Peripheral and central mechanisms of visceral sensitization in man

P. ANAND,* Q. AZIZ,† R. WILLERT† & L. VAN OUDENHOVE††

Abstract Visceral hypersensitivity (perception of gastrointestinal sensory events at a lower-than-normal threshold) is considered to be an important pathophysiological mechanism in the development of functional gastrointestinal disorders (FGIDs), such as irritable bowel syndrome, non-cardiac chest pain and functional dyspepsia. These disorders are associated with significant health care and socioeconomic costs due to factors such as repeated visits to consultants, hospitalizations and work absenteeism. Despite the presence of extensive evidence linking visceral hypersensitivity and FGIDs, the mechanism(s) underlying visceral hypersensitivity has not been fully elucidated. Suggested hypotheses include sensitization of afferent neurones, both at the level of the enteric and the (afferent) autonomic nervous system (peripheral sensitization), sensitization of spinal cord dorsal horn neurones (central sensitization) and psychosocial factors/psychiatric comorbidity influencing the processing of afferent signals at the level of the brain. Importantly, these hypotheses may be complementary rather than mutually exclusive. However, the degree to which each of these mechanisms contributes to the overall perception of visceral pain, and therefore the generation of symptoms, still remains unclear. This article discusses the mechanisms that may underlie visceral hypersensitivity, with reference to FGIDs. Understanding these mechanisms is essential in order to improve the diagnosis and treatment of patients with these disorders.

Keywords brain–gut axis, central and peripheral sensitization, functional gastrointestinal disorders, psychosocial factors, visceral hypersensitivity, visceral pain.

INTRODUCTION

Chronic unexplained symptoms are common to all medical specialties. They cause considerable morbidity and have enormous health care resource implications. Extensive investigations are conducted without benefit and inappropriate treatment leads to poor patient satisfaction. Understanding the pathophysiological mechanisms underlying these conditions has, therefore, become one of the major challenges for medicine in the 21st century.

Unexplained abdominal symptoms account for 40% of gastroenterological practice in the United Kingdom. These problems are usually classified as functional gastrointestinal disorders (FGIDs), a term that includes irritable bowel syndrome (IBS), non-cardiac chest pain (NCCP) and functional dyspepsia (FD). Despite extensive research in this area, the pathophysiology of FGID remains uncertain. The quest to identify an organic cause for symptoms leads to extensive investigation and frequent hospital attendance, which exerts a considerable financial strain on health service resources.1 A socioeconomic study estimated that the combined cost of health care utilization and job absenteeism related to FGID is $41 billion per annum in the eight leading Western economies.2

Heightened perception of gastrointestinal (GI) sensation [visceral hypersensitivity] is commonly observed in patients with unexplained abdominal pain.3 Studies using mechanical and electrical stimulation have
reproducibly demonstrated that patients with FGID have lower GI pain thresholds in comparison to healthy subjects. Furthermore, generalized somatic pain hypersensitivity is not apparent in FGID as studies have demonstrated that cutaneous pain thresholds in IBS and NCCP patients are similar or even higher than in controls, although lower cutaneous pain thresholds were also found in some studies.

Despite extensive research to understand mechanisms of visceral hypersensitivity in FGID, the dichotomy between a purely psychological explanation and a purely organic explanation remains unaddressed. Evidence for a purely psychological explanation comes from studies in FGID patients who have shown a high incidence of psychological/psychiatric problems, not only in tertiary care samples but possibly also in the community. Furthermore, experimental evidence from animal and human studies suggests that acute stress and anxiety alter GI function, and animal studies show that stress such as maternal separation in childhood can lead to visceral hypersensitivity in adult life. It is notable, however, that while psychological therapy improves well-being, it does not improve symptoms, suggesting that psychological problems may be more relevant to health-care-seeking behaviour rather than underlying the disease pathogenesis.

Evidence for a purely organic cause is provided by animal and human studies which clearly demonstrate that inflammation of the GI tract leads to visceral hypersensitivity due to increased sensitivity of afferent pathways. Furthermore, a third of FGID patients give a prior history of gut inflammation or injury such as gastroenteritis or abdominal surgery. However, patients who go on to develop FGID after gastroenteritis have a higher incidence of antecedent psychological problems, suggesting an interaction between psychological and organic aetiologies.

Functional dyspepsia, for example, has been consistently reported to be associated with abnormalities in gastric sensorimotor function (reviewed in Ref.25), including hypersensitivity to gastric distension, abnormal gastric compliance, impaired gastric accommodation to a meal and delayed gastric emptying. Moreover, psychosocial stressors including history of sexual or psychological abuse, personality traits (e.g. neuroticism) and psychiatric disorders (e.g. mood and anxiety disorders) as well as somatization are frequently present in FGID patients, in tertiary care samples and in the community. However, it is still unclear how psychosocial factors may interact with GI sensorimotor function in general and with visceral sensitivity in particular, in both health and FGID.

To explain the association between FGID, GI sensorimotor function and psychosocial factors, several potentially overlapping hypotheses have been put forward. Firstly, a direct [neuro]biological and possibly reciprocal interaction may exist between psychosocial factors or psychiatric disorders and GI sensorimotor function, the biological substrate for this being the reciprocal connection between the brain and the gut [brain–gut axis (BGA)]. This hypothesis implicates that psychosocial factors play a key role in FGID pathophysiology. Secondly, psychosocial factors or psychiatric disorders may influence gastric sensitivity through 'psychological' processes [arousal, anticipation/expectation, attention to/interpretation of bodily feelings and visceral-specific anxiety] that may influence symptom perception and/or symptom reporting. It should be noted, however, that it is well known from psychological, cognitive and pain neuroscience literature that these psychological processes have neurobiological correlates in the brain. Thirdly, psychosocial factors may not have a direct influence on GI sensorimotor function, but may only influence health care seeking or quality of life in FGID. Finally, FGIDs or visceral hypersensitivity and psychiatric or psychological disorders may be manifestations of a common [genetic] predisposition (e.g. serotonin transporter polymorphisms), this may lead to theories stating that they are nothing more than epiphenomena.

Based on current scientific evidence, a number of hypotheses have been proposed to explain the mechanism of visceral hypersensitivity. These include (i) sensitization of GI afferent nerves [peripheral sensitization (PS)], (ii) sensitization of spinal cord dorsal horn neurones [central sensitization (CS)], (iii) altered descending excitatory or inhibitory influences to the spinal cord nociceptive neurones [which may be influenced by psychological processes] and (iv) misinterpretation of non-noxious sensation as noxious due to cognitive and emotional biasing [hypervigilance], the result of psychiatric/psychological disorders.

The following sections summarize the role of PS/CS, altered descending influences on the spinal cord, psychological factors including hypervigilance and the putative interactions between these mechanisms.

**PERIPHERAL SENSITIZATION**

Tissue damage due to inflammation leads to sensitization of primary afferent nerves (PS) due to the release of inflammatory mediators, such as K+, H+, adenosine triphosphate (ATP), bradykinin, prostaglandins, serotonin and histamine. These inflammatory mediators reduce the transduction threshold of primary
afferents and recruit previously silent nociceptors.\textsuperscript{49} Inflammation also induces increased expression of sodium channels Nav 1.8 and 1.9 (SNS1 and 2), the vanilloid receptor transient receptor potential vanilloid type 1 (TRPV1 or VR1), the purine receptor P2X\textsubscript{3} and acid-sensing ion channels [ASICs].\textsuperscript{50–53} Furthermore, cytokines secreted by macrophages and mast cells contribute indirectly to nerve sensitization by upregulating the expression of nerve growth factor [NGF]\textsuperscript{54} and cause the release of cyclo-oxygenase metabolites\textsuperscript{55,56} and sympathomimetic amines. The consequence of these changes is an increase in pain sensitivity at the site of inflammation.\textsuperscript{49}

Immunocytochemical techniques have recently been used to demonstrate an upregulation of peptides, cytokines, TRPV1 receptor and neurotrophic factors in skin, urinary bladder and rectal biopsies of patients with chronic hypersensitivity states without evidence of overt inflammation.\textsuperscript{57} Uregulation of TRPV1 has also recently been demonstrated in patients with idiopathic vulvodynia,\textsuperscript{58} which further suggests that the TRPV1 receptor may be an important marker of afferent nerve sensitization. An upregulation of TRPV1, P2X\textsubscript{3} receptors and ASICs has also been identified in the inflamed human GI tract.\textsuperscript{59,61,63} Recent studies have demonstrated that patients with oesophagitis have an upregulation of both TRPV1\textsuperscript{59} and cytokine interleukin-8.\textsuperscript{60} These studies show that it is now possible to explore what receptors play an important role in mediating visceral hypersensitivity.

CENTRAL SENSITIZATION

Enhanced nociceptor input activates intracellular signalling cascades within spinal dorsal horn neurones. This leads to amplified responses to both noxious and innocuous inputs, due to facilitated excitatory synaptic responses and depressed inhibition.\textsuperscript{61–63} Facilitation is triggered by the presynaptic release of neurotransmitters and neuromodulators, such as glutamate, substance P [SP], brain-derived neurotrophic factor [BDNF] and prostaglandins. These neurotransmitters and neuromodulators activate ligand-gated ion channels including N-methyl-D-aspartate [NMDA] and a-amino-5-hydroxy-3-methyl-4-isoxazole propionic acid [AMPA] receptors, G-protein-coupled metabotropic receptors, neurokinin receptors and tyrosine kinase receptors, which then increase intracellular calcium via release from intracellular stores and calcium inflow. Subsequently, alterations in ion channel and receptor activity via calcium-dependent activation of protein kinase A [PKA], protein kinase C [PKC] and tyrosine kinases lead to phosphorylation of the NMDA receptors.\textsuperscript{49} This dramatically changes NMDA receptor kinetics and reduces its voltage-dependent magnesium block, thus augmenting its subsequent responsiveness to glutamate and increasing synaptic strength, enabling previously subthreshold inputs to activate the cell.\textsuperscript{54,65} This increase in gain alters receptive field properties and pain sensitivity, causing tissue hypersensitivity far beyond the site of injury that initiated CS.

In addition to producing CS, which occurs within seconds of appropriate activation of spinal dorsal horn neurones, nociceptive input also generates an activity-dependent change in transcription in dorsal root ganglion and dorsal horn neurones.\textsuperscript{66} These transcriptional changes occur in response to a complex mechanism involving the activation of transcription factors that lead to both an increase and a modification of constitutively expressed genes and also induction of novel genes. For instance, non-nociceptive afferents begin to express SP and BDNF after inflammation\textsuperscript{67} and this phenotypic shift results in allodynia, i.e. non-nociceptive tactile stimuli now induce pain. These changes take hours to manifest but, when established, lead to long-lasting changes in normal stimulus response characteristics.

A striking feature of the increase in synaptic efficacy characteristic of CS is that it includes not only those nociceptor central terminal synapses activated by the conditioning stimulus but also synapses made by low-threshold mechanosensitive A\β fibres on dorsal horn neurones.\textsuperscript{68,69} Low-threshold sensory fibres, activated by innocuous stimuli such as light touch, can now activate normally high-threshold nociceptive neurones at the dorsal horn, contributing to a reduction in pain threshold such that non-painful stimuli are now perceived as pain (allodynia), which is a direct consequence of an increased excitability of central nervous system [CNS] neurones. Although this pain is referred to the periphery, it arises from within the CNS. This central facilitation manifests within seconds of an appropriate nociceptive conditioning stimulus and can outlast the stimulus for several hours.\textsuperscript{70} If the stimulus is maintained, even at low levels, the CS persists. After peripheral nerve injury, for example, ongoing ectopic activity arising from sensory fibres in the injured nerve can elicit prolonged CS.\textsuperscript{71}

Such activity-dependent CS is extremely robust and has been reported in rodent, cat and primate dorsal horn neurones, including spinthalamic neurones.\textsuperscript{72–74} The behavioural consequences of CS can be readily detected in human psychophysical experiments. Intradermal injection of capsaicin, the pungent ingredient in chilli peppers, which activates the TRPV1 receptor, produces an intense but transient pain owing to activation of TRPV1-expressing nociceptors. This is
followed by heightened sensitivity to pinprick outside the region of the capsaicin injection (secondary mechanical hyperalgesia) and to low-threshold mechanosensory (brush) inputs [secondary mechanical allodynia], due to the induction of CS. Clinically, CS has been demonstrated to contribute to pain hypersensitivity in the skin, muscle, joints and viscera.

Central sensitization also plays a major role in the generation of acute postoperative and post-traumatic pain, migrainous and neuropathic pain. Interestingly, some clinical conditions, such as tension-type headache and fibromyalgia, appear not to be a reaction to a peripheral pathology but instead an expression of the presence of CS, but why CS manifests apparently spontaneously in these patients remains to be established.

ASCENDING AND DESCENDING VISCERAL PAIN PATHWAYS: THE BRAIN–GUT AXIS

Knowledge of the bidirectional communication system between the gut [enteric nervous system (ENS)] and the brain (CNS), classically termed the BGA is critical for understanding a putative influence of psychosocial factors on GI sensitivity and motor functions. The ENS and the CNS communicate through neural (autonomic nervous system), neuroendocrine (hypothalamo-pituitary-adrenal axis) and neuroimmune pathways, and these systems may highly interact, for example, through cytokine receptors on the vagus nerve. An extensive review falls beyond the scope of this article (see Refs), but we will provide a brief overview of the most important structures and their functions, with emphasis on the neural pathways involved in GI sensitivity.

Ascending pathways

Gastrointestinal sensory information is transmitted to the brain through vagal and spinal afferent nerves. Vagal afferents project to the nucleus of the solitary tract, which in turn projects to the thalamus (mostly via the parabrachial nucleus) and directly to regions regulating arousal and emotional, autonomic and behavioural responses including the hypothalamus, locus coeruleus (LC), amygdala and periaqueductal grey (PAG). From the thalamus, GI sensory signals are relayed to the cortical components of the ‘visceral sensory neuromatrix’ [see below].

First-order spinal afferent nerves make synapse in the dorsal horn of the spinal cord and second-order neurones project to the brain through the spinoreticular, spinomesencephalic, spinohypothalamic and spinothalamic tracts. The first three of these tracts mainly activate fast, largely unconscious and/or automatic responses to visceral sensory input [arousal, orientation, autonomic responses, prototype emotional and behavioural responses], thereby playing a key role in maintaining the homeostasis of the organism.

The spinothalamic tract projects to the ventral posterior lateral, medial dorsal and ventral medial posterior nuclei of the sensory thalamus, from which information is relayed to the somatosensory cortices (S1/SII) [lateral pain system], the anterior cingulate cortex [ACC] [medial pain system] and the insula, respectively. In these cortical regions, conscious and more complex processing takes place. The main function of SI/SII is to provide information about intensity and localization of the stimulus [sensory-discriminative pain dimension], whereas the ACC mainly processes pain affect [affective-motivational pain dimension]. The different subregions within the ACC are also important in generating autonomic, behavioural and descending antinociceptive responses to visceral pain, and in anticipation of or attention to aversive (visceral) stimuli. The insula is the ‘interoceptive cortex’ where all information about the internal state of the organism is processed, playing an important role in integrating visceral sensory and emotional information and in higher order control of autonomic visceromotor responses. Finally, the orbital prefrontal cortex is playing a key role in the integration of sensory information from different modalities (especially related to food and eating) and attributing affective, motivational, reward and hedonic valence to it. Furthermore, this region is also involved in the generation of and choice between autonomic and behavioural response patterns, and has been shown to be a putative biological substrate of cognitive influences [including placebo effect and expectation of relief] on emotions and the affective dimension of visceral pain.

Thus, different dimensions of visceral sensation and pain are processed at the different levels of the ascending part of the BGA as described. However, descending pathways originate at virtually all BGA levels to modulate the ongoing transmission of visceral sensory information, mainly at the level of the dorsal horn of the spinal cord.
ascending pathways, can be enhanced by stimulation of the reticular formation of the nucleus raphe magnus. This is part of the excitatory spino-bulbo-spinal feedback loop, which is conveyed within the ventrolateral funiculus to excite spinal cord neurones. The role of this excitatory pathway is thought to be to activate the descending antinociceptive system to the dorsal horn via the nucleus raphe magnus and to activate arousal and emotional responses via autonomic nuclei.

Central descending inhibitory pathways

Viscerosomatic spinal neurones can not only be excited by visceral afferent activity but also be inhibited. The inhibitory neurones are thought to have a modulatory role in visceral pain. For example, visceral stimulation can induce excitation of innervating neurones and inhibit non-innervating spinal neurones so that the ascending information within the cord is enhanced from this organ, hence making it easier to interpret the source of afferent information for the brain. Other viscerosomatic neurones that innervate the viscera can be inhibited by either a visceral or somatic input for up to 1 s, so that no afferent response to a further input during this period occurs. While their role is not fully understood, the activity of these viscerosomatic afferents may explain the intermittent rhythmic nature of abdominal colic. These neurones have also been implicated in explaining the phenomena called counterirritation, where the pain threshold in the viscera is increased following noxious somatic stimulation within its segmental spinal innervation.

Besides local spinal inhibitory pathways, it is well recognized that spinal noceceptive transmission is modulated by descending pathways from various supraspinal structures, including the nucleus raphe magnus, periventricular grey of the hypothalamus and the midbrain PAG. At cortical level, the ACC is the most important source of descending modulatory pathways, projecting to the amygdala and the PAG, which is probably the key pain modulatory region. On a lower brainstem level, the noradrenergic LC, the serotonergic raphe nuclei and the rostrolateral ventral medulla (RVM) receive input from the amygdala and the PAG, and project in turn to the dorsal horn of the spinal cord, where ongoing transmission of sensory information is modulated (gate mechanism). Throughout the whole descending modulatory system, from cortex (ACC) to PAG and spinal cord (dorsal horn), endogenous opioids are crucially involved, together with other neurotransmitters including serotonin and noradrenaline.

Thus, similar to its somatic counterpart, visceral nociceptive transmission is also subject to descending inhibitory modulation. This is evident in the responses of dorsal horn neurones to noxious colorectal distension, which were inhibited by electrical or chemical stimulation applied within the PAG. More recently, the visceromotor response (contraction of abdominal and hind limb musculature) and the spinal dorsal horn neuronal responses to colorectal distension have been shown to be modulated in a biphasic manner by chemical stimulation in the brainstem RVM. The interaction between the descending facilitatory and inhibitory systems from the RVM appears to produce a net facilitatory effect following tissue injury, perhaps as an evolutionary defence mechanism to enable protection of the injury. The neuromodulators producing these effects are only now beginning to be understood, but it appears that activation of NMDA receptors and production of nitric oxide are important in the descending facilitatory pathway, while non-NMDA receptors mediate the inhibitory descending pathways.

MECHANISMS OF VISCERAL PAIN HYPERSENSITIVITY

Although visceral pain hypersensitivity has been widely demonstrated in FGID, the pathophysiological mechanisms to account for such hypersensitivity are not well characterized. Alterations in the pain transmission pathways may occur throughout the BGA from the primary afferent, through the spinal cord to the brainstem and higher centres. Subsequent neural pathways from the brain to the gut via vagal and spinal efferents will modulate this sensory input, resulting in either a facilitatory or inhibitory response to the visceral stimulus.

Peripheral mechanisms

The development of PS of visceral afferent fibres has been shown to cause long-term sensorimotor disturbances of the gut when the inflammation subsides and neonatal visceral inflammation has been shown to cause long-term colonic hyperalgesia in adult rats. These results suggest that either persistent sensitization of primary afferent neurones or synaptic plasticity within the CNS can occur long after the resolution of the insult. It is therefore conceivable that a disorder now labelled as functional had an antecedent peripheral initiating event. Evidence for this hypothesis is seen in patients with postinfectious IBS (PI-IBS) who give a preceding history of GI infection prior to the
onset of their symptoms. Increased mast cells, T lymphocytes and expression of interleukin-1β have been observed in the large bowel in PI-IBS patients.20,112 Furthermore, recent data have shown close proximity of mast cells and nerves, with a correlation to abdominal pain severity in IBS patients.113 This suggests a neuroimmune interaction in IBS and is supported by demonstrations that SP can alter mast cell excitability and function via neurokinin-1 (NK-1) receptors on mast cells,114 with NK-1 receptor expression being influenced by interleukin-4 production from T lymphocytes.115 Therefore, changes in peripheral neuroimmune interactions may contribute to the pathophysiology and clinical expression of altered visceral pain hypersensitivity seen in FGID.116

Another potential mechanism for ongoing PS is nerve injury, as this is well known to cause long-lasting hyperalgesia in animal models of somatic neuropathic pain. Studies using a model of pelvic nerve damage in the rat have shown a reduced threshold to distension and increased spontaneous activity,117 suggesting that visceral nerve damage could significantly contribute to the afferent barrage arriving at the spinal cord in the absence of any peripheral inflammation. This could explain the anecdotal reports of patients with FGIDs pinpointing the onset of their symptoms to immediately after abdominal or pelvic surgery.

Peripheral inflammation could also potentially alter the phenotype of visceral afferent neurons such that an increased expression of ligand- or voltage-gated channels remains despite resolution of the inflammation. Candidate receptors include TRPV1, voltage-gated calcium or sodium channels and stretch activated potassium channels.118

TRPV1 receptors The TRPV1 receptor, which is activated by heat and capsaicin, plays an important role in visceral hypersensitivity. First cloned in 1997,119 the polymodal TRPV1 receptor belongs to the family of TRP receptors expressed particularly by small-sized afferent neurones and by mononuclear blood cells.120 TRPV1 is activated by capsaicin and its analogues, lipids, other molecules such as resiniferatoxin that contain a vanillyl moiety, and also by endocannabinoids including anandamide.119,121 Upon activation, a sensation of burning pain is evoked, along with release of the neuropeptides SP and calcitonin gene-related peptide. The receptor is also gated by noxious heat (>43 °C), and its mechanism potentiated by protons. It has been suggested that inflammatory and ischaemic hyperalgesia may in part be mediated by the enhanced TRPV1 response resulting from a decreased tissue pH and production of excess hydrogen ions.119

In humans, there is increasing evidence that TRPV1 is involved in gut hypersensitivity and pain. Topical capsaicin has been shown to be an effective treatment for idiopathic pruritus ani, with the probable mechanism being functional desensitization of nociceptors by capsaicin.122 Hypersensitivity is likely to result from inflammatory products driving phenotypical changes in sensory neurones expressing TRPV1, mainly via increased NGF and/or glial-derived neurotrophic factor [GDNF].

Transient receptor potential vanilloid type 1 receptor expression changes have also been linked with other gut hypersensitivity disorders. Patients suffering from rectal hypersensitivity and faecal urgency have been found to have an increase in the TRPV1-expressing nerve fibres when compared with controls, and these levels correlated with a decrease in threshold to rectal heat and distension.57 This group of patients was also found to have increased GDNF and trk-A-expressing fibres [Fig. 1, from Ref.123].

TRPV1 has been implicated in the mechanism of pain produced in gastro-oesophageal reflux disease [GORD]. In oesophagitis patients, the proportion of papillae positive for these nerve fibres was increased, suggesting that acid-induced inflammation may upregulate expression of acid-sensitive receptors such as TRPV1, hence contributing to the visceral hypersensitivity often seen in patients with GORD and chest pain.59 A recent study124 has revealed an increase in TRPV1-expressing nerve fibres in the oesophageal mucosa of patients with non-erosive reflux disease, further strengthening the hypothesis.

This could provide an explanation for the distressing burning symptom which these patients complain of during reflux episodes and in response to alcohol, hot beverages and foods, via stimulation of TRPV1. Interestingly, a trial of FD patients treated with red pepper125 resulted in patients initially complaining of epigastric pain, followed by an improvement of symptoms after prolonged treatment for a few days. This is similar to the effect seen with capsaicin treatment of pruritus ani described above, suggesting initial stimulation of TRPV1-expressing neurones, followed by desensitization.

Capsaicin induces ileal pain when applied via ileal stomata.126 Schmidt et al. conducted a study which revealed that perfusion of capsaicin in the human jejunum in healthy individuals induced pain and warmth sensation indicative of activation of capsaicin-sensitive receptors, probably TRPV1.127 In patients with painful inflammatory bowel disease [IBD], the number of TRPV1-expressing neurones is significantly increased in colonic mucosa.51
These studies in humans provide evidence of a role for TRPV1 in inflammation-induced pain and visceral hypersensitivity. The changes in expression are likely to be mediated by the effects of NGF, which is produced locally during inflammation. Nerve growth factor sensitizes TRPV1 receptors to protons, enhancing their effect, and also increases expression of TRPV1. Increased NGF and recently trk A expression have been reported in acute IBD. Ji et al. have shown that the increase in TRPV1 levels which occur 12–24 h after inflammation is by an NGF-mediated p38 kinase pathway. TRPV1 activity is modulated by inflammatory mediators including bradykinin and prostaglandins, probably by cAMP-dependent PKA or PKC-mediated phosphorylation of the receptor.

Possible mechanisms by which NGF can mediate hypersensitivity are summarized in Figs 2 and 3, reproduced with kind permission of The Lancet.

Acid-sensing ion channels Tissue damage, whether it be a result of trauma, infection, inflammation or ischaemia, results in local tissue acidosis and pain. Pain may be due to modulation of receptors, such as TRPV1, by acidic pH or by direct activation. A sodium selective channel, ASIC1, expressed by sensory neurones, is closed at a pH of 7.4, but is activated once the pH falls below 7.0. The related ASICs have been renamed as ASIC2a, ASIC2b and ASIC3.

These channels are likely to play a role in nociception and GI visceral hypersensitivity, but experimental evidence in humans is lacking at this stage. Yiangou et al. looked at ASIC expression in biopsies from actively inflamed Crohn’s disease patients and found that ASICs 1, 2 and 3 were all expressed in the enteric neurones. Interestingly, only ASIC3 expression was significantly upregulated in the inflamed specimens when compared with controls, suggesting a role for ASIC3 in inflammation and pain/GI hypersensitivity. As acid-sensing channels, they would be a likely candidate for oesophageal pain provoked by acid, but further studies are warranted.

ATP-gated ion channels Ion channels that are gated by extracellular ATP have been characterized on sensory neurones including those in the intestine in animal studies. Two types of receptors exist: P2X receptors are ATP-gated and P2Y are G-protein-coupled receptors. In the GI tract, ATP release may occur from a variety of sources including cell damage, sympathetic and extrinsic sensory neurones, and hence ATP-gated ion channels are a likely candidate for mediating GI nociception following inflammation, infection or injury.

P2X3 receptors, a subgroup of the P2X receptors, have been shown to be present in human enteric neurones. Yangou et al. also found that in inflamed IBD colonic biopsies, the levels of P2X3-expressing neurones were significantly increased. This human study implies that P2X3 have a role in inflammation, pain and dysmotility.

Voltage-gated sodium channels Voltage-gated sodium channels [VGSC], of which there are numerous in the central and peripheral nervous systems, are...
responsible for the rising phase of the action potential, by a voltage-dependent increase in sodium ion permeability. They are involved, along with potassium channels, in determining the excitability of sensory neurones. Classification into two types is possible: those sensitive to the potent puffer fish toxin tetrodotoxin (tetrodotoxin-sensitive) and those in the second group which are insensitive to tetrodotoxin (tetrodotoxin-resistant [TTXr], Nav 1.8 and 1.9). Tetrodotoxin-sensitive channels are found in all sensory neurones, but TTXr channels are preferentially expressed by nociceptor sensory afferents. Tetrodotoxin-resistant sodium channels are likely to play an important role in nociceptive transmission and there is particular interest in the TTXr VSGC α subunit SNS Nav 1.8. With respect to TTXr sodium channels, in vitro work has shown that a number of inflammatory mediators such as prostaglandin E2 (PGE2), serotonin and adenosine increase the rates of activation and inactivation, decrease the activation threshold and increase the size of the current, i.e. cause sensitization. Animal studies reveal that TTXr sodium channels play an established role in sensitization of afferents and development of inflammatory hyperalgesia, and although experimental data in human studies is lacking, they are likely to be a key player.
Mechanisms by which primary visceral afferent neurones contribute to visceral pain hypersensitivity may therefore include (i) peripheral inflammation, defined by ongoing cytokine expression in the absence of histological changes, (ii) visceral nerve damage and (iii) changes in the number or function of several ion channels, initiated and maintained by presently unknown means. All of these potential mechanisms could result in visceral pain hypersensitivity without further amplification of visceral afferent input to the CNS. However, it is more likely that the peripheral input adds to the CNS mechanisms, which also contribute significantly to visceral pain hypersensitivity.

Central mechanisms

Central sensitization Central sensitization is a key process in the development of persistent somatic pain hypersensitivity and there is an extensive body of literature that has highlighted the importance of SP, neurokinin B, PGE2 and the NMDA receptor in its development and maintenance at the spinal level.\textsuperscript{63} Evidence for a role of CS as a mechanism for the development and maintenance of visceral pain hypersensitivity comes from both animal and human studies.\textsuperscript{3,4,135–141}

Animal studies have demonstrated that following somatic inflammation, a positive correlation exists between visceral pain thresholds and increased afferent discharge of dorsal horn neurones demonstrating visceral somatic convergence.\textsuperscript{142–144} Spinal cFOS expression, used as a marker of dorsal horn activity, has also been shown to be increased following noxious colorectal distension\textsuperscript{145} and this is prevented by NMDA receptor antagonism.\textsuperscript{65,146}

To address the question whether inflammation/injury can induce CS in the human GI tract, a human model was developed, which demonstrated that infusion of hydrochloric acid into the healthy oesophagus reduced pain threshold not only in the acid-exposed region (PS) but also in the adjacent unexposed region (CSI). This effect was prolonged, lasting up to 5 h after 30 min of acid exposure. The duration and magnitude of CS was related to the intensity of acid exposure. Repeat exposure after recovery significantly potentiates the effect of the first infusion, suggesting that repeated injury can induce a progressive increase in hypersensitivity.

A major limitation of most visceral hypersensitivity studies is that they rely on subjective methods of reporting sensation.\textsuperscript{184} To overcome this, a commonly used neurophysiological technique, cortical evoked potentials (CEP), has been developed as a more objective correlate of oesophageal sensation. Cortical evoked potentials allows recording of cortical neuronal electrical fields generated in response to a peripheral nerve stimulus. Using this technique before and after acid infusion, a reduction in CEP latency was demonstrated, which suggests that facilitation of afferent pathway conduction accompanies the CS.\textsuperscript{140}

To explore whether the mechanisms of CS [in an oesophageal model] are similar to those described in animal studies, pharmacological studies have been used to block receptors involved in CS.

A recent study showed that administration of an antagonist at the PGE2-receptor EP1 prior to acid infusion blocks the subsequent development of oesophageal hypersensitivity, suggesting that prostaglandins play an important role in mediating PS and CS.\textsuperscript{185}

That inflammation-evoked prostaglandin release through induction of cyclo-oxygenase-2 (COX-2) at the site of injury/inflammation is an important mechanism for the development of PS has long since been established.\textsuperscript{55,56} Prostaglandins also play a role in spinal mechanisms of hyperalgesia (CS).\textsuperscript{186–190,192} Using a selective oral COX-2 inhibitor [Valdecoxib; Bextra\textsuperscript{a}, Pfizer Pharmaceuticals, no longer available on the UK and US market] in the oesophageal model of CS model in healthy subjects,\textsuperscript{191} it was demonstrated that the development and maintenance of acid-induced oesophageal hypersensitivity could not be attenuated by COX-2 inhibition.

Substance P and its receptor [NK-1] have been shown to have a role in pain and hyperalgesia,\textsuperscript{193–198} although results using NK-1 receptor antagonists [NK-1RA] in human somatic pain have been disappointing.\textsuperscript{199} Evidence for a role of SP in visceral nociception comes from several animal models, including NK-1 knockout mice, which have shown an effect of NK-1RA on reducing visceral hyperalgesia.\textsuperscript{200,201} In comparison to cutaneous afferents, a greater proportion of visceral afferent neurones contain SP [80% vs 25%]\textsuperscript{202} and the highest concentration of NK-1 receptors in lamina I of the spinal cord correlates with termination of visceral afferent neurones.\textsuperscript{203} Therefore, an oral selective NK-1 receptor antagonist was used in the oesophageal model of CS in healthy subjects to assess the role of SP in human visceral hypersensitivity.\textsuperscript{204} This demonstrated...
that the hypersensitivity induced in the proximal oesophagus [secondary allodynia] by acid infusion in the distal oesophagus was not prevented by prior treatment with the NK-1 receptor antagonist. Furthermore, the NK-1RA did not alter baseline sensory or pain thresholds in either the oesophagus or foot (somatic control), demonstrating that NK-1 receptor antagonism does not influence normal human nociceptive processing in keeping with previous published studies. Whether combination therapy with NK1 plus NK2 or NK3 receptor antagonists is efficacious remains to be seen.

Psychological factors: hypervigilance, anxiety, stress and abuse history Hypervigilance is a normal physiological state of the nervous system in response to perceived threat and enhanced arousal can be associated with enhanced sensitivity to visceral sensations, as seen in healthy individuals with sensations of palpitations, urgency and ‘butterflies’ in the stomach when experiencing fear. Evidence (from neuroimaging studies, among others) now suggests that some patients with FGIDs are chronically hypervigilant to physiological visceral stimuli in that they selectively attend to normally subthreshold visceral inputs147 [see below for overview of neuroimaging evidence].

Patients with IBS often present during times of increased personal stress [although it is unclear whether stress initiates the disorder],11,12 and evidence for a role of stress in visceral pain hypersensitivity comes from animal models of IBS where inducing stress using maternal separation,16 water avoidance148 or foot shocks14 causes the animals to develop visceral pain hypersensitivity to colonic distension during further periods of stress. Corticotropin-releasing factor [CRF] is implicated in stress-induced visceral pain hypersensitivity as it is released by the hypothalamus during increased limbic activity,13 activates the hypothalamic-pituitary-adrenal axis and results in increased cortisol production which may then have a role in facilitating intestinal sensitivity and in increasing general arousal.149,150 A recent study has given clinical support for the role of CRF in visceral hypersensitivity as a CRF antagonist improved motility while reducing visceral perception and anxiety in IBS patients;151 further studies are awaited.

What causes some to become hypervigilant or hyperresponsive to stress while others do not remains unclear, but as with the animal models above, a previous history of childhood adversity or significant life stressor appears to modulate your responses to stress and is implicated in your future risk of developing an FGID.152

In a recent study in a large sample of tertiary care FD patients, factor analysis was performed on dyspepsia symptoms to define patient subgroups; associations of symptoms with gastric pathophysiological mechanisms and psychosocial factors/psychiatric comorbidity were determined. An association between gastric hypersensitivity, epigastric pain and burning and neuroticism, somatization, history of psychological abuse as a child or adult and quality of life was found.29

Another study has recently shown that normosensitive and hypersensitive FD patients do not differ in state anxiety at the day of their barostat investigation, nor in trait anxiety. However, within the hypersensitive subgroup, a significant negative correlation was found between state anxiety on one hand and gastric discomfort thresholds, pain thresholds and compliance on the other. These findings indicate that state anxiety may influence gastric sensorimotor function within the hypersensitive subgroup of patients, rather than distinguish between hyper- and normosensitive patients.153

Moreover, a history of sexual abuse, especially in childhood, has been shown to be associated with lower gastric discomfort threshold in FD, whereas psychological and physical abuse are associated with altered thresholds for first perception.154 In a sample of female IBS patients, however, a history of severe sexual or physical abuse was found to be associated with higher rectal pain thresholds.155 More research on this methodologically difficult issue is needed and the relationship between different forms of abuse history and pain thresholds is complex,156 but evidence that especially early abuse experiences can alter visceral and somatic pain sensitivity is growing.

It has also been shown that help-seeking IBS patients with comorbid psychiatric disorders are more likely to develop psychiatric disorders [especially anxiety disorders] before the onset of IBS.157 This may suggest that psychiatric symptoms, especially anxiety, play a role in the development of IBS.157

Finally, the evidence for a beneficial effect of psychotherapy and hypnosis in FGIDs is increasing158–160 and these point towards central mechanisms playing an important role in visceral pain hypersensitivity as they are probably acting on the limbic system, to reduce the effects of stress on the BGA.

Endogenous pain modulation Descending CNS pathways from the RVM to the dorsal horn of the spinal cord are well described in somatic nociception161 where they have a tonic inhibitory effect.162 In contrast, spinal visceral nociceptive transmission has both descending facilitatory and inhibitory inputs that produce a net facilitatory effect.106 Alterations in this.
dynamic equilibrium between facilitatory and inhibitory inputs from the midbrain to the spinal dorsal horn neurones following central stress or peripheral inflammation could therefore result in enhanced visceral pain perception, due to enhanced descending facilitatory influences or reduced inhibitory inputs.\textsuperscript{163}

To date, only indirect evidence from brain imaging studies exists in patients with FGIDs to assess the contributions of these pathways to symptom generation [see next paragraph]. However, evolving neuroimaging techniques should soon enable more thorough investigation of brainstem processing of human visceral pain.

Evidence from brain imaging studies in visceral sensory and affective neuroscience The ‘visceral sensation/pain neuromatrix’ was outlined by numerous functional brain imaging studies assessing brain responses during oesophageal, rectal, and, to a lesser extent, gastric distension, mostly using barostat distension protocols. It consists of the cortical and subcortical regions described above.\textsuperscript{164} Moreover, a recent study confirmed the involvement of several distinct brainstem regions, including the PAG and RVM regions, in the processing of visceral sensation.\textsuperscript{165}

It is, however, interesting that affective neuroscience is providing growing evidence for the hypothesis that interoceptive neurohumoral signals, especially from the viscera, and the brain regions processing such signals, are crucial in the generation and regulation of emotions and feelings.\textsuperscript{93,166–168} In a recent positron emission tomography study, Damasio et al. induced four different emotions in healthy volunteers using autobiographical memory scripts. Importantly, scanning only started when the subjects indicated that they actually started feeling the emotion.\textsuperscript{166} Virtually all regions that are known to process (visceral) sensory information [brainstem nuclei, insula, ACC, SII and orbitofrontal cortex (OFC)] were found to be involved in the feeling of emotional states,\textsuperscript{166} providing a neurobiological link between emotions and (visceral) sensation.

Several recent studies have compared brain responses during GI distension between FGID patients and healthy controls [reviewed in Refs\textsuperscript{43,91}].

In IBS, abnormalities in brain processing of rectal sensory signals have mainly been found in the medial pain system [subregions of the ACC], providing evidence for abnormalities in the affective and/or cognitive dimension of the pain experience, which might be one aspect of a more generalized state of negative affectivity. Generally, these alterations in ACC activity can be explained in terms of altered visceral afferent input to the brain and/or abnormal affective or cognitive responses to visceral afferent signals at the level of the CNS itself. In a number of studies, IBS patients showed higher ACC activity during painful rectal distension when compared with healthy volunteers\textsuperscript{8,169–171} Upregulation of visceral afferent input or increased ACC response [due to increased anticipation, hypervigilance or negative affective reaction to the visceral sensory stimulus] may account for these findings. However, in an equal number of other studies, lower or absent ACC activity was found as a brain response to rectal distension in IBS patients when compared with controls,\textsuperscript{172–175} which may be due to failure to activate descending antinociceptive pathways originating at the level of the ACC, ceiling effects or differential sensitization of the lateral, compared with the medial pain system in IBS.

It should be noted that heterogeneity in patient samples, stimuli applied and imaging methods used may at least partly account for the discrepancies in brain imaging findings in IBS.\textsuperscript{176} It is, for example, known that within the IBS group, a history of [childhood] sexual or physical abuse may alter brain responses to rectal distension.\textsuperscript{175} In conclusion, despite these discrepancies, there is a growing body of evidence supporting abnormal affective processing of visceral sensation in IBS patients. Furthermore, it has recently been shown that cognitive behavioural therapy (CBT) is associated with a reduction of baseline activity in the right subgenual ACC and the left medial temporal lobe [including the amygdala] of IBS patients, which was accompanied by improvements in GI symptoms, anxiety and worry. These brain activity changes may be the biological substrate of reduced attention to visceral stimuli or visceral-specific anxiety as a result of CBT in these patients.\textsuperscript{177}

In FD, brain imaging evidence is more sparse when compared with IBS. However, it was recently shown that in healthy volunteers, painful proximal gastric distension activates regions that are generally consistent with the visceral sensory ‘neuromatrix’,\textsuperscript{178,179} although there has been some debate regarding the role of somatosenory cortices.\textsuperscript{180,181} Functional dyspepsia patients who are hypersensitive to gastric distension showed similar activation of the lateral pain system [sensorimotor cortex] when compared with controls, although at far lower intragastric pressures and volumes [for similar pain or discomfort scores].\textsuperscript{182} This may provide a biological substrate for their hypersensitivity, but does not necessarily mean that central cognitive or affective processes are not involved [for an excellent account of this topic, see Ref.\textsuperscript{37}]. Furthermore, no activation of the medial pain
system was found, which may again be explained in several ways, as described above. A recent functional magnetic resonance imaging study, Phillips et al. showed that, in healthy volunteers, non-painful oesophageal stimulation is associated with greater neural activity in the dorsal ACC and the insula during fearful, compared with neutral emotional context produced by presentation of fearful or neutral facial expressions. Furthermore, discomfort, anxiety and activity in dorsal ACC and insula were significantly higher when high-intensity, compared with low-intensity fearful expressions were presented. The same group also found that selective and divided attention modulate the cerebral processing of oesophageal sensation in somatosensory and anterior cingulate cortices in a different way. These studies provide further evidence for an important influence of cognitive and affective factors on visceral sensation and its neurobiological correlates.

FUTURE POTENTIAL TARGETS FOR TREATMENT OF FGID

Recent progress in our understanding of sensory transduction of visceral afferents within the GI tract and spinal dorsal horn has resulted in potential new therapeutic targets being identified. Of these, the most promising targets on visceral afferents to reduce PS are the TRPV1 proton-activated receptor, the protease-activated receptor-2 via mast cell tryptase release, NMDA receptor subunit antagonists and VGSC, particularly Nav 1.8 and Nav 1.9, which have recently been shown to play an important role in the development of visceral hyperalgesia. Other potential targets include NGF acting at trk receptors which not only sensitize sensory neurones peripherally, but also mediate CS via increased gene transcription, prostanooid receptor antagonists acting peripherally and spinally at EP receptors and neuropeptidin receptor antagonists. Modulation of the descending facilitatory and inhibitory pathways also provides a novel area for therapeutic manipulation of visceral pain processing, with recent work suggesting a role for endogenous cannabinoid receptors. However, it must be recognized that these potential targets described above are aimed predominantly at modulating sensory visceral nociception and not the motor disturbances or altered brain processing of the stimuli such as serotonin agonists/antagonists or CRF antagonists. As our understanding of the pathophysiology of FGIDs increases, treatments will become targeted at specific underlying mechanisms and so provide more effective therapy than is currently available.

SUMMARY

An integrated biopsychosocial understanding of changes in intestinal sensory, motor and CNS activities is providing a conceptual model for FGID. In this model, higher neural centres are modulating peripheral intestinal sensory and motor activities and spinal sensory input, while the processing of these inputs by the brain can be influenced by psychological distress, which contributes to the generation of symptoms.

Several independent lines of evidence point towards an important role of psychosocial factors in FGID in general and an interaction between psychosocial factors and visceral sensitivity in particular. This yields support for a true biopsychosocial model of FGID, where we have to take the complex reciprocal relationship between psychosocial factors (including hypervigilance, stress, history of abuse and psychiatric comorbidity) and GI sensory and motor function (through their respective biological substrates) into account. This is necessary if we want to fully understand and elucidate the pathophysiology of FGID, as also in order to provide the best possible multidisciplinary care for this patient population that is often notoriously difficult to treat.

Visceral pain hypersensitivity is recognized as a characteristic feature in patients with FGID. Its pathophysiological basis is a combination of sensitized visceral afferent pathways, alterations in cortical processing of visceral afferent inputs and changes in descending modulatory inputs from the brainstem to the spinal cord and enteric neurones via the vagus nerve. However, the amount each step contributes to the overall perception of visceral pain hypersensitivity and hence symptom generation in individual patients still remains unclear.

With a continual increase in our knowledge of the mechanisms responsible for symptom generation in FGIDs, it is likely that the current classification system based on patients’ symptoms will eventually change to be based on the underlying pathophysiological mechanisms. Greater understanding of the receptors (and eventually the genes) involved in each step will then ultimately lead to improved diagnosis and subsequent treatment of these disorders. However, as with all new therapeutic advancements, the promise of potential new treatments for visceral hypersensitivity in FGIDs will only be effective in the context of a patient-centred biopsychosocial plan of care with an effective patient–doctor relationship.

ACKNOWLEDGEMENTS

P. Anand thanks Dr A. Akbar and Mr P. Facer for helping with the text and figures. L. Van Oudenhove is...
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