortunately, the diagnosis and treatment of chronic visceral pain is difficult, in part because our understanding of the underlying pathophysiologic basis is incomplete. In this review, we highlight recent advances in peripheral pain signaling and specific physiologic and pathophysiologic preclinical mechanisms that result in the sensitization of peripheral pain pathways. We focus on preclinical mechanisms that have been translated into treatment approaches and summarize the current evidence base for directing treatment toward these mechanisms of chronic visceral pain derived from clinical trials. The effective management of chronic visceral pain remains of critical importance for the quality of life of suffers. A deeper understanding of peripheral pain mechanisms is necessary and may provide the basis for novel therapeutic interventions.

Keywords: Visceral Pain; Inflammatory Bowel Disease; Irritable Bowel Syndrome; Histamine; Serotonin; Microbiome; Abdominal Pain; Inflammation.

Pain, defined as an unpleasant sensory and emotional experience associated with or resembling that associated with actual or potential tissue damage, can be acute or chronic.[1] It can originate from somatic (muscle, bone, or soft tissue) or visceral (thoracic, abdominal, or pelvic organs) structures.[1] Visceral pain is one of the most challenging clinical conditions facing patients and their health care providers. It is extremely common. Abdominal pain is a key reason that patients with gastrointestinal disorders, such as inflammatory bowel disease (IBD) or disorders of gut-brain interaction (DGBI), including irritable bowel syndrome (IBS) or functional dyspepsia (FD), seek medical attention.[2,3] More than 70% of patients with IBD experience abdominal pain during an acute flare,[4] and between 20% and 60% report chronic abdominal pain.[5] Chronic visceral pain is a hallmark of some DGBI, which affect up to 40% of adults, primarily women, worldwide.[6]

The diagnosis and treatment of chronic visceral pain is difficult, largely because it is poorly localized and difficult to describe due to the relatively small density of nerve terminals in the viscera and the divergent projections into the spinal cord,[7] and because the pathophysiology remains incompletely understood. Chronic visceral pain is, thus, a significant burden to patients and is associated with anxiety, depression, decreased quality of life, and increased direct and indirect health care costs.[5,8,9] IBS alone is estimated to cost the United States (US) ∼US $350 million each year for outpatient clinic visits, not including diagnostic testing, medications, nonpharmacologic therapies, or indirect costs due to lost productivity.[10] Unfortunately, these challenges have been further amplified by the opioid crises.[11,12] This highlights the continued need for advances in understanding of the pathophysiology of visceral pain to enable both effective and safe therapies.

Chronic visceral pain is a disorder of the microbiota-gut-brain axis, and central and peripheral mechanisms both contribute to its pathogenesis (Figure 1). Triggers include stress, psychological comorbidities, such as anxiety or depression, diet, low-grade intestinal inflammation, and microbial dysbiosis.[4,13–15] Most abdominal pain signaling

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Abbreviations used in this paper: **HT, 5-hydroxytryptamine; CB1, cannabinoid receptor 1; cGMP, guanosine 3′,5′-cyclic monophosphate; DGBI, disorders of gut-brain interaction; DOR, δ-opioid receptor; FD, functional dyspepsia; FDA, Food and Drug Administration; FMT, fecal microbial transplant; FODMAP, fermentable oligosaccharides, disaccharides, monosaccharides and polyols; GABA, γ-aminobutyric acid; GC-C, guanylate cyclase-C; GPCRs, G protein-coupled receptors; IBD, inflammatory bowel disease; IBS, irritable bowel syndrome; IBS-D, inflammatory bowel disease with diarrhea; KOR, κ-opioid receptor; MOR, μ-opioid receptor; RCT, randomized controlled trial; SCFA, short-chain fatty acid; SNRI, serotonin norepinephrine reuptake inhibitor; SSRI, selective serotonin reuptake inhibitor; TCA, tricyclic antidepressant; TRP, transient receptor potential; TRPV1, transient receptor potential vanilloid 1; US, United States.**

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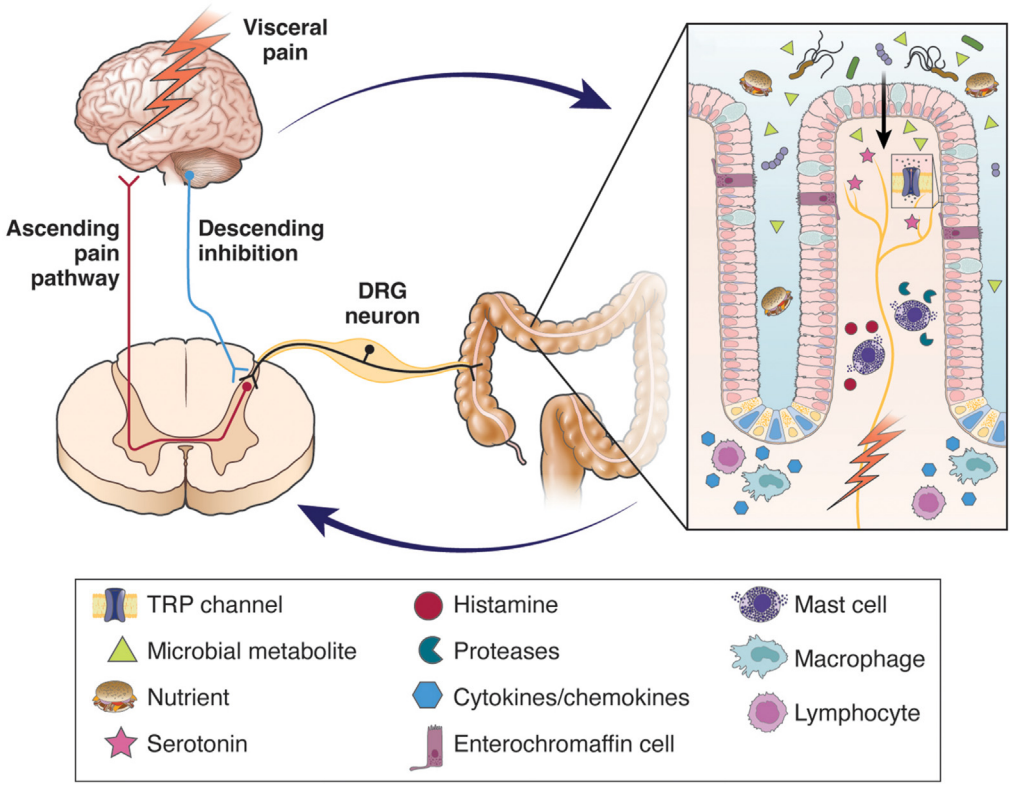

Figure 1. Chronic visceral pain is a disorder of the gut-brain axis. Nociceptors have cell bodies that lie in the dorsal root ganglia (DRG) and pseudounipolar axons that connect the intestine and the spinal cord. These synapse with second-order neurons in the spinal cord and with central ascending pathways thereafter. Nociceptive neurotransmission in the spinal cord is modulated by descending pathways. (*Inset*) At the level of the mucosa, nociceptive terminals are both mechanosensitive and chemosensitive and are stimulated by luminal factors (eg, microbial products and nutrients) as well as by host mediators released due to infection, inflammation, or tissue damage (eg, serotonin, histamine, proteases, chemokines, and cytokines). These mediators can act indirectly via the epithelium/enterochromaffin cells or can stimulate nociceptors directly if there is a breakdown in the mucosal barrier. This results in sensitization of ion channels such as TRP, resulting in increased visceral pain.

originates from nociceptors (pain-sensitive neurons), called visceral primary afferent nerves, whose cell bodies lie in the dorsal root ganglia and which have pseudo-unipolar axons connecting the intestine and the spinal cord.[16] Nociceptors synapse with second-order neurons in the thoracolumbar and lumbosacral spinal cord[17] and thereafter with central ascending pain pathways. Nociceptive neurotransmission in the spinal cord is modulated by descending pathways originating from the hypothalamus and midbrain.[18]

Sensitization of nociceptors, defined as a decrease in the threshold for stimulation and an increase in the magnitude of the response,[19] can occur peripherally, in the central nervous system, or both. This results in hyperalgesia, a heightened response to painful stimuli, and allodynia, which is pain arising from nonpainful stimuli.[19] Central sensitization may also result in comorbid pain involving different organ systems,[20] a discussion of which is beyond the scope of this review.

At the level of the periphery, nociceptive nerve terminals are found in muscle and serosa as well as in the mucosa.[7] Nociceptors are mechanosensitive and are stimulated by stretch or distention.[16] These actions are mediated by a variety of mechanosensitive ion channels, such as the transient receptor potential (TRP) receptors, including TRP vanilloid 1 (TRPV1) and 4, and TRP ankyrin 1, the 2-pore domain potassium channel family, the degenerin/epithelial sodium channel family, including the acid-sensing ion channels 1, 2, and 3, and the piezo-type mechanosensitive ion channel component 2 (Piezo-02).[21,22]

Nociceptors at the mucosal level are also chemosensitive and are stimulated by luminal factors, such as microbial products and nutrients, as well as by chemical mediators released during tissue infection, inflammation, or damage. These include bacterial toxins, neurotransmitters, proteases, bioactive amines, such as histamine, and serotonin, neurotrophins, adenosine-5′-triphosphate, chemokines, and cytokines (Figure 1, *inset*).[13,23] Luminal products can either stimulate nociceptors directly, particularly if there is associated breakdown in the mucosal barrier as seen in both IBD and IBS,[24,25] or indirectly via the epithelium or enteroendocrine cells.[26]

Chemical compounds and luminal products can, in turn, stimulate pronociceptive G protein-coupled receptors (GPCRs) or lead to increased expression and activation of ion channels, such as TRP or voltage-gated sodium and calcium channels, or can decrease potassium channel activation and expression, resulting in peripheral sensitization. In turn, nociceptors can release neurotransmitters, such as substance P and calcitonin gene-related peptide, which augment the inflammatory

response in the periphery and activate second-order neurons in the spinal cord, leading to neurogenic inflammation[13,23] (Figure 1, *inset*).

Building on this pathophysiological framework, this review will focus on recent advances in visceral peripheral pain neurotransmission and mechanisms that result in sensitization of afferents in patients with IBD or painful DGBI. It will discuss specific physiologic and pathophysiologic preclinical peripheral mechanisms that have been translated into receptor-based treatment approaches for visceral pain in clinical trials. Some of these treatments have targeted advances in the physiology of nociceptors or intermediary cells, or both, whereas others target new understanding of pathophysiologic mechanisms of specific disorders.

Mechanistic Advances and the Resulting Therapies

Guanylate Cyclase-C and Visceral Pain

Guanylate cyclase-C pharmacology and preclinical studies. The enterocyte receptor guanylate cyclase-C (GC-C) plays an essential role in fluid secretion, barrier function, and nociception. Drugs such as linaclotide and plecanatide have taken advantage of this homeostatic system to treat visceral pain. GC-C is found on the apical surface of enterocytes throughout the gastrointestinal tract and is activated by the paracrine hormones uroguanylin and guanylin.[27] Activation of GC-C triggers enzymatic conversion of guanosine-5'-triphosphate to guanosine 3′,5′-cyclic monophosphate (cGMP), which in turn regulates activity of the apical cystic fibrosis transmembrane conductance regulator, leading to increased luminal chloride and bicarbonate secretion and a secondary increase in intestinal motility.[27] Genetic mutations in the guanylate cyclase 2C gene (*GUCY2C*) have been found in patients with congenital secretory diarrhea[28] and may predispose patients to IBD,[29] whereas dysregulated GC-C expression has been implicated in the pathophysiology of both IBD[30] and IBS.[31] Sex differences have not been reported.[32]

Epithelial GC-C signaling has a key role in nociception. Linaclotide, a minimally absorbed GC-C agonist, decreased the visceral motor response to colorectal distention in both acute colitis and stress-induced models of visceral hypersensitivity. The effects of linaclotide were abolished in GC-C–knockout animals, confirming its specificity.[33] Linaclotide[34] or direct application of cGMP[34,35] to an ex vivo preparation of nociceptor afferents decreased response to circumferential stretch in control animals as well as in acute colitis[35] and in postinflammatory[34] models of visceral pain. GC-C expression was not found on nociceptors,[34,35] suggesting its antinociceptive effects were indirect. Indeed, linaclotide[34] and uroguanylin[35] both stimulated cGMP release from cultured epithelial cells.[35] The effects of linaclotide were abolished in ex vivo preparations where the mucosa was removed.[34]

These studies suggest that epithelial GC-C activation causes basolateral cGMP secretion, which decreases nociceptor activity, providing a biological mechanism for the clinical effects of GC-C agonists. We note that a recent study has challenged the dogma that enterocyte-derived cGMP is the main antinociceptive mediator of GC-C activation,[36] as discussed in section 6.

Clinical trials. Linaclotide and plecanatide have been tested in multiple randomized controlled trials (RCTs) in IBS with constipation, summarized in a prior meta-analysis (for summary of all trials discussed see Table 1).[37] Both were more efficacious than placebo in the effect on abdominal pain, according to the US Food and Drug Administration (FDA)-recommended end point for abdominal pain in IBS with constipation, consisting of a $\geq$30% improvement from baseline for $\geq$50% of weeks. However, delayed-release forms of linaclotide, developed based on the premise that ileocecal delivery of the drug targets abdominal pain without affecting bowel habit, were not superior to placebo over most abdominal pain measures in a phase II RCT.[38]

Peripherally Acting Opioids and Visceral Pain

Pharmacology and preclinical studies. Opioids signal through 4 GPCRs: μ-opioid receptors (MORs), δ-opioid receptors (DORs), κ-opioid receptors (KORs), and nociceptin opioid receptors.[39] The analgesic effect of conventional opioids can be strong (eg, oxycodone, morphine) or weak (eg, codeine) and predominantly result from activation of MORs, although DORs and KORs also play a role. On nociceptors, these receptors trigger GPCR-Gi/o protein signaling leading to the recruitment of multifunctional intracellular proteins, called β-arrestins, and sustained signaling by endosomes.[40] This signaling modulates ion channels and, ultimately, inhibits action potential firing. Receptor expression is increased in inflammatory conditions, including active IBD, possibly leading to altered signaling.[41]

Conventional opioids can exhibit potent analgesic actions, particularly for acute pain, but are limited by their adverse effect profile, including cognitive impairment, respiratory depression, nausea, constipation, and addictive potential.[42] Analgesic tolerance leads to dose escalation and consequently greater risk of these potentially life-threatening adverse effects. Dose escalation is also implicated in the development of a paradoxical switch in signaling, leading to opioid-induced hyperalgesia, a poorly understood condition.[43] The opioid crisis has hastened the search for safer alternatives, including peripherally restricted opioids that lack addictive potential and central adverse effects such as respiratory depression and cognitive impairment.

Strategies to develop peripherally acting opioids are being explored to identify safe, yet effective, analgesics for visceral pain. Access to the central nervous system can be restricted, for example, by creating charged molecules, and several compounds display peripheral analgesic actions,[44,45] including loperamide, a MOR agonist.[46] To date, however, these do not exhibit sufficient analgesic effects to be clinically useful to treat visceral pain.

Another strategy is to target opioid receptor heterodimers, such as eluxadoline,[47] a MOR agonist and DOR antagonist with weak affinity for KORs. MORs and DORs are

Table 1. Summary of Evidence for Efficacy of Available Treatments Directed Against Peripheral Mechanisms of Abdominal Pain in Their Effect on Abdominal Pain as an End Point

Treatment studied	Condition	No. of studies	No. of patients	Comparator	Reported effect
Linaclotide, 290 μg q.d.	IBS-C	3 RCTs summarized in a meta-analysis[37]	2447	Placebo	RR of abdominal pain persistence = 0.79 (95% CI, 0.73–0.85)
Plecanatide, 6 mg or 3 mg q.d.	IBS-C	2 RCTs summarized in a meta-analysis[37]	2194	Placebo	RR of abdominal pain persistence = 0.84 (95% CI, 0.78–0.90) and 0.87 (95% CI, 0.81–0.93), respectively
Loperamide	IBS-D Unselected patients with IBS	2 RCTs[54,55]	24 60	Placebo Placebo	Abdominal pain score 3.0 vs −0.14, $P < .05$ 2.2 days with abdominal pain vs 8.3 days, $P < .01$
Eluxadoline, 100 mg or 75 mg b.i.d.	IBS-D	4 RCTs summarized in a meta-analysis[56]	2758	Placebo	RR of abdominal pain persistence = 0.89 (95% CI, 0.83–0.96) and 0.95 (95% CI, 0.88–1.04), respectively
Psyllium (up to 10 g/d)	Unselected patients with IBS	2 RCTs[89,90]	80 178	Placebo Placebo	Abdominal pain mild or absent in 52.5% vs 57.5%, N/S RR of adequate relief of abdominal pain at 1, 2, and 3 months = 1.60 (95% CI, 1.13–2.26), 1.44 (95% CI, 1.02–2.06), and 1.36 (95% CI, 0.90–2.04), respectively
Bran (up to 10 g/d)	Unselected patients with IBS	1 RCT[90]	190	Placebo	RR of adequate relief of abdominal pain at 1, 2, and 3 months = 1.13 (95% CI, 0.81–1.58), 1.22 (95% CI, 0.86–1.72), and 1.70 (95% CI, 1.12–2.57), respectively
Low FODMAP diet	IBS IBD	12 RCTs summarized in a meta-analysis[91] 2 RCTs[92,93]	914 52 89	BDA dietary advice Habitual diet Sham diet Sham diet Habitual diet	RR of abdominal pain persistence = 0.78 (95% CI, 0.57–1.06) RR of abdominal pain persistence = 0.72 (95% CI, 0.47–1.10) RR of abdominal pain persistence = 0.51 (95% CI, 0.30–0.87) Abdominal pain severity score 22 vs 30, $P = .098$ and 36 days with abdominal pain vs 38 days, $P = .78$ OR for improvement in abdominal pain frequency = 2.97 (95% CI, 1.12–7.89)
Rifaximin, 550 mg t.i.d. for 2 weeks	Nonconstipated IBS	2 RCTs summarized in a meta-analysis[56]	1260	Placebo	RR of abdominal pain persistence = 0.95 (95% CI, 0.89–1.01

Table 1. Continued

Treatment studied	Condition	No. of studies	No. of patients	Comparator	Reported effect
FMT	IBS with bloating	2 RCTs[96,97]	62	Placebo	Abdominal pain score 2.80 vs 3.88 at baseline with FMT, $P = .001$, compared with 3.57 vs 3.79 at baseline with usual treatment, $P = .205$
	Unselected patients with IBS	1 RCT[98]	165	Placebo	Abdominal pain score 166.8 and 186.3 posttreatment with 60 mg and 30 mg FMT, respectively, vs 307.0 with placebo, $P < .001$
	UC		20	Usual treatment	Abdominal pain score 0.9 vs 4.5 at baseline with FMT, $P = .026$, compared with 1.8 vs 4.9 at baseline with usual treatment, N/S
Gelsectan	IBS-D	1 RCT[99]	60	Placebo	Number of patients with totally to slightly unacceptable abdominal pain reduced from 67% at baseline to 0% at 4 weeks with gelsectan vs 83% to 60% with placebo, statistical significance not reported
Probiotics	All in unselected patients with IBS	32 RCTs[100]	3469	Placebo	RR of abdominal pain persistence = 0.72 (95% CI, 0.64–0.82)
Combination probiotics		11 RCTs[100]	1183	Placebo	RR of abdominal pain persistence = 0.59 (95% CI, 0.45–0.76)
Lactobacillus-containing strains		5 RCTs[100]	1482	Placebo	RR of abdominal pain persistence = 0.64 (95% CI, 0.45–0.90)
Saccharomyces cerevisiae I-3856		3 RCTs[100]	389	Placebo	RR of abdominal pain persistence = 0.78 (95% CI, 0.64–0.95)
Bifidobacterium-containing strains		3 RCTs[100]	212	Placebo	RR of abdominal pain persistence = 0.33 (95% CI, 0.23–0.47)
Bacillus-containing strains					
Ketotifen (titrated from 2 mg to 6 mg b.i.d.)	Unselected patients with IBS	1 RCT[109]	60	Placebo	7% of patients reporting severe abdominal pain vs 28%, $P = .02$
Ebastine 20 mg o.d.	Unselected patients with IBS	1 RCT[110]	55	Placebo	Relief of abdominal pain in 41% vs 20%, $P = .19$
	Nonconstipated IBS	1 RCT[111]	202	Placebo	≥30% improvement in abdominal pain in 37% vs 25%, $P = .081$
Disodium cromoglycate, 600 mg/d	IBS-D	1 RCT[112]	43	No treatment	≥50% improvement in abdominal pain in 77% vs 28%, $P = .002$
Peppermint oil (usually 2 capsules t.i.d.)	Unselected patients with IBS	7 RCTs summarized in a meta-analysis[119]	748	Placebo	RR of abdominal pain persistence = 0.76 (95% CI, 0.62–0.93)

Table 1. Continued

Treatment studied	Condition	No. of studies	No. of patients	Comparator	Reported effect
Red pepper (capsaicin)	Unselected patients with IBS FD	1 RCT[120] 1 RCT[121]	50 30	Placebo Placebo	Abdominal pain score 1.9 vs 2.7 at baseline with red pepper, compared with 2.3 vs 2.4 at baseline with placebo, reported as "statistically significant" Abdominal pain score 1.61 posttreatment vs 2.37, $P < .05$
Alosetron, 1 mg b.i.d.	IBS-D	6 RCTs summarized in a meta-analysis[56]	2606	Placebo	RR of abdominal pain persistence = 0.83 (95% 0.78–0.88)
Ramosetron, 5 μg or 2.5 μg o.d.	IBS-D	5 RCTs summarized in a meta-analysis[56]	1928	Placebo	RR of abdominal pain persistence = 0.82 (95% CI, 0.75–0.89) and 0.75 (95% CI, 0.65–0.85), respectively
Ondansetron, 12 mg q.d, bimodal release or titrated up or down from 4 mg o.d.	IBS-D	3 RCTs summarized in a meta-analysis[127]	327	Placebo	RR of abdominal pain persistence = 0.95 (95% CI, 0.74–1.20)
Tegaserod, 6 mg b.i.d.	IBS-C FD	Pooled analysis of 4 RCTs[128] 2 RCTs[129]	2886 1360 1307	Placebo Placebo Placebo	OR for abdominal pain response = 1.38 (95% CI, 1.14–1.67) Abdominal pain response rate 44.9% vs 40.0%, $P = .027$ Abdominal pain response rate 44.0% vs 42.3%, $P = .51$
SSRIs (eg, escitalopram, 10 mg o.d.)	Unselected patients with IBS FD	5 RCTs summarized in a meta-analysis[132] 1 RCT[133]	262 195	Placebo Placebo	RR of abdominal pain persistence = 0.82 (95% CI, 0.58–1.16) Upper abdominal pain score 1.4 posttreatment vs 1.2, N/S
TCAs (eg, amitriptyline, 10–30 mg o.d., or imipramine, 50 mg o.d.)	Unselected patients with IBS FD	4 RCTs summarized in a meta-analysis[132] 1 RCT[134] 2 RCTs[133,135]	171 463 194 107	Placebo Placebo Placebo Placebo	RR of abdominal pain persistence = 0.53 (95% CI, 0.34–0.83) OR for $\geq$30% improvement in abdominal pain = 1.66 (95% CI, 1.12–2.46) Upper abdominal pain score 1.1 post-treatment vs 1.2, N/S Epigastric pain score 0.96 vs 1.24 at baseline with imipramine, $P = .026$, compared with 0.96 vs 1.13 at baseline with placebo, $P = .13$
SNRIs (eg., venlafaxine 150 mg o.d.)	Unselected patients with IBS	1 RCT[136]	30	Placebo	Frequency of abdominal pain or discomfort score 3.87 vs 4.93, $P = .03$

Table 1. Continued

Treatment studied	Condition	No. of studies	No. of patients	Comparator	Reported effect
Oloroinab, 10 mg to 100 mg t.i.d.	IBS with abdominal pain	1 RCT[147]	273	Placebo	56.5%, 59.7%, and 56.7% of 10 mg, 25 mg, and 50 mg t.i.d., respectively, achieved a ≥30% improvement in abdominal pain vs 52.9% with placebo, N/S
	Crohn's disease with abdominal pain	1 randomized, open-label study[148]	14	N/A	Change in abdominal pain score from baseline of −4.61 with 25 mg t.i.d. and −4.57 with 100 mg t.i.d.
Pregabalin, 75 mg o.d., or titrated up from 75 mg b.i.d.	Unselected patients with IBS	1 RCT[154]	85	Placebo	Abdominal pain score 28 posttreatment vs 40, $P = .008$
	FD	1 RCT[155]	72	Placebo	Epigastric pain score 3.0 posttreatment vs 4.0, $P = .01$

BDA, British Dietetic Association; b.i.d., twice daily; CI, confidence interval; IBS-C, IBS with constipation; N/A, not applicable; N/S, not significant; o.d., once daily; OR, odds ratio; q.d., once daily; RR, relative risk; t.i.d., 3 times daily.

coexpressed on nociceptors innervating the intestine, and eluxadoline shows high binding affinity for MOR/DOR heterodimers in cell assays[48] and functional interaction between receptors. However, there has been sparse mechanistic study in whole-animal models to clarify the role of this interaction further.[49]

There are other promising strategies to develop safe opiates, such as enhancing endogenous opioids (eg, enkephalinase inhibitors), by developing pH-sensitive opioid analogues,[50] which are only active at sites of inflammation and thus lack the adverse effect profile and addictive potential of conventional opioids. Combinations of subthreshold opioids and cannabinoid receptor 1 (CB1) agonists can provide strong analgesia[51] without adverse effects. Novel delivery systems using nanoparticles of between 1 and 100 nm in diameter, containing opioid cargoes,[52] target intracellular signaling in endosomes and can be delivered intrarectally to act locally within the inflamed colon. To date, most of these strategies are based on preclinical studies and none have been tested adequately in humans. Finally, female rodents are less sensitive to opiate analgesia,[53] and whether these strategies have sex-specific effects would be important to evaluate.

Clinical trials. Few trials have been conducted with new opioid-related drugs in visceral pain, largely due to the negative impact of the opioid crisis. Despite widespread use of loperamide in clinical practice, there is little evidence for this. One 13-week RCT, recruiting patients with IBS with diarrhea (IBS-D), reported pain scores were significantly lower with loperamide.[54] In a second 3-week trial that recruited IBS of all subtypes, the number of painful days was reduced significantly with loperamide, but only in patients with alternating bowel habit.[55] Both trials used historical definitions of IBS, did not conform to guidance for design of treatment trials in DGBI, and many participants did not report abdominal pain at all. More rigorous trials of loperamide are needed, although it is unlikely these will ever be conducted.

In contrast, eluxadoline has been tested rigorously in phase III RCTs at 2 doses, 75 mg or 100 mg twice daily, with data pooled in a prior meta-analysis.[56] Only 100 mg twice daily was superior to placebo for the FDA-recommended end point for abdominal pain, but benefit was modest. In addition, there have been safety issues, with episodes of acute pancreatitis and sphincter of Oddi dysfunction reported.

The Microbiome and Visceral Pain

Advances in pathophysiology. The involvement of gut microbiota in the development of visceral pain is largely based on preclinical studies measuring pain thresholds after transfer of human stool microbiota into germ-free rodents or administration of live biotherapeutics (probiotics) or antibiotics, or both, in rodent models. For instance, germ-free rats colonized with stool microbiota from individuals with IBS display decreased pain thresholds in response to rectal distention.[57] Further insights have been gained from studies involving gnotobiotic mice, revealing the role of

commensal microbes in maintaining normal excitability of gut intrinsic neurons.[58]

Perturbing the gut microbiome during early life using vancomycin leads to visceral hypersensitivity in rats.[59] Conversely, administration of live biotherapeutics, such as *Faecalibacterium prausnitzii, Lactobacillus paracasei* NCC2461, or *Lactobacillus GG*, reduces visceral hypersensitivity and intestinal permeability in preclinical models that alter the early-life microbiome.[60,61] Unlike in early life, antibiotic administration improves visceral hypersensitivity in adult mice,[62] suggesting potential age-dependent effects of the microbiome.

Interestingly, visceral pain responses to colorectal distention vary across the estrous cycle in female mice, but this effect is lost in germ-free animals. Ovariectomy caused visceral hypersensitivity in specific pathogen-free, but not germ-free mice, suggesting an interaction between sex hormones, visceral pain, and the microbiome.[63]

Building on insights from animal models, human studies exploring fecal microbiome changes in patients with IBS have found specific taxa that positively (Proteobacteria)[64] or negatively (*Bifidobacterium* spp) correlate with the severity of pain.[65] Although human microbiome studies have focused largely on the colon, changes in small-intestinal microbial composition, rather than bacterial numbers, appear to differentiate patients with abdominal pain from healthy controls.[66] However, the role of small-intestinal microbiota in the pathophysiology of abdominal pain remains unclear. Together, although findings from preclinical models and human studies underscore a role of the gut microbiome, whether these changes are causal to the development of visceral hypersensitivity or a consequence of changes in diet and gastrointestinal motility is unknown.

Gut microbiota-derived metabolites, neurotransmitters, toxins, and cell wall components have emerged as potential factors underlying the pathophysiology of visceral hypersensitivity. These bioactive compounds can (1) sensitize sensory neurons indirectly by stimulating either enteroendocrine cells, which release serotonin, or immune cells, which release chemokines and cytokines, both of which act on distinct neuronal populations, (2) disrupt the intestinal barrier, allowing passage of potentially noxious stimuli, and (3) activate sensory neurons directly, particularly in instances where barrier function is compromised (Figure 2). Most bacteria-derived compounds are pleiotropic, acting via multiple signaling pathways. Thus, they exert wide-ranging effects. Furthermore, gut microbiota can both synthesize and use neurotransmitters, encompassing excitatory, such as glutamate, histamine, dopamine, and norepinephrine, and inhibitory neurotransmitters, such as γ-aminobutyric acid (GABA).[67] These neurotransmitters allow intercommunication among microbiota members and the host.

Enterochromaffin cells are the primary cell type responsible for peripheral serotonin production. They are polymodal chemosensors, capable of detecting specific luminal signals via an array of receptor pathways and translating them to the enteric nervous system by modulating serotonin-sensitive primary afferent nerves.[68] Catecholamine neurotransmitters, such as norepinephrine and dopamine, initiate the adrenoceptor alpha 2A (Adrα2A) and the transient receptor potential cation channel subfamily C member 4 (TRPC4) signaling cascade.

On the other hand, short-chain fatty acids (SCFAs) and branched-chain fatty acids, such as isovaleric acid and, to a lesser extent, butyrate, activate the olfactory receptor 558 and P/Q type Cav channel within enterochromaffin cells.[68] A multitude of bacterial metabolites, including butyrate, also augment serotonin synthesis within enterochromaffin cells.[69] The role played by serotonin in modulating visceral pain, as well as the critical role of enterochromaffin cells in isovalerate-induced visceral hypersensitivity, is discussed further below.

Pathogen-associated molecular pattern molecules, which include bacterial cell wall components such as lipopolysaccharide, bind to pattern recognition receptors such as Toll-like receptors, are present on immune cells and sensory neurons. Pathogen-associated molecular pattern molecules contribute to visceral hypersensitivity by influencing nociceptors directly or by affecting immune cells indirectly, leading to peripheral sensitization.[70,71] Diet-derived metabolites from bacterial fermentation, such as SCFAs, indole and indole derivatives, and kynurenine, also modulate visceral nociception. Butyrate exerts antinociceptive effects[72] via peroxisome proliferator-activated receptors suppressing the activity of nuclear factor κ-light-chain-enhancer of activated B cells, involved in pain and inflammation.[73,74]

Butyrate also augments intestinal barrier function via activation of hypoxia inducible factor,[75] regulates immune cells via free fatty acid 2/3 receptors,[76] and drives epigenetic changes. Tryptophan is converted by microbes to kynurenic acid[77] or to indole derivatives,[78] both of which exert anti-inflammatory effects via G protein-coupled receptor 35 and aryl hydrocarbon receptor, respectively,[79,80]

Gut bacteria play an important role in determining the luminal bile acid and protease pool. Bile acid metabolites, including deoxycholic acid, regulate pain through the activation of G protein-coupled bile acid receptor 1, and are present in both primary sensory neurons and macrophages. Proteases contribute to visceral hypersensitivity by targeting intestinal barrier function[81] as well as by signaling directly through protease activated receptor 2, present on neurons.[82] The luminal protease pool depends on the balance between bacterial proteases[83] and suppression of host proteases by bacteria harboring β-glucuronidases.[81]

The identification of distinct microbiota-driven mechanisms opens the door for novel therapeutic strategies. Currently, microbiota-targeted interventions largely focus on augmenting intestinal barrier function. In preclinical studies, fiber maintained both microbial diversity and barrier function,[84] and a diet low in fermentable oligosaccharides, disaccharides, monosaccharides and polyols (FODMAP) was found to preserve barrier function by decreasing lipopolysaccharide-mediated mast cell activation.[85]

Clinical trials. There are a multitude of methods to manipulate the microbiome, and thereby microbial metabolites, as a means of treating abdominal pain. SCFA enemas

have been studied in IBD, but trials have not reported an effect on abdominal pain.[86–88] Fiber has been assessed in IBS, but few trials report abdominal pain outcomes.[89,90] One 12-week RCT found there was no benefit of psyllium, a soluble fiber, over placebo,[89] but in another trial of psyllium, bran, or placebo, significant improvements in abdominal pain occurred with both psyllium and bran at several time points.[90]

A network meta-analysis of 12 trials studied the effect of a low FODMAP diet on abdominal pain.[91] It was superior to a sham diet but was not superior to standard British Dietetic Association dietary advice for IBS or habitual diet. In contrast, in a RCT comparing a 4-week low FODMAP diet with a sham diet in patients with quiescent IBD with persistent gastrointestinal symptoms, abdominal pain severity and days with abdominal pain did not differ.[92] In another 6-week trial of a low FODMAP diet vs normal diet in patients with IBD in remission with ongoing gastrointestinal symptoms, response for abdominal pain frequency, but not severity, was significantly higher with the low FODMAP diet.[93] Abdominal pain response rates with rifaximin, a minimally absorbed antibiotic, according to the FDA-recommended end point, were reported in a meta-analysis.[56] There was no benefit with rifaximin over placebo.

Although there have been multiple RCTs of fecal microbial transplant (FMT) in both IBS and IBD, summarized in prior meta-analyses,[94,95] few report impact of FMT on abdominal pain. Two RCTs of FMT in IBS studied this end point.[96,97] One 12-week trial of a single FMT via nasojejunal tube in IBS with predominant bloating reported abdominal pain scores were significantly reduced.[96] In the second RCT, 30 mg or 60 mg of a single FMT via gastroscopy led to a significant reduction in abdominal pain at 3 months vs placebo.[97] One RCT comparing FMT with usual therapy in active ulcerative colitis reported abdominal pain scores improved significantly with FMT at 2 weeks compared with baseline, but also improved significantly in the usual therapy arm.[98]

Gelsectan, a prebiotic with mucoprotective and bifidogenic effects, which may reinforce the intestinal barrier, was studied in 1 crossover trial.[99] The number of participants with totally to slightly unacceptable abdominal pain was reduced from baseline to 4 weeks compared with placebo. Finally, in a meta-analysis certain combinations of probiotics, *Lactobacillus*-containing strains, *Saccharomyces cerevisiae* I-3856, and *Bifidobacteria*- and *Bacillus*-containing strains improved abdominal pain, but certainty in the evidence was low to very low across the studies, with heterogeneity between individual trials in most analyses.[100]

Histamine and Visceral Pain

Pharmacology and preclinical studies. Histamine functions as a paracrine signaling molecule that can activate nociceptors in the gastrointestinal tract (Figure 3). It is a member of the biogenic amine family and is synthesized from L-histidine exclusively by L-histidine decarboxylase.[101] Histamine signaling to nociceptors in the gut could originate from 2 sources: intestinal tissue or the lumen. In tissue, it is stored in high concentrations, predominantly in mast cells, but also in basophils and eosinophils. However, other cells in the gut also express histidine decarboxylase, including macrophages, neutrophils, platelets, and dendritic cells, and can synthesize and release histamine but do not store it.[102] In the lumen of the gastrointestinal tract, there are 3 possible sources: synthesis by microbiota, ingestion of histamine-rich foods, and histamine released from tissues that permeates into the lumen.

Histamine is metabolized by 2 dominant pathways, histamine-*N*-methyltransferase and diamine oxidase,[103] resulting in *N*-methyl histamine and imidazole acetaldehyde metabolites, respectively. These metabolites also exhibit biological activity (eg, *N*-methyl histamine), and the preponderance of the pathways may differ between mast cells and microbiota.

Histamine, and possibly some metabolites, can activate 4 GPCRs, H_1 to H_4.[101] These GPCRs signal intracellularly via Gq and cyclic adenosine monophosphate to modulate passive and voltage-gated ion channels on nociceptors and other effector pathways in nonneuronal cells. The distribution of these receptors within the gut suggests histamine activates pain signaling directly through activation of neurons and indirectly via immune cell activation (Figure 3).

In humans,[104] H_1 receptors are found on connective tissues cells, immune cells, enterocytes, smooth muscle cells, and nerves, H_2 receptors are found on gastric parietal cells, enterocytes, immunocytes, enteric nerves, and smooth muscle cells, and H_4 receptors are expressed on immune cells, blood vessels, nerves, and enterocytes. H_3 receptors have yet to be identified in humans.

Previous studies highlight the role of histamine in patients with IBS, demonstrating increased levels of histamine (and proteases) in mucosal biopsy specimens from patients compared with healthy controls, evidence of mast cell activation, and the ability of histamine in tissue supernatants from patients to exaggerate activation of rodent and human nociceptive neurons.[105] Sex differences have not been described. More recent studies show increases in duodenal eosinophils in patients with FD, another source of tissue histamine, implicating a role in abdominal pain in this disorder.[106] The triggers resulting in abnormal mast cell-histamine signaling observed in these patients have been unclear, but recent preclinical studies suggest multiple possible etiologies, as outlined below.

When mice develop a self-limiting bacterial colitis and are exposed simultaneously to a food antigen, and then reexposed to the food antigen alone after resolution of infection, they lose oral tolerance to the food antigen.[14] This leads to visceral hypersensitivity and mast cell activation with histamine release. This exaggerated pain signaling was blocked by an H_1 receptor antagonist, which inhibits histamine signaling to neurons, and by an IgE antibody, which prevents mast cell activation. Tissues exhibited elevated IgE levels, consistent with loss of oral tolerance, but there was no systemic increase in IgE, highlighting immune activation was confined to the intestine.

Injection of common antigens into the rectal mucosa in patients with IBS caused greater wheal and flare responses,

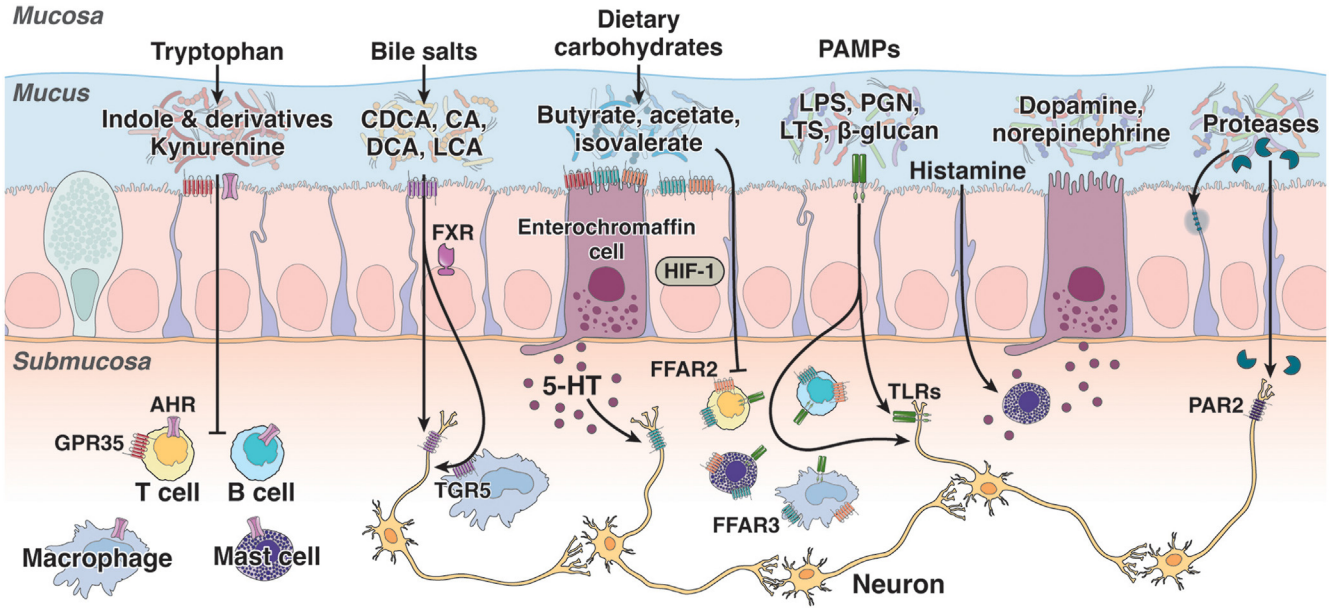

Figure 2. Mechanisms underlying gut microbiome-driven visceral nociception. Gut microbiome-derived products can sensitize peripheral nociceptors directly or act indirectly by stimulating immune cells or enterochromaffin cells, or both, to release cytokines, chemokines, or serotonin, or a combination of these, respectively. The gut microbiome can also modulate intestinal barrier function by altering the luminal bile acid and protease pool or through metabolites such as butyrate. AHR, aryl hydrocarbon receptor; CA, carboxylic acid; CDCA, chenodeoxycholic acid; DCA, deoxycholic acid; FFAR, free fatty acid receptor; FXR, farnesoid X receptor; GPR35, G protein-coupled receptor 35; LCA, lithocholic acid; LPS, lipopolysaccharide; LTS, leukotrienes PAMPs, pathogen-associated molecular patterns; PAR2, protease activated receptor 2; PGN, peptidoglycan; TGR5, Takeda G protein-coupled receptor 5; TLR, Toll-like receptor.

compared with healthy volunteers, consistent with the hypothesis that some patients with IBS are sensitized to food antigens.[14] Recent preliminary studies suggest psychological stress can also induce loss of oral tolerance to food antigens and lead to mast cell-histamine–mediated visceral hypersensitivity in both the small intestine and colon, a feature observed in many patients with IBS.[107]

Histamine production by the microbiota may be stimulated by poorly absorbed complex carbohydrates. In a study using germ-free mice to create a humanized IBS mouse model with fecal microbiota from patients with IBS and healthy controls,[108] mice given fecal samples from patients with IBS who were high histamine producers, based on stool and urine samples, exhibited visceral hyperalgesia. This exaggerated pain signaling was blocked by H_1- and H_4-receptor antagonists, suggesting several signaling pathways were involved (Figure 3). Some of the histamine in the luminal samples could originate from the host (eg, mast cell degranulation). However, high histamine producers were found to have microbial species, including *Klebsiella aerogenes,* in their stool that could make up to 100 times more histamine than those without.

Clinical trials. Drugs targeting histamine receptors or stabilizing mast cells have not been well studied in gastrointestinal diseases. An 8-week trial of ketotifen, a H_1-receptor antagonist, in IBS reported a significant improvement in abdominal pain over placebo.[109] Ebastine, another H_1-receptor antagonist, has been assessed in IBS.[110,111] A 12-week proof-of-concept RCT found rates of relief of abdominal pain were numerically higher with ebastine, but not significantly so. In a subsequent phase IIb placebo-controlled trial, rates of abdominal pain improvement were higher with ebastine, although this was not significant.[111] The effect of the mast cell stabilizer disodium cromoglycate on abdominal pain was studied in a RCT in IBS-D.[112] In this 6-month study, compared with no treatment, significantly more patients randomized to disodium cromoglycate experienced abdominal pain improvement.

Transient Receptor Potential Vanilloid 1 and Visceral Pain

Pharmacology and preclinical studies. TRPV1 is a nonselective ligand-gated cation channel that is highly enriched in gastrointestinal tract nociceptors. It is activated by polymodal stimuli, including mechanical stretch, noxious heat, low pH, exogenous chemical irritants, such as capsaicin (the active ingredient in chili peppers), and endogenous lipid metabolites of arachidonic acid (eg, the endocannabinoid anandamide[113]). Selective ablation of colon-projecting TRPV1-expressing neurons decreased nociception in response to colorectal distention in mice,[22] highlighting its critical role in visceral pain. Estrogens can modulate lumbosacral dorsal root ganglia TRPV1 expression, suggesting a potential mechanism for sex differences in visceral pain.[114]

Sensitization of TRPV1 by inflammatory mediators, such as histamine,[110] is one of the key pathways in mediating

peripheral visceral hypersensitivity, as discussed above. However, TRPV1 expression does not necessarily correlate with receptor sensitization. For example, in patients with IBS who were hypersensitive to rectal distention, rectal application of capsaicin caused increased pain perception. No change in TRPV1 expression was noted when comparing hypersensitive and normosensitive patients with IBS, suggesting that although TRPV1 expression is important, additional factors, such as receptor sensitization or central factors, or both, are necessary in mediating visceral pain.[115]

Although most of the studies have focused on TRPV1 sensitization in IBS and FD,[113] TRPV1 may also play a role in chronic visceral pain in patients with IBD in remission. Rectal TRPV1 expression was increased in patients with IBD in endoscopic remission with chronic visceral pain and correlated with patient-reported symptoms.[116] Visceral hyperalgesia was TRPV1-dependent in postinflammatory mice,[117] and they also displayed increased SCFA-producing microbiota and stool SCFA content. These microbial-derived SCFAs increased capsaicin-evoked calcium responses in the postinflammatory state, suggesting that microbial metabolites can sensitize TRPV1.[118]

Clinical trials. Peppermint oil, as well as being a smooth muscle relaxant, may have effects on TRPV1 signaling. A meta-analysis showed it was more efficacious than placebo for abdominal pain.[119] However, benefit was modest, with heterogeneity between studies, and most trials did not use FDA-recommended end points. Although capsaicin stimulates the TRPV1 receptor, leading to worsening abdominal pain, repeated administration downregulates the receptor. A 6-week trial in IBS showed abdominal pain scores were significantly lower, compared with baseline, in patients receiving red pepper pills compared with those receiving placebo.[120] A similar 5-week study in FD demonstrated a significant reduction in epigastric pain scores with red pepper vs placebo.[121] However, patients in both trials randomized to red pepper dropped out due to pain exacerbations.

Serotonin (5-Hydroxytryptamine) and Visceral Pain

Pharmacology and preclinical studies. The monoamine neurotransmitter 5-hydroxytryptamine (5HT) plays an integral role in initiation of intrinsic gut reflexes regulating motility, secretion, and vasodilation. It also participates in the pathogenesis of visceral pain via afferent nerve 5-HT_3 and 5-HT_4 receptors; drugs that modulate these receptors have been used extensively in the treatment of visceral hypersensitivity.[122] The actions of 5HT are terminated by the serotonin selective reuptake transporter, a peripheral target of selective serotonin reuptake inhibitors (SSRIs), tricyclic antidepressants (TCAs), and serotonin norepinephrine reuptake inhibitors[122] (SNRIs).

Genetic polymorphisms in the 5-HT_3 receptor and the serotonergic synthetic enzyme, tryptophan hydroxylase, are associated with increased IBS susceptibility, whereas SSRI transporter polymorphisms are associated with both IBS and FD.[123] Multiple studies report changes in 5HT synthesis, reuptake, and release in IBS,[124] suggesting dysregulated 5HT signaling contributes to the pathophysiology of visceral pain. Surprisingly, few studies evaluating the role of TCAs in visceral pain have been performed in rodent models.

Although 5HT can be secreted by enteric neurons and mucosal mast cells, most of the body's 5HT is synthesized and stored by enterochromaffin cells.[122] Enterochromaffin cells are electrically excitable, and display axon-like basal processes, forming functional connections with extrinsic and intrinsic afferent neurons, termed neuropods.[68,125] Enterochromaffin cells function as luminal sensory transducers, releasing 5HT in response to dietary nutrients and microbial products, as well as mucosal distortion via mechanosensitive Piezo-02 channels.[126] 5HT release by enterochromaffin cells, thus initiates intrinsic gut reflexes and stimulates extrinsic nerves.

A recent study evaluated the role of a mucosal afferent-enterochromaffin cell circuit in the pathogenesis of visceral hypersensitivity using transgenic mice,[26] where enterochromaffin cells could be activated or silenced selectively. Direct activation of enterochromaffin cells elicited 5HT release and was sufficient to cause both acute and chronic visceral hypersensitivity to colorectal distention. Remarkably, activation of enterochromaffin cells was sufficient to elicit anxiety-like behavior in mice. These effects were inhibited by the 5HT3 antagonist alosetron, which decreased mucosal afferent activity. Conversely, silencing activity of enterochromaffin cells attenuated 5HT release and visceral hypersensitivity mediated by the microbial metabolite, isovalerate, in male mice. The mucosal afferent-enterochromaffin cell circuit demonstrated high tonic activity in female, but not male, mice suggesting a sex-specific contribution to pain signaling. Together, these data demonstrate that the enterochromaffin cell-mucosal afferent circuit plays an essential role in pathogenesis of visceral hypersensitivity.[26]

It is possible that the GC-C pathway also regulates 5HT secretion from enterochromaffin cells. GC-C is expressed not only by enterocytes but also by a subtype of monoamine synthesis-expressing neuropods enriched in the proximal intestine of mice.[36] GC-C enriched neuropods formed functional connections with nociceptors in cocultures and caused spontaneous nociceptor activation, which was abolished by linaclotide. The antinociceptive effects of linaclotide on the response to colorectal distention were lost in mice that were deficient in neuropod GC-C.[36] Thus, it is possible that enterochromaffin GC-C activation regulates 5HT tone, but whether this mechanism is active in vivo is unclear.

Clinical trials. The efficacy of the 5HT_3-receptor antagonists alosetron and ramosetron, according to the FDA-recommended end point for abdominal pain, has been reported in multiple trials in IBS-D, pooled in a meta-analysis.[56] Ramosetron, 2.5 μg daily and 5 μm daily, and alosetron, 1 mg twice daily, were superior to placebo, although alosetron has been associated with ischemic colitis. Varying doses of ondansetron, another 5HT_3-receptor antagonist with a long history of safety, were assessed in 3 trials in IBS-D, summarized in another meta-analysis.[127] The drug was not superior to placebo for pain.

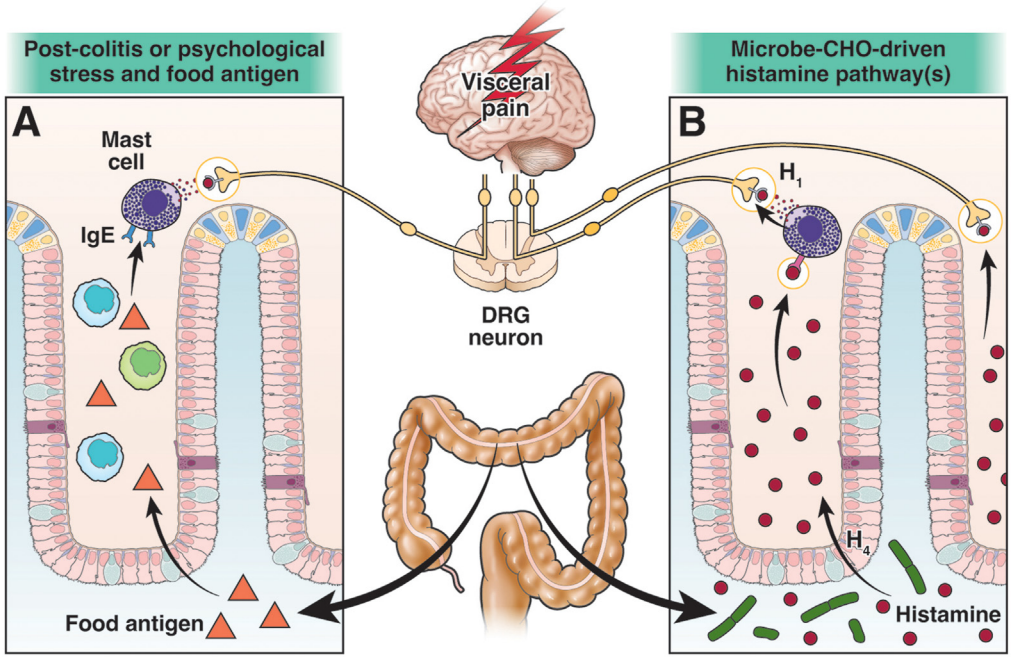

Figure 3. Novel mechanisms causing increased histamine signaling to intestinal nociceptors. (*A*) This schema shows that after combined food antigen (*red* triangle) and acute self-limiting colitis or combined food antigen and psychological stress exposure, reexposure to food antigen alone triggers increased IgE release within the intestine (which is not systemic), causing mast cell degranulation within the intestinal wall. The ensuing histamine release causes nociceptor sensitization and increased pain signaling. (*B*) This schema shows that ingestion of poorly absorbed complex carbohydrates (CHO) (eg, FODMAPs) can stimulate microbial production of histamine. Patients with *Klebsiella aerogenes* produce up to 100 times more histamine than those lacking this bacterium in their stool samples. Luminal histamine stimulates H_4 and H_1 receptors, leading to mast cell degranulation with ensuing nociceptor sensitization and increased pain signaling. DRG, dorsal root ganglia.

For $5HT_4$-receptor agonists, in a pooled analysis of data from 4 RCTs in IBS, tegaserod, 6 mg twice daily, was more efficacious for pain than placebo.[128] In two 6-week trials of tegaserod in FD, 6 mg twice daily was superior to placebo for abdominal pain in 1 RCT but not the other.[129] Safety issues arising from cardiovascular and cerebrovascular ischemic events led to the withdrawal of tegaserod. Although it was reintroduced briefly, tegaserod is now no longer available. Prucalopride was assessed in chronic constipation and was efficacious,[130] but no RCTs report its efficacy in improving abdominal pain, and it has never been tested in IBS or FD.

Although SSRIs, TCAs, and SNRIs are antidepressants, in the context of treating abdominal pain, they act as gut-brain neuromodulators involving, at least in part, 5-HT[131]; discussion of the central actions of these compounds is beyond the scope of this review. SSRIs have been assessed in IBS and FD, with no impact on abdominal pain in IBS in a prior meta-analysis,[132] and a reduction in pain scores in FD in a single RCT of escitalopram, 10 mg once daily, but with no benefit over placebo.[133] TCAs, however, were more efficacious than placebo for abdominal pain in IBS in a meta-analysis of 4 RCTs[132] and more recently in a 6-month trial in 463 patients.[134] In one 12-week trial in refractory FD, imipramine led to a significant reduction in epigastric pain scores vs placebo,[135] but another trial of amitriptyline demonstrated no benefit.[133] The SNRI venlafaxine was assessed in a single 12-week RCT in IBS; abdominal pain frequency scores were reduced significantly compared with placebo.[136] An RCT of FD did not report its effect on abdominal pain.[137]

Cannabinoids and Visceral Pain

Pharmacology and preclinical studies. Cannabinoids are widely used alternative therapies to treat abdominal pain in both IBD and IBS.[138,139] The actions of cannabinoids are mediated via the endocannabinoid system, which regulates gastrointestinal motility, secretion, immune function, intestinal permeability, and visceral hypersensitivity.[140]

The classical components of the endocannabinoid system are the endogenous cannabinoid ligands, anandamide, and 2-arachidonoylglycerol, as well as their biosynthetic and degradative enzymes. These are found throughout the microbiota-gut-brain axis, including the epithelium, enterochromaffin cells, enteric nervous system, and immune system, as well as extrinsic afferent nerves, where they primarily exert an antinociceptive effect. Anandamide is also an agonist at TRPV1. Thus, endocannabinoids have both pronociceptive and antinociceptive effects, depending on the receptor.[140] Interestingly, commensal bacteria can produce endocannabinoid-like molecules,[141] although whether a microbial source of endocannabinoid-like molecules plays a role in visceral hypersensitivity is unknown.

In animal models of stress-induced visceral hypersensitivity and in postinflammatory models, CB1 and CB2

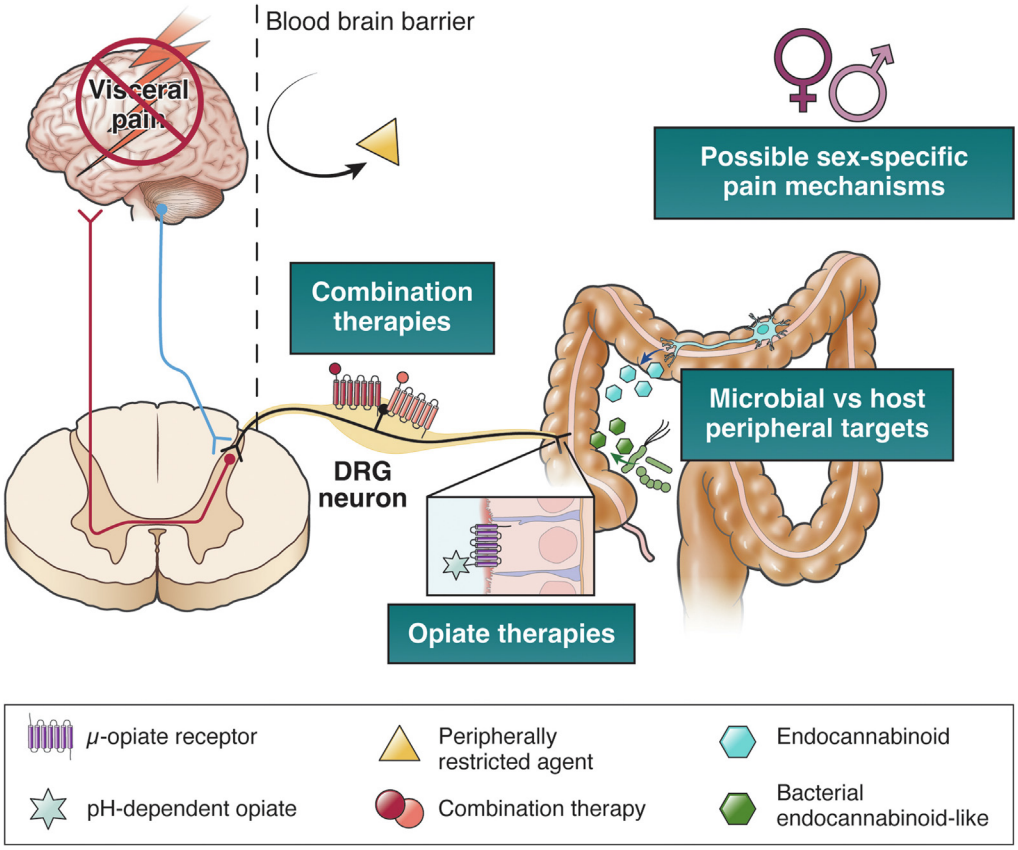

Figure 4. Areas for future investigation in peripheral mechanisms of visceral pain. There is a need to identify peripheral mechanisms underlying visceral pain and develop novel therapeutic agents to treat patients. Identification of microbial vs host sources of peripheral targets (eg, GABA, endocannabinoids, 5HT), is one such mechanistic area. Evaluation of the relative contribution of each pathophysiologic mechanism to nociceptor sensitization in individual patients, and thus use of combination therapies targeting these mechanisms, is key. For opiates, some promising strategies for the development of safe yet effective opiate therapies are the development of pH-sensitive opiate analogues active at the site of inflammation, use of peripherally restricted agents or subthreshold combinations of opiates, and CB1 receptor agonists. Because chronic visceral pain is more common in women, future studies should evaluate whether pain mechanisms are sex-specific and whether treatments should be used in a sex-specific manner. ♀, female; ♂, male.

agonists decrease the visceromotor response to colorectal distention.[140,142–144] Endocannabinoids can either exert their antinociceptive actions directly via CB1 and CB2 receptors expressed on nociceptors[142,145] or indirectly via down-regulation of mast cell or macrophage activation.[140] However, clinical use of cannabinoids is hampered by psychotropic adverse effects. Accordingly, there has been interest in synthesizing peripherally restricted cannabinoid receptor agonists.[142–144]

A recent preclinical study of the peripherally restricted CB2 receptor agonist, olorinab, was performed in rodent models of acute colitis and postinflammatory visceral pain.[142] Olorinab reversed the colitis-induced hypersensitivity to colorectal distention in both the acute and postinflammatory state; no effects on visceral pain were seen when olorinab was given to controls. Olorinab was able to decrease mechanosensitivity of ex vivo afferent nerves in a dose-dependent manner, both in acute colitis and in the postinflammatory state, although CB2 expression was not up-regulated in afferent nerves compared with controls.[142] Unfortunately, only male mice were evaluated in this study, although CB2 expression is increased in female patients with IBS.[146] These data suggest CB2 receptors on visceral afferents are sensitized by inflammation and, in turn, play a regulatory anti-nociceptive role.

Clinical trials. In a 12-week phase II dose-ranging study of olorinab in IBS, the proportion of patients experiencing improvement in abdominal pain was not significantly higher with any dose studied.[147] However, in those with moderate to severe pain at baseline, abdominal pain scores were significantly improved with 50 mg 3 times daily. No placebo-controlled trials of this drug in IBD have been conducted, although an 8-week open-label randomized study recruiting patients with Crohn's disease who reported abdominal pain found a significant reduction in pain scores from baseline with olorinab.[148] There is no evidence for other drugs acting on cannabinoid receptors for treating abdominal pain in gastrointestinal disorders.[149]

γ-Aminobutyric Acid and Visceral Pain

Pharmacology and preclinical studies. Functional GABA receptors have been identified in the nerve terminals

of colonic afferents. The activation of GABA receptors ($GABA_A$ and $GABA_B$) by endogenous GABA decreases sensitivity of colonic afferents, whereas $GABA_A$ activation also reduces visceral pain perception.[150] Functional GABAergic transmission has also been found in nociceptors, producing strong analgesic effects.[151] In addition to endogenous production, certain bacteria expressing glutamate decarboxylase, can produce GABA from glutamate.[67] In rodent models, GABA-producing bacteria have an analgesic effect in stress-induced[152] and fecal-retention[153] models of visceral hypersensitivity.

Clinical trials. There has been 1 RCT of pregabalin in both FD and IBS, but no trials in other painful gastrointestinal disorders and no trials of gabapentin. Abdominal pain scores were significantly lower in patients assigned to titrated pregabalin vs placebo in a 12-week trial in IBS.[154] Similarly, pregabalin, 75 mg daily, was superior to placebo for epigastric pain scores in an 8-week trial in FD.[155]

Conclusions

Chronic visceral pain represents a substantial burden to patients. Despite the potential for the evidence-based treatments described above, a need remains for the development of novel therapeutics to treat sensitization of peripheral pain pathways effectively. Future directions should include the identification of microbial vs host sources of peripheral targets (eg, GABA, endocannabinoids, 5HT), similar to current work evaluating histamine (Figure 4).

With respect to the microbiome, research should avoid observational-based community profiling and focus on mechanistic approaches evaluating how microbiota or microbial products, or both, interact with nociceptors. Methods to test the relative contribution of each pathophysiologic mechanism to the sensitization of peripheral nociceptors and their role in overlapping pain syndromes in individual patients is also required. This would allow the use of specific drug combinations to target multiple mechanisms synergistically.

Given the sex bias of chronic visceral pain, future studies should evaluate whether pain mechanisms are sex-specific or whether treatments should be used in a sex-specific manner. Identifying whether differing or similar peripheral mechanisms are involved in the development of chronic visceral pain in patients with IBD vs painful DGBI will be important. Finally, evaluation of the relative contribution of peripheral vs central sensitization to symptoms would be important to individualize patient therapy. Continued multidisciplinary collaboration between clinician-scientists and bench-based scientists with the use of innovative reverse translational approaches is necessary to advance this field, identify new target pathways, and improve the clinical management of patients.

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Conflicts of interest
These authors disclose the following: Stephen Vanner is cofounder of pHarm Therapeutics Inc, a company that is developing pH-sensitive analgesics that selectively target sites of inflammation where pain originates. Purna C. Kashyap is an ad hoc consultant for Pendulum Therapeutics and Intrinsic Medicine and holds the patent US20170042860A1 "Methods and materials for using *Ruminococcus gnavus* or *Clostridium sporogenes* to treat gastrointestinal disorders" for use of tryptamine producing bacteria to treat gastrointestinal disorders. Mayo Clinic and Purna C. Kashyap have a financial interest related to use of tryptamine-producing bacteria. The remaining authors disclose no conflicts.

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