Role of Principal Ionotropic and Metabotropic Receptors in Visceral Pain

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Visceral pain is the most common form of pain caused by varied diseases and a major reason for patients to seek medical consultation. It also leads to a significant economic burden due to workdays lost and reduced productivity. Further, long-term use of non-specific medications is also associated with side effects affecting the quality of life. Despite years of extensive research and the availability of several therapeutic options, management of patients with chronic visceral pain is often inadequate, resulting in frustration for both patients and physicians. This is, most likely, because the mechanisms associated with chronic visceral pain are different from those of acute pain. Accumulating evidence from years of research implicates several receptors and ion channels in the induction and maintenance of central and peripheral sensitization during chronic pain states. Understanding the specific role of these receptors will facilitate to capitalize on their unique properties to augment the therapeutic efficacy while at the same time minimizing unwanted side effects. The aim of this review is to provide a concise review of the recent literature that reports on the role of principal ionotropic receptors and metabotropic receptors in the modulation of visceral pain. We also include an overview of the possibility of these receptors as potential new targets for the treatment of chronic visceral pain conditions.

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Key Words
Ligand-gated ion channels; Receptors, metabotropic glutamate; Visceral pain

Introduction

Chronic visceral pain is one of the most frequent and debilitating disorders in the general population, critically impacting economy and quality of life.1 Epidemiologic studies fail to identify the number of patients with this condition, perhaps due the lack of clear pathologic features associated with visceral pain conditions.2 In spite of this problem, surveys have shown prevalence rates among adults of 25% for intermittent abdominal pain, 20% for chest pain and 16-24% for pelvic pain in women.3 Functional gastrointestinal disorders underlie the most prevalent forms of visceral pain. Irritable bowel syndrome (IBS) is one functional gastrointestinal disorders characterized by abdominal pain, discomfort and altered bowel habits and creates tremendous pressure on the healthcare system affecting an estimated 10-15% of Europe and U.S. populations with consequent costs estimated to exceed US$ 40 billion.4,5 Pain is also one of the presenting
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Visceral pain pathway.

MPG, major pelvic ganglion; PN, pelvic nerve; STT, spino-thalamic tract.

Figure.

Symptoms in about 50-70% of patients experiencing the initial onset or exacerbations of inflammatory bowel disease. Even though visceral pain may be the response to noxious stimuli as distension or inflammation, the severity of pain does not always reflect the severity of the condition causing the pain. Although the precise pathophysiology of chronic visceral pain is still far from being elucidated, recent reports indicate that this disorder might be associated with a dysregulation at multiple levels of the brain-gut axis and might involve both the central nervous system (CNS) and the peripheral nervous system (Figure). In addition, aberrant central processing, as well as abnormalities within the stress responsive systems has also been reported to cause visceral pain. Moreover, early-in-life exposure to noxious and/or inflammatory stimuli has been reported to enhance the susceptibility of the organism to subsequent pathological challenges in the adult life by producing long-lasting neuroanatomical and neurophysiological changes in the nociceptive system. Nevertheless, our understanding of the etiology, pathophysiology and natural history of chronic abdominal pain has significantly increased in the past decade, in part because of the new tools to investigate the nervous system.

Although development of visceral pain is considered to be an important defensive mechanism, development of hypersensitivity represents a significant clinical problem and is likely to be one of the major factors involved in the pathogenesis of abdominal and chest pain in functional bowel disorders. Research over the years has shown that the pathophysiology of the visceral pain is extremely complex. Hence, development of effective analgesics for the treatment of visceral pain has become an ongoing challenge for the pharmaceutical industry. For years, treatment recommendations for visceral pain were the same as that for somatic pain. However, of late it is widely accepted that visceral pain processing is distinctly different from somatic pain and as a result it should be treated differently from somatic pain. Currently, analgesics (opiates, nonsteroidal anti-inflammatory drugs, and benzodiazepine), antispasmodics and antidepressants are the most common medications for acute as well as chronic visceral pain conditions; yet they are always associated with the prominent adverse effects like addiction and constipation. Furthermore, the development of analgesic tolerance, inadequate pain relief and altered pain sensitivity with prolonged opioid use has also proved an unfortunate obstacle for their clinical applications. Complementary and alternative medicinal approaches like probiotics, herbal supplements, and electro-acupuncture are also being widely used to treat visceral pain although with varying degree of success.

For better understanding of the increase of visceral sensitivity in patients, a broader knowledge of the molecular components involved in the pathophysiology visceral pain is necessary. Accumulating evidence from extensive clinical and basic research in the last decade have shown the involvement of several receptors and ion channels in the development and maintenance of chronic visceral pain conditions. Receptors and channels have always been traditional targets for drug development to battle against a variety of diseases including chronic pain. Based on the epidemiological evidence and convincing basic science data, there is growing interest in the pharmaceutical industry to expand basic science and clinical research data and to explore specific potential targets for visceral pain therapy for this underserved patient population. Hence, a comprehensive review of the major re-
Table. List of Ionotropic and Metabotropic Receptors and Their Ligands

| Receptor               | Agonist                      | Antagonist                           |
|------------------------|------------------------------|--------------------------------------|
| GABA A receptor        | GABA, Muscimol, HZ166        | Baclofen                              |
| GABA B receptor        | GABA, Baclofen, gamma-hydroxybutyrate, CGP7930, ADX71441, ADX71943 | Phaclofen, CGP-35348                 |
| NMDA receptor          | NMDA, glutamate, aspartic acid | AP-5, AP-7, Ketamine                 |
| Kainate receptor       | Kainic acid                  | LY382884, CNQX, DNQX                 |
| AMPA receptor          | AMPA, glutamate              | NBQX, Kynurenic acid                 |
| Opioid receptor        |                              |                                      |
| MOR                    | Morphine, oxycodone          | -                                    |
| KOR                    | Oxycodone, asimadoline       | -                                    |
| DOR                    | Enkephalin, cannabidiol, tetrahydrocannabinol | Buprenorphine |
| Cannabinoid (CB) receptor | CB1 Dronabinol               | -                                    |
| CB2                    |                              |                                      |
| Serotonin (5-HT) receptor | 5-HT1 DPAT                   | -                                    |
|                        | 5-HT2                        | -                                    |
|                        | 5-HT3                        | -                                    |
| TRP channels           | TRPV1 SB 366791              |                                      |
|                        | TRPA1                        | -                                    |
|                        | TRPV4                        | -                                    |
|                        | TRPC4                        | -                                    |
|                        |                              |                                      |
|                        |                              |                                      |

GABA, gamma aminobutyric acid; NMDA, N-methyl-D-aspartate receptor; AMPA, α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid receptor; MOR, μ-opioid receptor; KOR, κ-opioid receptor; DOR, δ-opioid receptor; DPAT, 8-hydroxy-DPAT hydrobromide; TRP, transient receptor potential.

Receptors and ion channels involved in the modulation of visceral pain is necessary. While discussing all the receptors and ion channels will be beyond the scope and limitation of this article, we have limited our focus on the recent reports which have strongly indicated a role for these receptors and ion channels in modulating chronic visceral pain. These channels and receptors might in future be considered as potential targets for novel analgesics to treat visceral pain (Table). The contribution of ionotropic (membrane ion channels) and metabotropic (coupled to second messenger systems) receptors to visceral nociceptive processing is discussed below.

**Gamma Aminobutyric Acid Receptors**

Gamma aminobutyric acid (GABA), the major inhibitory neurotransmitter in the brain, is essential for the overall balance between neuronal excitation and inhibition that is vital to normal brain function. GABA produces neuronal inhibition by acting on a diversity of membrane-bound receptors. These receptors can be divided into 2 major types: ionotropic receptors that are ligand-gated ion channels (GABA_A receptors), and metabotropic receptors that are G-protein coupled receptors (GABA_B receptors). GABA_A receptor channels are ubiquitous in the mammalian CNS mediating fast inhibitory neurotransmission by becoming permeant to chloride ions. Similarly, GABA_B receptors are expressed by almost all neurons as well as glial cells in the CNS and their activity influences many neural systems and behavioral states. Several recently published reports indicate that both GABA_A and GABA_B receptors are involved in the pathophysiology of chronic pain. While majority of the reports confirm the role of GABA receptors in somatic pain, few studies have also reported on their involvement in visceral pain. GABA_A receptors have been widely reported to be involved in several models of somatic pain and are actively pursued as potential targets for novel analgesic drugs. In contrast, their role in visceral pain condition was confirmed only recently. Sengupta et al reported that down regulation of microRNA-mediated GABA_A receptor subunit in adult spinal cord following neonatal cystitis was responsible for the development of chronic visceral pain in rats. Microinjection of muscinol, (GABA_A receptor agonist), into raphe magnus of rats reduced colorectal distension evoked suppression of withdrawals indicating that evidence that raphe magnus neurons contribute to this antinociception via the GABA_A receptors.

Studies have shown that GABA_B receptor is involved in modulating mechanosensory traffic through the vagus nerve and...
nociception. While the exact mechanism is not known, GABAB receptors and clinical conditions to test somatic and visceral sensitivity. Activation of GABAB receptors were found to down regulate calcium channels which may be the mechanism of antinociceptive effect. Activation of GABAB receptors were found to down regulate calcium channels and activate the inward rectify potassium channels which was found to improve repolarization of neurons. In rodent studies, baclofen was found to reduce afferent firing from a colorectal distension which is a well-established and widely used model for noxious visceral stimuli. This effect was found to occur in a dose-dependent manner and was unrelated to smooth muscle relaxation or improved colon compliance. Several other studies also reported that baclofen significantly reduces the visceromotor responses from colorectal distension. Baclofen was also found to inhibit the expression of c-fos in the dorsal horn to experimental colitis and capsaicin-induced bladder irritation in rodent models of visceral pain. Hara et al reported that intrathecal diltiazem (a calcium channel blocker) in combination with muscimol (GABA	extsubscript{A} agonist) or baclofen (GABA	extsubscript{B} agonist) was found to potentiate the GABA agonists-induced visceral antinociception without increasing motor paralysis. The authors report that when muscimol was administered with diltiazem, the increase in the threshold for colorectal distension was significantly larger than muscimol alone. Similarly, the colorectal distension threshold after the combination of baclofen and diltiazem also showed a significantly larger increase than that seen after baclofen alone. In addition to that, motor paralysis observed with muscimol did not increase when muscimol was co-administered with diltiazem. Brusberg et al reported that activation of GABA	extsubscript{B} receptors was found to produce antinociceptive effects in a rat model of mechanically induced visceral pain (noxious colorectal distension). Despite these effects, clinical use of baclofen has been limited by its short duration of action, narrow therapeutic margin and side effects, including sedation, dizziness, nausea, muscle weakness and mental confusion.

Positive allosteric modulation (PAM) of GABA receptors may represent a valid approach in the treatment of visceral pain conditions, with the possibility of an improved safety profile compared to the effect of agonist. Kalinichev et al demonstrated that the GABA B receptor positive allosteric modulator, ADX71441, significantly improved micturition indices and cystometry variables in 2 models of overactive bladder, a disorder caused in part by urothelial dysfunction, increased excitability of the detrusor and abnormal functioning of neuronal circuits serving the micturition reflex. Similarly, ADX71943, a novel, potent and selective GABA	extsubscript{B} receptor PAM was recently reported to reduce pain-associated behaviors in acetic acid induced abdominal writhing test. This effect was blocked by GABA	extsubscript{B} receptor antagonist CGP63360. ADX71943 also reduced pain in the formalin induced paw withdrawal in mice and rats. Allosteric modulators for GABA	extsubscript{A} receptors have also been developed and tested in several models of pain. While they were found to offer effective analgesic effect in rodent models of somatic pain, their efficacy in visceral pain models is yet to be established. HZ166, a new PAM agonist with preferential activity at GABA	extsubscript{A} receptors showed a dose-dependent anti-hyperalgesic effect in mouse models of neuropathic and inflammatory pain which was triggered by chronic constriction injury of the sciatic nerve and by subcutaneous injection of zymosan. At doses producing maximal antihyperalgesia, HZ166 was found to be devoid of sedation and motor impairment which is usually seen with muscimol and showed no loss of analgesic activity during chronic treatment period. Recently, Paul et al showed that the antihyperalgesic effect mediated by GABA	extsubscript{A} receptors occurs exclusively via a genuine spinal action and does not involve supraspinal sites. These findings further support the use of allosteric modulators rather than using traditional ligands as they are devoid of side effects.

**Glutamate Receptors**

The excitatory amino acid, glutamate, is a major neurotransmitter in the mammalian central nervous system and plays an important role in nociceptive processing, via a diverse set of membrane receptors which include both ionotropic and metabotropic. Activation of these receptors is responsible for basal excitatory synaptic transmission and many forms of synaptic plasticity such as long-term potentiation (LTP) and long-term depression, mechanisms that are thought to underlie learning and memory process.

**Ionotrophic Glutamate Receptors**

Glutamate acts on 3 ionotropic receptor subtypes: N-meth-
N-Methyl-D-aspartate receptors

NMDA receptors are calcium channels activated by glutamate and are slowly desensitized once activated. These high-calcium permeable channels generate synaptic neuroplasticity, wide dynamic range neuronal responses and gene expression within the CNS. NMDA receptors display a number of unique properties that distinguish them from other ligand-gated ion channels.

First, the receptor controls a cation channel that is highly permeable to monovalent ions and calcium. Second, simultaneous binding of glutamate and glycine, the co-agonist, is required for efficient activation of NMDA receptor. Third, at resting membrane potential the NMDA receptor channels are blocked by extracellular magnesium and open only on simultaneous depolarization and agonist binding. Native NMDA receptors are composed of NR1, NR2 (A, B, C, and D), and NR3 (A and B) subunits. Co-expression studies have demonstrated that formation of functional NMDA receptor channels requires a combination of NR1, an essential channel-forming subunit, and at least one of the NR2 subunits.

NMDA receptor activation has long been reported to be associated with visceral hyperalgesia. Due to slow inactivation, NMDA receptors produce LTP which is reported to generate chronic pain. NMDA receptors are found on enteric extrinsic afferents from colon and bladder; these afferents release calcitonin gene-related peptide and substance P once the receptor is activated. NMDA receptors within the rostral ventromedial medulla are important in modulating pain.

The study demonstrated that subpopulations of spinal neurons (short latency-sustained neurons) are sensitized as a result of neonatal somatic pain and the function of these neurons is predominantly influenced by NMDA receptors. In the CNS, hippocampal N-methyl D-aspartate receptor subtype 2B (NR2B)-NMDA receptors were found to be responsible for the facilitation of CA1 LTP via tyrosine phosphorylation, which leads to visceral hypersensitivity. In this study, the tyrosine kinase inhibitor (genistein) was found to significantly restrict the induction of LTP via inhibition of tyrosine phosphorylation of NR2B subunit, resulting in the inhibitory effect on LTP maintenance. A similar finding on the role of tyrosine phosphorylation of NR2B subunit was also reported to play a crucial role in central sensitization of chronic visceral pain. Zhuo suggested increased expression of NR2B subunit of NMDA receptor and glutamate receptor 2 (GluR2) of AMPA receptor might be involved in the key mechanisms for long-term synaptic plastic changes responsible for visceral hypersensitivity in rats. Further, activation of NMDA receptors has been shown to be critical to the manifestation of wind-up in spinal neurons recorded and plays a major role in amplification of a nociceptive input in post-inflammatory conditions. In clinical studies, Verne et al. reported on the involvement of NMDA receptor in the development of somatic hypersensitivity in IBS patients suggesting a role for these receptors in convergence-facilitation mechanism observed in a subset of IBS patients who display enhanced pain sensitivity to rectal and cutaneous heat stimulation.

Kainate receptors

Kainate receptors are ionotropic receptors that respond to the neurotransmitter glutamate. Despite the key role played by glutamate in transmission of nociception, the role of kainate receptors on nociceptive transmission of the viscera has not been investigated. While postsynaptic kainate receptors are involved in excitatory neurotransmission, presynaptic kainate receptors have been implicated in inhibitory neurotransmission by modulating release of the inhibitory neurotransmitter GABA through a presynaptic mechanism. There are 5 types of kainate receptor subunits, GluR5, GluR6, GluR7, KA1, and KA2 which are similar to AMPA and NMDA receptor subunits and can be arranged in different ways to form a tetramer. The ionotropic kainate receptor subunit, GluR5 (GluK1), is expressed in many regions of nervous system related to sensory transmission. Recent study has also shown that GLUR1 receptor in the spinal cord regulates neurotransmitter release from the GABAergic and glycinergic interneurons in the spinal cord. In naïve rats, intrathecal administration of GLUR5-selective antagonist, LY382884 was found to attenuate capsaicin- and carrageenan-induced mechanical allodynia in rats. This suggests that under normal conditions, there is a role of these receptors in spinal nociceptive transmission. Spinal administration of MSVIII-19 (a selective ligand for the GluK1 receptor) reverses hypersensitivity in several models of pain in mice, supporting the clinical potential of GluK1 antago-
nants for the management of pain. Similarly, kainate receptor subunit GluR6 was reported to play an important role in visceral pain. Administration of GluR6 antisense oligonucleotides (ODNs) can suppress the expression of GluR6 in rat spinal cord, and subsequently found to alleviate the formalin-induced pain. The results of this study suggest that kainate subunit GluR6 plays a pivotal role in the visceral pain of rectum induced by formalin injection, and it may serve as a potential target in designing new therapy for visceral inflammatory pain.

**AMPA receptors**

AMPA receptor is a non-NMDA-type ionotropic transmembrane receptor for glutamate that mediates fast synaptic transmission in the CNS. AMPA receptors are composed of various combinations of 4 subunits (GluR1- GluR4), and only AMPA receptors that lack the GluR2 subunit are permeable to Ca2+. AMPA receptors are reported to be involved in neuropathic pain conditions; however information on their role in visceral pain is limited. Studies performed by Chen et al. provide direct evidence that peripheral nerve injury induces postsynaptic GluA1 accumulation in cingulate cortical neurons and inhibits postsynaptic GluA1 accumulation. Banerjee et al. reported that esogephalic acid exposure in rats significantly increased expression of GluA1, pGluA1Ser831, and phosphorylated CaMKIIThr286, in the cortical membrane preparations. Electrophysiology studies showed that microinjection of IEM-1460 (open-channel blocker of AMPA receptors) near the recording site significantly attenuated acid-induced sensitization of cortical neurons suggesting a role for AMPA receptors in acid reflux induced esogephalic pain.

**Metabotropic Glutamate Receptor**

Metabotropic glutamate (mGlu) receptors are G-protein coupled receptors that have been subdivided into 3 groups, based on sequence similarity, pharmacology and intracellular signaling mechanisms. Group I mGlu receptors (mGlu1 and mGlu5) are coupled to PLC and intracellular calcium signaling, while group II (mGlu2 and mGlu3) and group III receptors (mGlu4, mGlu6, mGlu7, and mGlu8) are negatively coupled to adenyl cyclase. Lindström et al. first reported that the mGluR5 antagonists (MPEP and MTEP) inhibit colorectal distension-evoked visceromotor responses and cardiovascular changes in conscious rats suggesting that mGluR5 participates in mediating mechanically evoked visceral nociception in the gastrointestinal tract. Crock et al. demonstrated that activation of metabotropic glutamate receptor 5 (mGluR5) in the central nucleus of the amygdala (CeA) induces bladder pain sensitization by increasing CeA output. Optogenetic activation of the CeA neurons expressing mGluR5 receptor was found to produce a robust increase in the visceral pain response indicating the involvement of mGluR5 receptors in visceral pain. The CeA-localized effects on responses to bladder distention were associated with changes in extracellular signal-regulated kinases 1/2 (ERK1/2) phosphorylation in the spinal cord demonstrating that mGluR5 activation leads to increased CeA output that drives bladder pain sensitization. Liu et al. showed that Activation of mGluR7 in the nucleus tractus solitarii had anti-nociceptive effects, whereas activation of mGluR8 in the nucleus tractus solitarii had pro-nociceptive effects on cardiac-evoked muscle hyperalgesia and this facilitatory effect was found to be dependent on vagal afferents.

**Opioid Receptors**

Opioid receptors are a group of G protein-coupled receptors with opioids as ligands. They are activated both by endogenously produced opioid peptides and by exogenously administered opioid drugs, such as morphine, which are not only among the most effective analgesics known but also highly addictive drugs of abuse. To date, 4 types of opioid receptors have been identified: μ (mu for morphine), κ (kappa for ketocyclazocine), δ (delta for deferens given that it was originally discovered in the vas deferens of mice). The use of opioid analgesics has a long history, dating back several millennia. In spite of extensive research and wide spread use of opioids for the treatment of various forms of pain, controversies still linger. The mu, delta and kappa opioid receptors are all involved in nociception and are thus obvious candidates for a targeted drug development in the clinical treatment of both acute and chronic pain.

Kappa and mu opioid receptors are found on visceral afferents. Distribution along the gastrointestinal tract indicates that endogenous opiate peptides have a modulating function for both gastrointestinal motility and secretory function. Su et al. documented that responses of mechanosensitive pelvic nerve afferent fibers innervating the colon are inhibited by kappa-opioid receptor agonists having varying affinities for putative kappa-opioid receptor subtypes. Su et al. also reported that kappa, but not mu or delta opioid receptor agonists attenuated the responses of bladder sensitive pelvic nerve afferent fibers to urinary bladder distension suggesting a role for these receptors in modulating bladder pain. Sengupta et al. reported a peripheral upre-
gulation of kappa-opioid receptors in rats following in colonic inflammation produced by intracolonic instillation of trinitrobenzene sulfonic acid. Clinical studies with human volunteers found that oxycodone which is thought to be a kappa and mu receptor agonist significantly blocked visceral pain better than morphine which has little kappa receptor activity. Recently, Fichna et al. showed that the activation of the endogenous nociceptin/orphanin (N/OFQergic) system by using a new potent non-peptide nociception orphanin peptide agonist, SCH 221510, displays a significant antinociceptive effect in 2 mouse models of visceral hypersensitivity mimicking IBS pathology. Moreover, the same group has previously reported an antinociceptive effect of the same drug when orally administered in a mouse model of trinitrobenzene sulfonic acid-induced colitis. In vitro afferent recordings from mouse splanchic high-threshold nociceptors from mice with colonic inflammation we found that asimadoline (peripherally restricted selective KOR agonist) dose-dependently inhibited colonic nociceptors. Furthermore, asimadoline also dose dependently inhibited colonic nociceptors from chronic visceral hypersensitivity, an effect that was prevented by the prior application of a KOR antagonist. Overall, these data indicate KOR expression is functionally upregulated during inflammation and chronic visceral hypersensitivity.

Serotonin Receptors

Serotonin (5-HT) is an important neurotransmitter in the brain-gut interaction, with 80% of the total body 5-HT located in the gastrointestinal (GI) tract. Approximately 95% of the human body’s serotonin is produced and stored in enterochromaffin cells in the intestinal epithelium. However, small amounts of 5-HT are also present in serotonergic neurons of the enteric nervous system where 5-HT takes part in the slow and fast neurotransmission. Serotonin exerts its biological activity through interaction with different receptors, currently classified into 7 groups on the basis of their structure, transduction mechanism and pharmacological profile: 5-HT1-7. Most of these receptors are expressed in the GI tract, and their stimulation plays different roles (either inhibitory or excitatory) in the control of intestinal motility and secretion. The 5-HT3 receptor is coupled to an ion channel, whereas 5-HT 1,2,4,5,6,7 receptors are coupled to G proteins. Although the 5-HT receptors are extensively investigated, conflicting results exist regarding the role of these receptors in somatic and visceral nociceptive processing.

Mickle et al. reported that the 5-HT1A receptor agonist DPAT produces pronociceptive effects, primarily via the activation of presynaptic 5-HT1A receptors in GABAergic neuron to restrict GABA release and thereby disinhibiting the excitatory glutamatergic neurons in the spinal cord. The 5-HT1 receptor agonist tegaserod has also been reported to modulate visceral pain. Tegaserod produces analgesia via activation of supraspinal 5-HT1 receptors which triggers the release of opioids at supraspinal site, which in turn activates the descending noradrenergic pathways to the spinal cord to produce analgesia. Systemically and centrally administered alosetron (5-HT3 antagonist), reversed the mechanical somatic hypersensitivity and prevented the development of visceral hyperalgesia, suggesting a centrally mediated effect.

Cannabinoid Receptors

The cannabinoid (CB) receptors found in mammals are CB1 and CB2, both members of the superfamily of G protein-coupled receptors. CB1 receptors are found primarily in neurons of the brain and GI tract extrinsic and intrinsic nervous system. CB2 receptors have been identified through immunohistochemical studies in most neurons of the ileum enteric nervous system of mice and in peripheral immune cells. There is a variety of published data to show that cannabinoid receptor agonists have an anti-nociceptive effect in inflammatory and visceral hyperalgesia in animal models, however these observations are not convincingly supported by human studies.

Feng et al. reported on the role of endocannabinoids in 5-HT mediated modulation of visceral nociception in a rat model suggesting that vagal 5-HT1 receptor-mediated duodenal anandamide release contributes to acute luminal 5-HT-induced antinociception via CB1 signaling. In contrast, decreased anandamide is associated with hyperalgesia upon chronic 5-HT treatment. Similarly, taranabant, an inverse agonist of CB1 receptor was reported to reduce abdominal pain and increases intestinal transit in mice. While several animal studies have been reported on the analgesic effect of CB receptor agonists, some clinical studies argue against centrally acting CB agonists as tool to decrease visceral hypersensitivity. In a case controlled study involving ten IBS patients and 12 healthy volunteers, mixed CB1/CB2 receptor agonist delta-9-tetrahydrocannabinol (dronabinol) did not alter baseline rectal perception to distension compared to placebo in healthy or IBS patients. Similarly, after sigmoid stimulation there were no significant differences between placebo and delta-9-tetrahydrocannabinol in sensory thresholds of discomfort indicating non-involvement of these receptors in visceral sensation. These results necessitate the need to further
investigate the role of CB receptors in somatic and visceral pain conditions.

**Transient Receptor Potential Ion Channels**

The transient receptor potential (TRP) family of ion channels is TRPV1, TRPV2, TRPV3, TRPV4, TRPM 8, and TRPA1. These channels are, in general, thermoreceptors found on poorly myelinated and nonmyelinated afferents arising from the dorsal root ganglia, nodose ganglia, and the CNS. TRPA1. These channels are calcium permeable, nonselective channels with 4 identical subunits and a central pore. TRPA1 is highly expressed on primary afferents. TRPV1 has been demonstrated to be upregulated in upper gastrointestinal disorders such as gastroesophageal reflux disease and pancreatitis. TRPV1 expression was found to be upregulated not only in those with inflammatory bowel disease in remission but also with irritable bowel type symptoms. TRPA1 is upregulated in experimental colitis and deletion of TRPA1 expression in a mouse gene knock-out model reduced colitis-induced mechanical hypersensitivity. Charrua et al. also reported on the synergistic effect of TRPV1 and TRPV4 receptors although they are expressed in different bladder afferent populations. While low doses of TRPV4 antagonist (RN1734) and TRPV1 antagonist (SB366791) had no effect on bladder hyperactivity induced by lipopolysaccharide. The synergistic activity of antagonists for these receptors in very low doses may offer the opportunity to treat lower urinary tract symptoms while minimizing the potential side-effects of each drug. Jurik et al. reported that constitutive genetic deletion of TRPV1 or peripheral TRPV1 deletion reduced acetic acid-evoked abdominal constrictions, without affecting referred abdominal hyperalgesia or allodynia in an acute pancreatitis induced visceral pain model. Additionally, intracerebral TRPV1 antagonism by SB 366791 significantly reduced chemical and inflammatory spontaneous abdominal nociceptive responses, as observed by reduced expressions of nociceptive facial grimacing. In addition to the established role of cerebral TRPV1 in anxiety, fear, or emotional stress, this study demonstrates for the first time that TRPV1 in the brain modulates visceral nociception by interfering with the affective component of abdominal pain. The transient receptor potential canonical subfamily 4 (TRPC4) ion channel, which is involved in the tissue-specific and stimulus-dependent regulation of intracellular Ca²⁺ signaling also plays a role in visceral pain. Westlund et al. reported that rats with TRPC4-knockout mutation displayed tolerance to visceral pain induced by colonic mustard oil exposure, but not somatic or neuropathic pain stimuli. Behavioral studies showed that ML-204 (TRPC4 antagonist) inhibited visceral pain-related behavior in a dose-dependent manner without noticeable adverse effects. These data provide evidence that TRPC4 is required for detection and/or transmission of colonic mustard oil visceral pain sensation.

**Conclusion**

It is now widely accepted that chronic visceral pain is mediated by unique peripheral and central mechanisms. This article, based on a literature review, focused on the involvement of the major ionotropic and metabotropic receptors in the pathophysiology of chronic visceral pain. Considerable advances in research have provided an insight into many of the molecular mechanisms involved and animal studies using clinically relevant models of visceral pain along with in vitro studies are expanding the field of knowledge. In summary, various ionotropic and metabotropic are widely reported to be involved in visceral pain modulation and is likely to involve a number of interactive components and pathways. The involvement of these molecular components in visceral hypersensitivity in experimental models and patients provides a deeper insight and complexities involved and the challenges associated with the treatment of visceral pain. Many of the potential targets discussed here need to be translated into clinical practice with further good clinical trials.

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