Central Processing of Noxious Stimuli in Patients with Irritable Bowel Syndrome Compared to Healthy Controls

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ABSTRACT

James Steven Heymen: Central Processing of Noxious Stimuli in Patients with Irritable Bowel Syndrome Compared to Healthy Controls
(Under the direction of Kathleen C. Light, Ph.D.)

Irritable Bowel Syndrome (IBS) is a complex disorder of unknown etiology. Research into the pathophysiology of IBS suggests the involvement of psychological, hormonal, immunological, genetic, cardiovascular, and autonomic nervous system factors, as well as peripheral and central sensitization of pain signals in the etiology and/or maintenance of IBS. Visceral hyperalgesia is consistently observed in IBS patients. However, recent investigations have found evidence of somatic hyperalgesia, not seen in earlier studies, suggesting the possibility of a dysfunction in central pain regulatory mechanisms.

Evidence suggests a role for central sensitization in IBS pain. Psychophysical investigations into dysregulation of the endogenous pain regulatory mechanisms of temporal summation and diffuse noxious inhibitory controls (DNIC) have been consistently demonstrated in other chronic pain conditions such as Fibromyalgia and Temporomandibular Disorder, which show high comorbidity with IBS.

The primary objective of this investigation was to explore the role of central sensitization in IBS pain by assessing both efferent (DNIC) and afferent (temporal summation) central modulation of nociception in IBS patients. Group differences in psychological, autonomic nervous system, and general pain measures were also assessed.

Forty eight pre-menopausal females (27 with IBS) participated in this investigation.

No group differences were seen in temporal summation or the DNIC effect on temporal summation. Similarly, no group differences were seen in any general pain measures or in sympathetic tone. IBS subjects reported significantly greater stress than Controls on measures of; state anxiety, depression, catastrophizing, and anger-out expression. IBS subjects also demonstrated significantly lower levels of DNIC than Controls during noxious tonic conditioning stimuli. However, non-noxious conditioning stimuli also produced an apparent DNIC effect in a counterbalanced design. After controlling for non-specific effects occurring in the non-painful conditioning protocol (distraction, and psychological measures associated with DNIC), IBS subjects continue to show deficient DNIC ($p < 0.01$).

This is the second investigation that has attempted to account for non-specific effects in the investigation of DNIC. Only by controlling for non-specific effects, can evidence of deficient DNIC can be attributed to dysregulation in endogenous analgesic mechanisms. Further studies are needed to elucidate whether deficient DNIC is a cause or consequence of IBS pain.

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β	beta (slope)
η^2	eta squared (effect size)
C	Celsius
ANOVA	analysis of variance
ANCOVA	analysis of covariance
IBS	Irritable Bowel Syndrome
THR	Pain Threshold
TOL	Pain Tolerance
APR	Average Pain Rating
MPR	Maximum Pain Rating
WPR	Water Pain Rating
DNIC	Diffuse Noxious Inhibitory Controls
RR	Rate of Rise
NMDA	N-methyl-D-aspartate
CGRP	calcitonin gene-related peptide
FMS	fibromyalgia syndrome
TMD	temporomandibular disorders
MRI	magnetic resonance imaging
fMRI	functional magnetic resonance imaging

Chapter 1

Introduction

Irritable Bowel Syndrome (IBS) is characterized by chronic abdominal pain that is associated with changes in bowel function and is one of the most common reasons for visits to primary care physicians or gastroenterologists (2.4 to 3.5 million visits/year) in the United States. Approximately 15 % of the U.S. population suffers from IBS (Drossman et al., 1993) and is more prevalent in women, with estimates ranging from a 2:1 ratio in community samples (Saito et al., 2000) to a 4:1 ratio in treatment seeking populations (Camilleri, 1999). In 1998, IBS accounted for direct health care costs totaling $1.6 billion (American Gastroenterological Association, 2001), and has been linked to a doubling of health care visits per year, from 4.5 to 9.0 (Levy et al., 2000b). This may be due to the fact that only 50% of IBS patients are satisfied with the treatments for IBS that are currently available to them (Whitehead W.E. et al., 2004; Thompson et al., 1997).

IBS patients, by definition, do not demonstrate any observed pathological condition. Although recent evidence suggests a role for serotonin reuptake transporter polymorphisms (Kim et al., 2004; Pata et al., 2002; Yeo et al., 2004), or dysregulated immunological activity (Gwee et al., 1999; Gwee et al., 2003; O'Mahony et al., 2005; Spiller, 2003) in some IBS subgroups, the etiology of IBS is unknown.

Psychological Influences on Irritable Bowel Syndrome

Psychological stress is known to precipitate symptoms in patients with IBS (Whitehead et al., 1992). Although psychosocial factors affect motility in IBS and healthy controls, stress increases motility more so for patients with functional gastrointestinal disorders than for healthy controls (Drossman et al., 1997). In addition, levels of chronic life stress predict clinical outcome of IBS (Bennett et al., 1998) and psychological symptoms define severity and status in IBS (Drossman, 1999).

The evidence regarding the influence of psychological factors on IBS is mixed. It is known that IBS patients have an increased prevalence of psychiatric disorders, such as anxiety, depression, personality disorders, and somatization, compared to the general population. Gwee and colleagues (1996; 1999) showed that patients with acute gastroenteritis were more likely to develop IBS if they had higher levels of anxiety, somatization, and depression than those who did not develop IBS. However, Chun et al. (1999) reported that rectal hyperalgesia was not related to psychological distress, healthcare seeking behaviors, or general chronic functional pain disorders.

In a review of the evidence for psychological influences on perception of abdominal pain, Whitehead and Paulson (1998) reported that 66% of IBS patients report pain at abnormally low rectal distensions. This finding did not correlate with anxiety or depression in IBS patients. IBS patients even rated sham distensions as painful, but control subjects did not. When psychological factors were controlled for, there was no difference between IBS and controls in pain ratings of sham distensions. This may indicate psychological factors played a role in IBS subjects identifying sham distensions as painful. In addition, manipulating attention and arousal with stress or relaxation, had a substantial effect on pain reports by IBS subjects.

Studies have shown that how a person responds to stress also influences pain sensitivity. Burns and colleagues (2004) demonstrated that an anger-out expression style was associated with higher pain intensity in patients not taking opioids, even after controlling for anxiety and depression. Similarly, Bruehl and colleagues (2003) found that greater anger-out scores on an anger management style questionnaire were associated with hypersensitivity to pain and with a reduced naloxone blockade effect, indicative of impaired opioid anti-nociception.

In a recent study, Granot and colleagues (2002) reported that scores on the Pain Catastrophizing Scale predicted postoperative pain following elective abdominal surgery. Gracely and colleagues (2004) suggested that catastrophizing influences pain perception, by increasing attention and anticipation, and emotional responses to pain. This was based on the fMRI findings that greater catastrophizing scores were associated with elevated activity in brain areas involved in anticipation of pain (medial frontal cortex and cerebellum), attention to pain (dorsal anterior cingulate cortex and dorsolateral prefrontal cortex), emotional aspects of pain (claustrum) and motor control, in FMS patients, after controlling for effects of depression.

Edwards and colleagues (2004) reported that elevated catastrophizing scores were unrelated to experimental pain thresholds during clinical pain conditions. However, after the pain was successfully treated, baseline catastrophizing scores were inversely related to pain threshold and pain tolerance measures. This suggests that catastrophizing may be a consequence of suffering from chronic pain. France and colleagues (2002a) showed that catastrophizing scores on the Coping Strategies Questionnaire were not related to the nociceptive flexion reflex, RIII, but was correlated with pain ratings in young healthy

subjects, suggesting that the relationship between catastrophizing and increased pain reports is not related to differential modulation of spinal nociception.

Psychiatric disorders and psychological distress are commonly associated with IBS, although a causal relationship has not been established. Houghton and colleagues (2002) found that hypnotherapy was successful in normalizing abnormal visceral sensitivity and reduced pain in IBS patients. This may suggest a role for the central nervous system in regulating visceral pain.

It has been suggested that IBS is actually several different disorders resulting from different etiologies (Rodrigues et al., 2005) Inflammation and immune dysfunction has recently been linked to IBS and genetic polymorphisms have also recently been identified in IBS patients (Pata et al., 2002; Kim et al., 2004; Yeo et al., 2004). Others have suggested that IBS is part of a broader disorder of pain dysregulation that includes other chronic pain disorders such as fibromyalgia (FMS) and temporomandibular disorders (TMD) (Siddall & Cousins, 2004; Aaron et al., 2000; Kleinbohl et al., 1999). Up to 80 % of IBS patients also have other gastrointestinal chronic pain disorders, such as dyspepsia or upper gastrointestinal pain. In addition, many patients with IBS also have somatic chronic pain conditions, such as FMS, and TMD and Veale and colleagues (1991) has shown that approximately 70% of FMS patients have chronic visceral pain.

The majority of IBS patients have been found to be hypersensitivity to balloon distension of the rectum, demonstrating higher pain ratings (Naliboff et al., 1997), lower visceral pain thresholds (Mertz et al., 1995) , and pain tolerance (Whitehead et al., 1990) than controls. Although not all IBS patients demonstrate this hypersensitivity, visceral hyperalgesia is considered to be a biological marker for IBS (Chun et al., 1999; Mertz et al., 1995). Whitehead and colleagues (1998) reported that approximately two-thirds of IBS

patients have visceral hypersensitivity to rectal distensions. It is not known whether visceral hyperalgesia is a result of peripheral or central sensitization. Those investigators that suggest that IBS pain results from central dysregulation of pain processing differ in whether the dysfunction is in the brain or in the spinal cord. Recent brain imaging studies show differences in pain centers in the brain during noxious rectal distensions (Verne et al., 2003a; Mertz et al., 2000; Wilder-Smith et al., 2004), while other studies have demonstrated spinal cord involvement in IBS pain based on alterations in the nociceptive flexion (RIII) reflex during rectal distensions (France et al., 2002b).

Contrary to earlier reports, most recent investigations have also demonstrated somatic hyperalgesia in IBS patients (Bouin et al., 2001; Verne et al., 2001; Verne et al., 2003b; Rodrigues et al., 2005). This new data suggests the possibility that IBS patients may suffer from a global dysregulation in central pain processing similar to that seen in other chronic pain disorders, such as Fibromyalgia (FMS) and temporomandibular disorders (TMD). Investigations to explore central sensitization in these and other chronic disorders have focused on two aspects of central sensitization, namely; 1) exaggerations in the wind-up phenomenon that facilitates ascending pain signals, and 2) deficits in DNIC (disinhibition), that normally provides tonic descending antinociceptive signals. Research consistently shows enhanced temporal summation of afferent pain signals as well as deficits in Diffuse Noxious Inhibitory Controls (DNIC) in FMS (Lautenbacher & Rollman, 1997; Staud et al., 2001; Staud et al., 2003) and TMD patients (Maixner et al., 1995; Maixner et al., 1998; Kashima et al., 1999; Sarlani et al., 2004) and TMD patients compared to healthy controls, leading these experts to conclude that this central alteration in pain signaling may be important in the onset or worsening of chronic pain symptoms (Edwards et al., 2003b; Staud & Rodriguez, 2006; Verne & Price, 2002).

Neurophysiology of Pain

Pain occurs when noxious thermal, mechanical, or chemical, stimuli excite peripheral terminals of specialized primary afferent neurons called nociceptors (Woolf, 2000). There are two types of primary afferent nociceptors, myelinated A-delta mechanosensitive and unmyelinated C-fiber Polymodal (thermal, mechanical, chemical) nociceptors (Price & Dubner, 1977). A-beta mechanosensitive afferent stimuli do not evoke pain in humans, even at high intensity.

A-delta afferents conduct 1st pain, which is an immediate, sharp, pricking pain at 3-30 meters per second (Willis, 1985; Price, 1972). C-fiber afferents conduct 2nd pain (1-1.5 sec later), which is a burning or throbbing type of pain at a much slower .5 to 2.0 meters per second (Price, 1972; Willis, 1985). There are three types of A-delta nociceptive afferents: 1) mechanical receptors high thresholds, and type 1 and type 2 mechano-thermal nociceptive afferents. Unmyelinated C-polymodal nociceptive afferents outnumber myelinated A-delta nociceptors, 3:1 (Price & Dubner, 1977), but in primate limbs, more than 90 % of cutaneous nociceptors are C-fiber polymodal neurons.

Nociceptive neurons elicit a variety of responses from the central nervous system including spinal cord withdrawal reflexes, autonomic and neuroendocrine responses, as well as behavioral consequences of general alertness, arousal, and orientation (Palmgreen et al., 2002). Secondary, or spinal nociception is primarily conducted by wide-dynamic-range neurons and some nociceptive specific neurons. Wide dynamic range neurons can detect temperature changes from 0.2 to 0.3 degrees in the painful range, from 45 to 51^{0} Celsius (C), which nociceptive specific neurons cannot. Nociceptive specific nociceptors conduct 1st pain, and 2) wide dynamic range neurons conduct heat, pinch, and cold sensations in response to 2nd pain, C-fiber signaling (Price & Dubner, 1977). Most Lamina V neurons are wide

dynamic range neurons, which do not differentiate tissue of origin and represent the integration of all afferent input to the dorsal horn.

Ascending Pathways from the Gut

Visceral pain is poorly localized for several reasons: 1) visceral afferents communicate to several dorsal horn connections across a wide rostral-ventral segment of the spinal cord, 2) visceral nociceptors converge with somatic afferents at the dorsal horn, and 3) visceral information travels vagal pathways to the brainstem leading to contrasting properties in the signaling process.

Visceral nociceptors are located within the mucosa and muscle layers of the gastrointestinal tract. Most are non-myelinated C-polymodal neurons, which respond to mechanical, chemical, and thermal stimulation. Also in the mucosa are myelinated, A-delta fibers, which primarily respond to mechanical and thermal stimuli (Crowell et al., 2005). Stimuli initiate a signal in these first order neurons, which pass through the splanchnic nerve to the sympathetic nervous system, through the dorsal root ganglion up to the dorsal horn. Secondarily, post-synaptic nerves begin in the dorsal horn, traveling up the spinothalamic tract, which crosses contralaterally, and spinoreticular tract within the spinal cord. Spinoreticular tract signals then synapses with thalamic and reticular formation nuclei in the pons and medulla. Finally, third order neurons continue to the limbic system and frontal cortex. In a parallel fashion, signals traveling through the spinothalamic tract travel through the aqueductal gray area of the midbrain, to the thalamus, where third order neurons then communicate to the sensory-motor cortex.

First order neurons release glutamate, which initially activates non N-methyl-D-aspartate (NMDA) receptors of the second order neurons. NMDA receptors are initially blocked by the presence of magnesium. Glutamate also triggers the release of peptides:

substance P, calcitonin gene-related peptide (CGRP), and neurokinen A, from the pre-synaptic primary afferent terminal, which depolarizes the post-synaptic membrane removing the magnesium block and allowing calcium to enter NMDA receptors (Mayer et al., 1999). Intra-cellular calcium then combines with calmodulin to create protein kinase, which leads to nitric oxide oxidase. Nitric oxide acts either as an intracellular messenger depolarizing the cell or as a neurotransmitter which causes presynaptic release of the primary visceral afferent (Mayer et al., 1999).

Descending Pathways

Descending pain regulatory signals originate in the insula, thalamus, anterior cingulate, pre-frontal cortex, amygdala, hypothalamus and periductal gray area. Descending signals travel through the reticular activating system to the dorsal horn in both inhibitory and facilitative pain modulation. There are multiple spinoreticular/reticulospinal loops on each side of the body that these pass information in both directions, and may be inhibitory or facilitatory. They connect the spinal cord to: the dorsolateral pontine tegmentum, the rostral ventral medulla, the dorsal medulla, the caudal medulla, and the lateral hypothalamus. Other descending pain modulating pathways include: the cerebral cortex connections to the nucleus gracilis and cuneatus, reticular formation, and the thalamus; the peri-aqueductal grey matter direct connections to the dorsal, or to the raphe nuclei of medulla, and then to dorsal horn; and the locus ceruleus connections to the dorsal horn.

The inhibitory pathway reduces nociception by releasing peptides and neurotransmitters that reduce the ascending pain signal. These include: serotonin and norepinephrine which activate the opioids, glycerine, gamma-aminobytric acid and cholecystokinen from interneurons to blunt the pain signal. The facilitative pathways release

additional glutamate, which increases NMDA receptor activation to increase pain signals (Mayer et al., 1999).

Visceral Hyperalgesia in Irritable Bowel Syndrome

Although visceral pain is the most common form of pain produced by disease, there has been far more research into the mechanisms for neurogenic or somatic pain (Cervero & Laird, 2004). The amount of visceral pathology is often not associated with the intensity of the pain in visceral pain states. For example, colitis and gastrointestinal ulcerations are not associated with significant pain, but pain is the central feature in functional gastrointestinal disorders, such as; IBS, dyspepsia, post-cholosistectomy syndrome, interstitial cystitis, and functional chest pain (Mertz, 2003), which show demonstrable no pathology. Visceral pain can lead to two forms of hypersensitivity: 1) Somatic pain in dermatomes that converge at the same dorsal horn neuron as the visceral afferents (referred hyperalgesia), and 2) enhanced sensitivity of the same or nearby viscera (visceral hyperalgesia). Giamberardino et al. (2000) suggests that the source of these sensory alterations must be the central nervous system because they often originate in healthy tissues. Considerable research suggests that enhanced excitability of spinal cord neurons substantially contribute to for visceral hyperalgesic states (Garrison et al., 1992; Hummel et al., 1997; Roza et al., 1998).

Although the etiology of IBS is unknown, patients have been shown to have visceral hypersensitivity to gut distensions. In addition, dyspepsia or upper gastrointestinal pain occur comorbidly in up to 80 % of IBS patients (Agreus et al., 1995). Ritchie, et al. (1973) first demonstrated lower pain thresholds in response to balloon distensions in the bowel in IBS patients compared to controls. Since this pioneering investigation, many researchers have confirmed this finding (Whitehead et al., 1980; Bouin et al., 2004; Hobday et al., 2000; Kwan et al., 2004; Mertz et al., 1995; Steens et al., 2002). Visceral hyperalgesia has been

consistently shown to be present in the majority of patients with IBS and is considered to be a biologic marker for IBS (Mertz et al., 1995), (Chun et al., 1999).

Somatic Hyperalgesia in Irritable Bowel Syndrome

Early studies found IBS patients to have higher or similar pain thresholds (THR) or pain tolerance (TOL) to noxious somatic stimuli compared to healthy controls (Cook et al., 1987; Accarino et al., 1995; Rossel et al., 1999; Whitehead et al., 1990; Zighelboim et al., 1995). However, most, but not all (Chang et al., 2000), recent investigations have found significant somatic hyperalgesia in IBS patients (Bouin et al., 2001; Verne et al., 2000; Verne et al., 2003c). These studies suggest the possibility of a central dysregulation in normal pain signal processing as a potential mechanism in the etiology of IBS pain (Verne & Price, 2002).

In addition, Rodriguez (2005) and Verne (2003b) demonstrated that somatic pain sensitization was not limited to afferents in the L4-L5 dermatome, and that there was not a sensitization gradient across three sites tested (face, arm, and leg). These results suggest that somatic hyperalgesia in IBS is not limited to areas of visceral/somatic convergence. Rodrigues (2005) suggested that spinal hyper-excitability is consistent with this wide distribution of somatic hyperalgesia. In addition, Verne and Price (2002) describe IBS s a common precipitant of central sensitization.

Sensitization

It is known that sensitization occurs in IBS patients after repeated rectal distensions, leading to hyperalgesia of the rectum (Whitehead & Palsson, 1998; McRoberts et al., 2001; Munakata et al., 1997b). Bouin (2002) has identified lowered rectal pain thresholds as a hallmark of IBS, stating, “Dysregulation in the neurobiology of visceral afferents and pain sensitivity control (inhibition) is believed to explain IBS symptoms. It is not known whether this is due to peripheral, to central sensitization, or both.

Peripheral sensitization occurs when polymodal C-fiber nociceptors and A-fiber mechanonociceptors increase their sensitivity after repeated noxious stimulation which is unique to the nociceptive system. Sensitization results in a decreased threshold, an increased response to supra-threshold stimuli, and spontaneous activity. Inflammatory mediators such as prostaglandin's, bradykinin, and adenosine triphosphate, sensitize primary afferents, especially C-fiber polymodal receptors, as well as recruit silent nociceptors. Recently, however, non-inflammatory mediators, such as glycerol sensitase, glutamate, and trypsin (released by stress) have been shown to trigger visceral pain and lower mechanical stimulation thresholds (Bueno & Fioramonti, 2002). The idea of an "inflammatory soup" may explain why medications with a variety of mechanisms of action have shown only partial benefit in treating IBS (Kirkup et al., 2001).

Central sensitization is the facilitation of central nervous system nociceptive neurons triggered by peripheral injury or nociceptive input from C-fibers (Woolf & Wall, 1986). It is characterized by reduced stimulation thresholds, an expansion of receptive fields, and an increase in background activity of spinal neurons. Strong or prolonged primary nociception causes plasticity leading to increased synaptic strength between the nociceptors and spinal neurons, so that smaller signals trigger pain (Woolf & Wall, 1986). Woolf (2004) suggested that chronic pain conditions like IBS and Fibromyalgia may be due to central sensitization.

Peripheral or Central Sensitization in Irritable Bowel Syndrome

The exact cause of visceral hyperalgesia in IBS patients is not known (Delvaux et al., 2004). Gastric hypersensitivity could be due to local or central mechanisms following an event (Gebhart et al., 2002). Munakata et al. (1997a) reported that repeated stimulation of sigmoid splanchnic afferents led to central sensitization manifested as hyperalgesia and viscerosomatic referral during rectal distensions and to spontaneous rectosigmoid

hyperalgesia in the absence of applied stimuli, concluding that repetitive sigmoid contractions may induce rectosigmoid hyperalgesia in IBS.

Visceral hyperalgesia, seen in IBS patients, may be due to dysregulation of inhibitory or facilitative central processes, or from peripheral dysregulation, or some combination of these. Recent investigations suggest that chronic pain may be primarily the result of central sensitization. Lembo et al. (1994) reported that lidocaine in the rectum did not affect hypersensitivity in IBS patients, indicating lumbarsplanchnic pathways are involved in visceral hypersensitivity. Lembo et al. (2000) also found that IBS patients may have a diminished central release of endogenous opioids in response to visceral stimuli, based on their response to Fentanyl.

On the other hand, Vase et al. (2003) showed hyperalgesia was due to either spinal or peripheral influences, but not brain function, after local anesthesia of the rectum reversed hyperalgesia. Local lidocaine reversed hyperalgesia in the rectum, suggesting that central sensitization is maintained by tonic stimulation from rectum or colon (Verne et al., 2003b). Chey et al. (1995) found that Octreotide reduced rectal perception of electrical stimuli and suggested that this was associated with inhibited cerebral and spinal evoked potentials indicating an effect on spinal afferent pathways. In a study evaluating brain function in response to a non-visceral stimulus, Bloomhoff et al. (2000) found subjects with IBS to show hyper-reactivity in forebrain event-related potentials to auditory stimuli. The authors observed that aberrant brain functioning in response to non-visceral stimuli in their investigation, as well as to visceral stimuli in many previous studies, might indicate that aberrant central processing may be an aspect of IBS.

In an animal model, Miranda et al. (2004) demonstrated convergence of visceral and somatic nociception by sensitizing gastrointestinal neurons using somatic pain stimuli. The

authors report that ionotropic glutamate receptors in the spinal cord are involved in sensitizing neurons that are sensitive to colorectal distension, concluding that "Sensitization occurs at the spinal level and is independent of supraspinal influences". Peles, Miranda, and colleagues (Peles et al., 2004) then demonstrated that nociceptive somatic stimuli sensitized viscero-somatic convergent spinal neurons that respond to rectal distensions, and that a selective NMDA antagonist and an α-amino-3-hydroxy-5-methylisoxazole-4- propionic acid antagonist significantly attenuated the sensitized response to rectal distension in both intact and spinalized animal (Peles et al., 2004)

Convergence was also demonstrated in a human model by Coffin and colleagues (2004) by testing the effects of rectal distensions on the somatic nociceptive reflex (RIII), which is an objective index of spinal nociceptive processes. IBS subjects were shown to have hyper-excitability of spinal nociceptive processes, when rapid rectal distensions reduced inhibitions of the somatic reflex RIII seen on electromyography. Slow-ramp distensions also facilitated RIII reflex in IBS subjects (Coffin et al., 2004; Roza et al., 1998).

Al-Chaer and colleagues (2000) developed an animal model of IBS and demonstrated visceral and somatic hyperalgesia in adult rats after subjecting them to rectal mustard oil as neonates demonstrating that increased wide dynamic range neuronal activity was involved in both visceral and cutaneous hyperalgesia. Kawasaki and Al-Chaer (2003) found increased firing of somatic neurons after colorectal distension and vice-versa demonstrating convergence of visceral and somatic afferents at shared dorsal horn sites. In another demonstration of central viscerosomatic convergence, electrical stimulation of the leg facilitated visceral hyperalgesia, which was then blocked by ionotropic glutamate receptor antagonists administered to the lower spinal cord (Miranda et al., 2004). Willert et al. (2004) prevented and then reversed esophageal hypersensitivity in the proximal esophagus induced

by acid infusion in the distal esophagus by administering ketamine, an NMDA antagonist, indicating central sensitization as a mechanism in visceral hypersensitivity.

Roza and colleagues (1998) identified changes in spinal neurons following nociceptive stimuli to the ureter of rats leading to visceral hyperalgesia. Further they found that continued noxious visceral input following induced visceral hyperalgesia created changes in the dorsal horn neurons that could not be explained by a generalized excitability and suggested that this central hyperalgesic state depends on alterations in both central inhibitory and facilitative activity.

In a magnetic resonance imaging (MRI) study, Verne et al. (2003b) found that both, visceral and cutaneous hyperalgesia was accompanied by increased activity in all levels of somatosensory afferent processing in the brain and concluded that hyperalgesia was due to increased afferent processing (which could include disinhibition) ascending to the brain and not due to selective increased activity at higher cortical levels (limbic and prefrontal areas). Verne et al. (2003b) also reported that MRI activity was not different (comparing IBS patients to healthy controls) during both, rectal noxious stimuli or during cutaneous noxious stimuli, leading the authors to conclude that the difference in pain ratings between IBS and healthy controls must be due to afferent signaling from the spinal cord or periphery. These findings are consistent with the idea that mechanisms involved in visceral and cutaneous hyperalgesia, at least in part, occur at the spinal level. Finally, in a review of visceral hyperalgesia studies, Verne and Price (2002) suggested that the frequency of extra-intestinal symptoms that are commonly seen in patients with IBS suggests a central hyperalgesic state.

Normal processing of noxious stimuli is complex, as it involves multiple central modulating systems that influence the pain signal before it reaches the thalamus. These include; ascending facilitation of nociception and descending modulation of these signals

which can be facilitative or inhibitory. Repetitive or intense noxious stimuli can result in sensitization of these pathways, both peripherally and centrally. Psychophysical investigations into abnormalities in pain processing in patients with chronic pain conditions have focused on afferent measures of temporal summation, and efferent DNIC effects.

Temporal Summation

Investigations to explore dysregulation of central pain mechanisms in chronic pain patients have focused on identifying exaggerations in temporal summation and deficits in DNIC. Temporal summation describes an increase in pain sensitivity in response to the 'wind-up' of second-order neurons, primarily in the dorsal horn. Wind-up is the increase in magnitude of second order nociceptive neurons in response to repetitive, brief, noxious stimuli (< .33 Hz) (Staud & Rodriguez, 2006) applied to C-fiber primary nociceptive afferents (Herrero et al., 2000; Mendell, 1966). It is consistent and very reproducible (Herrero et al., 2000). Even non-painful gut perception depends on temporal and spatial summation (Serra et al., 1998). This allows for stimuli that are not normally perceived to achieve conscious sensation. If a noxious stimulus arrives at the synapse in the dorsal horn more frequently than once every three seconds, the post-synaptic electrical discharge becomes more prolonged, which results in an increase in the severity of the pain. Temporal summation is the psychophysical increase in pain sensitivity of the wind-up phenomenon (Ren, 1994).

Although wind-up is exclusively evoked by unmyelinated afferent C-fibers under normal conditions, it is not a property of peripheral C-fibers, but is a property of the central synapses of these fibers with post-synaptic spinal wide-dynamic range neurons and to a lesser extent nociceptive specific (NS) neurons and as such, is exclusively a central phenomenon (Price et al., 1977). Primary A-delta and C-fiber afferents do not temporally summate, but actually are progressively suppressed. Adaptation of A-delta and C-fibers to

repeated brief heat suggests that wind-up must be a central phenomenon. Further, wind-up is shown to be a central phenomenon because C-fibers show a reduction in response amplitude and action potential (Price & Dubner, 1977) and because NMDA antagonists reduce it. Neuropathic pain has been alleviated by NMDA antagonists (Backonja et al., 1994; Eide et al., 1995); Felsby, 1995; Krictensen, 1992; Mathisen 1995). WIND-UP may underlie a continuation of pain from prolonged nociception despite a reduction in their number of action potentials in afferent C-fibers (Price, 1972; Price & Dubner, 1977).

Wind-up is primarily the result of increases in the neurotransmitters, glutamate and aspartate and the neuromodulator, substance P, in the synapse between the C-polymodal primary afferents and dorsal horn neurons (Xu et al., 1992). These neurotransmitters activate NMDA receptors and neurokinen-1 receptors leading to prolonged depolarization of dorsal horn neurons. This wind-up phenomenon reflects a sensitization in the dorsal horn which leads to hyperalgesia (Price, 1999). Wind-up causes dorsal horn neurons to remain partially depolarized, which may cause spontaneous firing, hyper-responsiveness to weaker somatic stimuli. This leads to expansion and increased sensitization of the receptive fields (Li et al., 1999).

Wind-up can lead to spatial summation (of mechanosensitive A-delta afferents, but not heat sensitive C-fibers by expanding the receptive fields leading to recruitment of more dorsal horn neurons. Repetitive noxious stimulation of C-fibers leads to prolonged discharge of dorsal horn neurons. If wind-up is perpetuated beyond several minutes this can lead to long-term potentiation. This long lasting increase in the efficacy of synaptic transmission can last from one hour to several months Aziz (2000). Wind-up is believed to be instrumental in the creation or maintenance of chronic pain (Price, 1991).

Wind-up is usually tested using noxious phasic heat stimuli (Maixner et al., 1998; Staud et al., 2001; Staud et al., 2003) with increases in pain ratings indicating increases in temporal summation. While numerous studies have demonstrated enhanced temporal summation in somatic pain conditions, and wind-up of visceral afferents have been repeatedly demonstrated in animal models, only one, recent investigation has explored temporal summation using visceral noxious stimuli in human subjects (Sarkar et al., 2006). Sarkar and colleagues (Sarkar et al., 2006) demonstrated enhanced temporal summation to visceral electrical stimulation of the upper esophagus after inducing central sensitization using acid infusion to the lower esophagus in the first human model of visceral temporal summation. This mechanism has yet to be explored in patients with IBS.

Diffuse Noxious Inhibitory Controls

Counter-irritation has been known to decrease pain for centuries (Price, 1999), including references from Hippocrates stating that a stronger pain inhibits a weaker pain. This counter-irritation phenomenon is called Diffuse Noxious Inhibitory Controls (DNIC). DINC mechanisms act like a barrier to prevent the spread of pain and keep it bearable (Pielsticker et al., 2005) by providing a tonic inhibitory influence. The benefits of DNIC are evident in dangerous circumstances, such as when in battle, during injury, and even during sports activities (Tracey, 2004) .

Deficits in DNIC are believed to play an important role in geriatric and chronic pain (Edwards et al., 2003a). DNIC occurs when wide dynamic range neurons in the dorsal horn of the spinal cord (nociceptive specific neurons are not affected by this type of control) are substantially inhibited by a second nociceptive stimulus administered anywhere in the body distinct from their excitatory receptive fields (Le et al., 1992). In normal conditions, these inhibitions can be triggered only by nociceptive conditioning stimulus (CS) from A-delta- or

C-peripheral fibers. The inhibitions are very potent, affecting all of the activities of the wide dynamic range neurons.

Testing DNIC requires administering phasic, noxious, test stimulus (TS), prior to and, during administering tonic, noxious, CS, typically, ischemic, cold pressure, or noxious heat pain, heteroptopically (Guieu et al., 1994; Kosek & Ordeberg, 2000b) (Lautenbacher & Rollman, 1997; Staud et al., 2003; Edwards et al., 2003a), however, some investigators have used tonic CS with a slow-ramp increases in temperature to identify threshold and tolerance levels (Sigurdsson, 1994). The term "diffuse" is derived form heterotopic inhibition, as opposed to homotopic inhibition, as in transcutaneous nerve stimulation (TNS). The difference between the pain that is experienced with the first phasic pain stimuli, without the conditioning stimulus, and with the second phasic stimuli during the concurrent tonic pain is described as the DNIC effect.

Dysregulation of pain modulation due to deficient DNIC is believed to play an important role in chronic pain conditions like Fibromyalgia (Lautenbacher & Rollman, 1997; Staud et al., 2003; Kosek & Hansson, 1997), TMD disorders (Sigurdsson & Maixner, 1994; Kashima et al., 1999; Maixner et al., 1997), osteoarthritis (Kosek & Ordeberg, 2000a), headache (Sandrini et al., 2006), and low back pain (Peters et al., 1992), but not all (Johannesson et al., 2006; Leffler et al., 2002a; Leffler et al., 2002b), leading experts to conclude that this central alteration in pain signaling may be important in the onset or worsening of symptoms. In these subjects, DNIC was shown to be absent or significantly diminished compared to controls. Lautenbacher (1997) showed that concurrent painful tonic thermal stimuli induced DNIC to electrical and heat pain in controls, but not in Fibromyalgia subjects (at non-painful sites). The protection offered by properly functioning DNIC appears to be lacking in these Fibromyalgia patients. According to Lautenbacher, the nature of

Fibromyalgia, or any chronic pain condition, points to a deficient DNIC process reflecting decrements in endogenous analgesic systems.

In all but one DNIC investigation of chronic pain conditions that are reported in the literature (Wilder-Smith et al., 2004), DNIC was tested using phasic noxious TS, including; electrical (Lautenbacher & Rollman, 1997; Pielsticker et al., 2005; Sandrini et al., 2006), heat (Maixner et al., 1995; Staud et al., 2003), and pressure (Kosek & Hansson, 1997) stimuli, with noxious tonic stimuli including: ischemic pain (Kashima et al., 1999; Kosek & Hansson, 1997; Kosek & Ordeberg, 2000a; Maixner et al., 1995), hot (Staud et al., 2003) or cold water submersion of the hand or foot (Johannesson et al., 2006; Sandrini et al., 2006), or tonic heat thermode contact (Lautenbacher & Rollman, 1997; Pielsticker et al., 2005) as the CS. DNIC has also been tested in geriatric populations using heat (Edwards et al., 2003a; Washington et al., 2000), and electrical TS (Washington et al., 2000) and cold water submersion as CS (Edwards et al., 2003a; Washington et al., 2000) and found elderly to have deficient DNIC.

In the only study to investigate DNIC mechanisms in IBS patients Wilder-Smith (2004) demonstrated significant differences in fMRI, in brain centers known to control autonomic, emotion, and descending modulatory responses to pain in IBS subjects compared to controls (Wilder-Smith et al., 2004). These changes corresponded to a compromised DNIC effect in visceral pain ratings from rectal distensions, before and during the administration of a conditioning (foot in ice water) stimulus.

In the Wilder-Smith study (2004) as well as several other DNIC investigations changes in average pain ratings (APR) during heterotopic counter-irritation, compared to no counter-irritation protocols were used as a measure of the DNIC effect (Edwards et al., 2003b; Price & McHaffie, 1988; Staud et al., 2001), and changes in maximum pain ratings (MPR) during counter-irritation (Edwards et al., 2003b; Staud et al., 2001). Still others

compared test stimulus threshold (THR) or tolerance (TOL) values during counter-irritation compared to no counter-irritation conditions (Kashima et al., 1999; Kosek & Hansson, 1997; Kosek & Ordeberg, 2000b; Lautenbacher & Rollman, 1997; Maixner et al., 1995).

This investigation tested DNIC effects for heat pain THR and TOL during slow-ramp ascending method of limits paradigm, as well as for APR and MPR during phasic heat pain TS. In addition, a second conditioning stimulus that was not noxious in nature (neutral temperature water hand submersion) was include to attempt to isolate non-specific effects, such as distraction, that is known to play a role in the results of DNIC investigations. This strategy has been used by others (Edwards et al., 2003a; Sigurdsson & Maixner, 1994; Roby-Brami et al., 1987), however, these investigators simply identified a lack of significant group differences in DNIC during the non-noxious CS that was seen in the noxious conditioning stimuli condition. Edwards and colleagues (Edwards et al., 2003b) improved on this concept in a post-hoc analysis by subtracting the change in pain ratings that occurred during non-noxious conditioning from the DNIC effect found in the noxious conditioning protocol to yield a "controlled measure of the degree to which thermal pain ratings change as a function of heterotopic cold pain". This is an elegant way to remove non-specific effects from the change score in pain ratings that have nothing to due with the physiological effects of counter-irritation.

Summary and Goals

IBS is a complex disorder of unknown etiology. Research into the pathophysiology of IBS over the last decade has demonstrated the involvement of many factors contributing to IBS symptoms. Psychological, hormonal, immunological, genetic, cardiovascular, and autonomic nervous system factors, as well as peripheral and central sensitization of pain signals are believed to be involved in the etiology and/or maintenance of IBS. Visceral hyperalgesia is

consistently observed in IBS patients; however, recent investigations have found evidence of somatic hyperalgesia, not seen in earlier studies. This new data suggests the possibility that IBS patients may suffer from a global dysregulation in central pain processing.

There is increasing interest in research in central nervous system processes, in both somatic and visceral pain (Tracey, 2004). There is considerable evidence suggesting that central sensitization is likely to play a major role in IBS pain. Psychophysical investigations into abnormalities in central pain processing have been observed in TMD and FMS patients, both of which have significant comorbidity with IBS. Whitehead and colleagues (2002) reported that as many as 77% of IBS patients also suffer from FMS and 64 % of IBS also suffer from TMD (Whitehead et al., 2002; Veale et al., 1991). Veale et al. (1991) also reported that approximately 70% of FMS patients have chronic visceral pain. It seems likely that the central dysregulation in central pain processing seen in FMS and TMD patients may also be important in the onset or worsening of symptoms in IBS.

Investigations into central pain dysregulation in IBS may lead to improvements in diagnosis, and ultimately, to novel therapies for patients with IBS. The primary objective of the proposed investigation is to explore the role of central sensitization in IBS pain by assessing both efferent (DNIC) and afferent (temporal summation) central modulation of nociception in IBS patients.

Chapter 2

Methods

Participants

Potential subjects were recruited from the UNC Gastroenterology Division of the Department of Medicine (IBS) and through university-wide email and flyer advertisements (IBS and healthy controls) and screened by telephone interview to confirm that they met inclusion criteria.

Inclusion: Subjects were pre-menopausal women, 18 years of age and older. IBS subjects met Rome II criteria (Drossman et al., 2000) for IBS and were currently suffering from painful symptoms of IBS.

Exclusions: Exclusionary criteria included the following: menopause, pregnancy or nursing, major clinical depression or anxiety disorder, hypertension, history of abnormal electrocardiogram, heart disease, kidney disease, diabetes, seizures, asthma, or thyroid disorder. Individuals taking analgesics, narcotics, or antidepressants were excluded from participation. Other medications were evaluated on a case-by-case basis. Healthy subjects did not have a history of any chronic pain conditions.

In order to control for non-specific effects unrelated to central pain regulatory mechanisms, several controls were instituted regarding subject selection and testing protocols. In addition to excluding males and post-menopausal females, subjects were scheduled for their visit during the follicular phase of their menstrual cycle (days 4-8) to control for possible gonadal hormone influences on pain perception. To minimize

sympathetic nervous system influences on pain perception, all subjects were instructed to refrain from consuming caffeine and nicotine for at least two hours prior to testing and all subjects were tested at approximately the same time in the afternoon to control for fluctuations in cortisol. Attempts were made to minimize psychological influences on pain perception by excluding individuals with an anxiety or depressive disorder. In addition, a battery of questionnaires to identify group differences in mood, perceived stress, and stress coping strategies were administered and significant group differences were included as covariates in the appropriate statistical analyses. It is well known that distraction plays a role in psychophysical investigations that include counter-irritation such as DNIC. To minimize these effects, a control condition, providing the same sensory input, except for nociception, was employed as the "counter-irritation" stimulus.

This investigation was approved by the University of North Carolina Medical Internal Review Board (05-MED-841). This investigation lasted approximately six months. Each subject's participation lasted from two to two and one half hours, for a single visit. Each subject was paid $35 for completing the study. Following a review of the informed consent form, participating subjects were asked to complete several questionnaires.

Outcome Measures

Psychological Measures

Questionnaires were used to identify the basic demographic characteristics of all subjects and to provide a standardized psychosocial evaluation in addition to acquiring information about the participant's health history and current pain (See Appendix A). Demographic data and health history were acquired by the Demographic/medical history questionnaire (Levy et al., 2000a). Psychosocial assessment covered four major areas: cognitive-behavioral adjustment to pain, general psychological status, pain coping/pain responsiveness, and perceived levels

of stress. Characterization of current clinical pain was obtained with the Irritable Bowel Syndrome Severity (Francis et al., 1997) and the Physical Symptoms Questionnaire (Palsson et al., 2002). Psychological status was measured with the State-Trait Anxiety Inventory (Spielberger et al., 1983), the Anger Expression Inventory (Spielberger et al., 1995), and the Beck Depression Inventory (Beck, 1961). The Perceived Stress Scale (Cohen et al., 1983) measured recent exposure and responses to stress and pain coping style was assessed by the Catastrophizing subscale of the Coping Strategies Questionnaire (CSQ) (Hirsh et al., 2006).

Following completion of the questionnaires, all subjects were given a brief exposure to the heat stimuli apparatus and the ice water conditioning stimulus in order to minimize apprehension regarding the TS. This consisted of a practice trial for each of the three heat pain testing procedures (THR, TOL, and phasic heat pain stimuli) and a brief exposure to the CS, by submerging their right hand in 12^0 C water for approximately 10 seconds.

Sympathetic Nervous System Measures

After the subject's height and weight were acquired, the subjects right arm was instrumented with heart rate and blood pressure monitors (Acutracker, Suntech Instruments). After a 10-minute rest, baseline blood pressure and heart rate were acquired as baseline measures. Immediately after each pain testing protocol heart rate and blood pressure were reacquired.

General Pain Sensitivity Measures

1) Pain Threshold (THR) was the temperature at which a non-painful stimulus first became painful in an ascending method of limits paradigm.
2) Pain Tolerance (TOL) was the temperature at which a stimulus reached the maximum temperature that an individual was able or willing to tolerate in an ascending method of limits paradigm.

3) Average Pain Rating (APR) (Edwards et al., 2003b) was a measure of the average pain ratings given during the series of noxious phasic stimuli on a 0 to 100 scale, with 0 representing no pain, and 100 representing the most intense pain imaginable.
4) Maximum Pain rating (MPR)(Staud et al., 2001) (Edwards et al., 2003b) was the highest pain rating given during the phasic noxious stimulus series on a 0 to 100 scale, with 0 representing no pain, and 100 representing the most intense pain imaginable.
5) Water Pain Ratings (WPR) were acquired on a 0 to 100 scale, with 0 representing no pain, and 100 representing the most intense pain imaginable, immediately after the completion of the administration of the heat stimulus, but prior to removal of their other hand from the water.

Measures of Central Pain Modulation

1) Temporal Summation was assessed using two measures a) the rate of rise (RR)(Bhalang et al., 2005), or the slope of the least squares regression line of pain ratings during the first six phasic noxious stimuli provided at a rate of once every three seconds on a 0 to 100 scale (temporal summation is known to plateau after 5-6 stimuli in a train), and, b) Delta (Bhalang et al., 2005; Edwards, 2005), which subtracts the first pain rating on a 0 to 100 scale, with 0 representing no pain, and 100 representing the most intense pain imaginable, from the peak pain rating across phasic stimuli trains yielding the degree of increase during the series.
2) DNIC was assessed using two different test stimulus conditions during two different types of CS. First, changes in measures of THR and TOL during a slow-ramp heat TS (Edwards et al., 2003b; Sigurdsson & Maixner, 1994) were acquired with CS at 12^0 C, and again using 32^0 C circulating water. Second, DNIC effects were acquired during

phasic noxious heat stimuli for APR and MPR without counter-irritation to these same measures during CS at 12^0 C, and again using 32^0 C circulating water (Staud et al., 2001).

3) The DNIC effect on Temporal summation was assessed by comparing a) the difference in RR without counter-irritation to the RR measure during counter-irritation (using both 12^0C and 32^0 C as a conditioning stimulus), and b) by comparing the difference in Delta without counter-irritation to the Delta measure during counter-irritation (using both 12^0 C and 32^0C as a conditioning stimulus).

Heat pain threshold and tolerance temperatures were assessed at three locations (then averaged) on the glabrous surface of the left forearm for all subjects. Temporal summation and DNIC were assessed at two positions, which were counter-balanced between subjects on the palmar surface of the left hand at two different temperatures, one being established just above the individual tolerance temperatures acquired from the previous test and one set at 50^0 C for all subjects. DNIC was assessed using the slow-ramp test stimulus, as well as the phasic heat pain stimuli at both thermode temperatures using two different conditioning water temperatures, which were counter-balanced between subjects, one being held constant at a noxious temperature (12^0 C) and one held constant at a neutral temperature (32^0 C) to control for non-specific effects. Finally, the effect of DNIC on temporal summation was assessed at both thermode temperatures and using both water temperatures. The subject's participation included a total of 18 minutes of actual testing, alternating with 36 minutes of rest periods, in addition to 45-60 minutes to complete a review of the informed consent form and questionnaires. All subjects completed all of the pain testing protocols and there were no adverse events.

Pain Testing Protocols

Pain Threshold and Pain Tolerance

Session B1: Heat pain thresholds (THR) were obtained using a controlled thermal device held in continuous contact with the skin of the glabrous surface of the left forearm in a slow-ramp protocol for each subject (Medoc Medical Instruments Inc.). The thermal stimuli were delivered using a thermode (30 by 30-mm) contactor. Heat stimuli began at 38^0C and increased in 0.5^0 C/second increments in an ascending method of limits paradigm. Inter-trial thermode temperature returned to 38^0C. To ensure participant safety, a maximum temperature of 51°C was used, which is 2°C below the maximum that is recommended to avoid tissue damage by the manufacturer, as well as previous investigators (Edwards et al., 2003a). Subjects rated the intensity of the thermal stimuli by clicking a mouse to establish pain thresholds. Subjects were told that they may discontinue the heat stimuli at any time by clicking on the mouse, which immediately discontinued the thermal stimuli, or by verbally requesting the session be stopped. Three trials, one centimeter apart, moving distally beginning one cm from the break in the elbow, and separated by 10 second rests, were performed and averaged for threshold (maximum duration of 30 seconds each). Blood pressure and heart rate were acquired immediately following the end of sensory testing. Differences in pain threshold were compared between groups.

Session B2: Following a three-minute rest, heat pain tolerance (TOL) were obtained in the same manner as THR temperatures except that subjects were instructed to identify the maximum heat pain that the were able or willing to tolerate by clicking the mouse which discontinued the heat stimuli (maximum duration of one minute each).

Temporal Summation

Temporal summation was assessed using two different thermode temperatures.

Session W1: Following a three-minute rest, the first temporal summation protocol was performed. During this procedure, the peak temperature of the thermode remained constant, at the mean pain tolerance temperature (rounded up to the nearest whole number) established in Session B2 for the duration of the temporal summation protocol. Baseline temperatures were 10^0C below individualized peak temperatures. Each heat pulse was delivered at a rate of one pulse every three seconds to ensure temporal summation (maximum 8 trials). Subjects rated the intensity of the pain from the thermal stimuli on a 0 to 100 scale, with 0 representing no pain, and **1OO** representing the most intense pain imaginable. Subjects were informed that the procedure would be terminated if they give a rating of 100 or verbally request that the stimuli be discontinued. Heart rate and blood pressure were reacquired immediately following the end of sensory testing. Differences in the RR (slope) from the first six pain ratings and in Delta (subtracting the first pain rating from highest pain rating) were compared between groups as measures of temporal summation.

Session W2: Following a three-minute rest, a second temporal summation protocol was performed at a different site on the palm (order was also counter-balanced between subjects). During this procedure, the temperature of the thermode remained at 50^0 C for each pulse peak for the duration of the temporal summation protocol. As in session W1, subjects rated the intensity of the pain from the thermal stimuli on a 0 to 100 scale, and subjects were informed that the procedure would be terminated if they give a rating of 100 or verbally request that the stimuli be discontinued. Heart rate and blood pressure were acquired immediately following the end of sensory testing. Differences in the RR and in Delta were compared between groups as measures of temporal summation.

Diffuse Noxious Inhibitory Controls

DNIC effects were assessed in two manners: 1) using TS in a slow ramp ascending method of limits for THR and TOL using 12^0 C (noxious) and again using 32^0C (non-noxious) CS (Sessions D1 - D4), and 2) using phasic TS at two different, but constant temperatures (individualized to TOL temperatures from session B2, and 50C) using the same two CS above (12^0C and 32^0 C) in separate tests (Sessions WD1 - WD4).

Session D1: Following a three-minute rest, heat pain THR was obtained using the slow ramp administration of the test stimulus, as in sessions B1 (above), respectively, while a tonic conditioning stimulus was applied to the right hand. The conditioning stimulus for assessing DNIC was ice water submersion (12^0 C) of the right hand up to the wrist, in a circulating bath. Blood pressure and heart rate was acquired prior to, and immediately following the end of sensory testing. Changes in THR (temperatures) were compared between groups.

Session D2: Following a three-minute rest, heat pain TOL was obtained using the slow ramp administration of the test stimulus, as in sessions B2 (above), respectively, while a tonic conditioning stimulus was applied to the right hand. The conditioning stimulus for assessing DNIC was ice water submersion (12^0 C) of the right hand up to the wrist, in a circulating bath. Blood pressure and heart rate was acquired prior to, and immediately following the end of sensory testing. Changes in TOL (temperatures) were compared between groups.

Session D3: Following a three-minute rest, the protocol for D1 was duplicated except that the temperature of the water used for the conditioning stimulus was a neutral, non-noxious temperature (32^0 C) to control for non-specific effects. Changes in THR (temperatures) were compared between groups.

Session D4: Following a three-minute rest, the protocol for D2 was duplicated except that the temperature of the water used for the conditioning stimulus was a neutral, non-noxious

temperature (32^0 C) to control for non-specific effects. Changes in TOL (temperatures) were compared between groups. Subjects were instructed to focus their attention of the slow-ramp heat pain test stimulus through the DNIC protocol and not on the conditioning stimulus.

To prevent order effects, the DNIC protocols using the two water temperatures as CS counter-irritation were counter-balanced. In addition, the position of the thermode for the tolerance temperature and 50^0 C thermode temperature was also counter-balanced at the base of the thumb and the heel of the hand for all phasic pain testing protocols.

Session WD1: Following a three-minute rest, heat pulses were administered at the individualized temperature used in session W1, while a tonic conditioning stimulus was applied to the right hand. The conditioning stimulus for assessing DNIC was ice water submersion (12^0 C) of the right hand up to the wrist, in a circulating bath. Phasic heat pain testing commenced after the subject's right hand was submerged in ice water for 20 seconds. The tonic noxious stimulation was delivered continuously to maintain a moderate level of pain during the assessment of phasic heat pain ratings. The duration of the DNIC protocols were less than 1 minute (24 seconds of phasic heat testing and 20 seconds of the conditioning stimulus prior to the initiation of the test stimulus). Subjects were instructed that they may stop the tonic cold pain or phasic heat stimuli at any time if the discomfort is greater than they wish to endure. Blood pressure and heart rate were acquired immediately following the DNIC testing. Changes in APR and MPR from baseline (session W1) to testing during counter-irritation (session WD1) were compared between groups.

Session WD2: Following a three-minute rest, heat pulses were administered at peaks of 50^0 C for all subjects, while a tonic conditioning stimulus was applied to the right hand. The conditioning stimulus for assessing DNIC was ice water submersion (12^0 C) of the right hand up to the wrist, in a circulating bath. Phasic heat pain testing commenced after the subject's

right hand was submerged in ice water for 20 seconds. The tonic noxious stimulation was delivered continuously to maintain a moderate level of pain during the assessment of phasic heat pain ratings. The duration was less than 1 minute of phasic heat testing, as in session WD1. Subjects were instructed that they may stop the tonic pain or phasic heat stimuli at any time if the discomfort is greater than they wish to endure. Blood pressure and heart rate were acquired immediately following the DNIC testing. Changes in APR and MPR from baseline (session W2) to testing during counter-irritation (session WD2) were compared between groups.

Session WD3: Following a three-minute rest, the protocol for WD1 (thermode at tolerance temperatures) was duplicated except that the temperature of the water used for the conditioning stimulus was a neutral, non-noxious temperature (32^0 C) to control for non-specific effects. Changes in APR and MPR from baseline (session W1) to testing during counter-irritation (session WD3) were compared between groups.

Session WD4: Following a three-minute rest, the protocol for WD2 (thermode at 50^0 C) was duplicated except that the temperature of the water used for the conditioning stimulus was a neutral, non-noxious temperature (32^0 C) to control for non-specific effects. Changes in APR and MPR from baseline (session W2) to testing during counter-irritation (session WD4) were compared between groups.

To prevent order effects, the DNIC protocols using the two water temperatures as CS counter-irritation were counter-balanced. In addition, the position of the thermode for the tolerance temperature and 50^0 C thermode temperature was also counter-balanced at the base of the thumb and the heel of the hand for all phasic pain testing protocols. Subjects were instructed to focus their attention of the phasic heat pain test stimulus through the DNIC protocol and not on the conditioning stimulus.

The Effect of Diffuse Noxious Inhibitory Controls on Temporal Summation

Changes in temporal summation measures, RR and Delta, during sessions W1 and WD1 (thermode at individualized tolerance temperatures, conditioning stimulus at 12^0 C) were assessed as measures of the effect of DNIC on temporal summation. In addition, changes in RR and Delta during sessions W2 and WD2 (thermode at 50^0 C, conditioning stimulus at 12^0 C) were assessed as measures of the effect of DNIC on temporal summation.

To control for non-specific effects these same measures of temporal summation (RR and Delta) were assessed comparing W1 and WD3 (thermode at individualized tolerance temperatures, conditioning stimulus at 32^0 C) as well as comparing RR and Delta during W2 and WD4 (thermode at 50^0 C, conditioning stimulus at 32^0 C) sessions. Again, the order of the conditioning stimulus protocols was counter-balanced between subjects and subjects were instructed to focus their attention of the phasic heat pain test stimulus through the DNIC protocol and not on the conditioning stimulus.

Data Analysis

Significant differences between patient and control groups in endogenous pain mechanism measures have been demonstrated in comparable investigations of TMD (Kashima et al., 1999; Maixner et al., 1997; Sigurdsson & Maixner, 1994), and FMS (Staud et al., 2001; Lautenbacher & Rollman, 1997; Kosek & Ordeberg, 2000b) using small to moderate total sample sizes ranging from 10 to 51 (mean sample size = 31). Thus, the sample size for this investigation was more than adequate to demonstrate significant group effects.

The first step in the data analyses was to calculate descriptive statistics and measures of the distribution of all the variables. Groups were compared by analysis of variance (ANOVA) with respect to all demographic variables. If there were significant differences,

these variables became covariates in subsequent analyses. Alpha was set at $p<.05$ for all analyses (SPSS, 15.0).

Primary Outcome Measures of Central Pain Modulation

Temporal Summation

ANOVA was used to compare group differences in temporal summation measures (RR and Delta) at both thermode temperature settings (individualized thermode temperature and 50^0 C).

Diffuse Noxious Inhibitory Controls

Separate Repeated-measures analysis of variance (ANOVA) were performed with Group as a between-subjects factor and Time (protocols including the conditioning stimulus vs. those without) for each DNIC measure: changes in THR, TOL, and changes in APR and MPR for both thermode temperature conditions (individualized thermode temperature and 50^0 C) during both counter-irritation conditions (12^0 C and 32^0 C water submersion of the other hand).

However, in order to control for non-specific effects, such as distraction, which is known to play a major role in DNIC effects, a Repeated-measures ANOVA was performed entering the APR for the non-noxious and for the noxious conditioning stimuli protocol for Time. This is precisely the same as comparing the difference in the DNIC effect during 12^0 C to the DNIC effect during 32^0 C CS. According to Edwards and colleagues, (2003b),

> "This gives a controlled (i.e. effects of the neutral water temperature session are subtracted) measure of the degree to which thermal pain ratings change, specifically as a result of heterotopic cold pain and provides measures of the decrease in average heat pain ratings during 12^0 C relative to 32^0 C CS."

This analysis was performed on the DNIC measures to determine group differences for APR, MPR, THR, and TOL measures.

Diffuse Noxious Inhibitory Controls Effect on Temporal Summation

Repeated-measures analysis of variance (ANOVA) were performed for changes in each measures of temporal summation (RR and Delta) for both thermode temperature conditions (individualized thermode temperature and 50^0 C) and both CS (12^0 C and 32^0 C).

Secondary Outcome Measures

ANOVA was used to compare group differences for all General Pain Sensitivity Measures (APR, MPR, THR, and TOL). In addition, ANOVA was used to compare group differences for all psychological measures (anxiety, depression, anger expression, perceived stress, and catastrophizing) and sympathetic nervous system outcome measures. ANOVAs were used to compare group changes in sympathetic nervous system activity (systolic blood pressure and heart rate). Significant differences between IBS and control subjects in sympathetic nervous system activity and psychological measures were used as covariates in ANCOVAs for secondary explanatory analyses.

Chapter 3

Results

Descriptive Information

Forty eight subjects were recruited for this investigation. This included 21 healthy controls (HC) and 27 patients with IBS. Five IBS patients also had another chronic pain condition (3 with migraines, 2 with TMD). There were no significant differences ($p > 0.05$) between the IBS and HC groups on any demographic variables. Eighty-five % (IBS) and 81% (HC) of subjects were college graduates. Forty-eight % (IBS) and 29% (HC) of those were graduate students. Groups were age matched (HC = 28.5 years and IBS = 28.9 years). The groups mean Body Mass Index was also virtually the same (HC = 23.9 and IBS = 24.1). IBS patients included 25 Caucasian, one African American, and one Asian. Healthy controls included 14 Caucasians, 3 African American, one Asian, two Native Americans, and one Sudanese.

IBS subjects reported a mean symptom severity score of 251.9 on the Irritable Bowel Severity Scale (Francis et al., 1997). This represents a moderate severity of IBS symptoms. This included five subjects who rated the symptoms as mild, thirteen as moderate and nine as severe.

Secondary Outcome Measures

Psychological Outcome Measures

ANOVA demonstrated that IBS patients reported significantly greater stress than did HC (Table 4.1) as reflected in scores on the state anxiety index of the Spielberger State Trait Anxiety Index ($p = 0.008$), the anger-out subscale on the Anger expression Inventory ($p =$

0.039), and the Catastrophizing subscale of the CSQ ($p = 0.005$) and the Beck Depression Inventory ($p = 0.026$).

After controlling for psychological measures of IBS subjects having other chronic pain conditions, group differences remained significant for the state anxiety measure ($p = 0.006$), the catastrophizing coping strategy ($p = 0.01$) and demonstrated a strong trend for the anger-out measure ($p = 0.058$).

Sympathetic Nervous System Outcome Measures

There were no group differences ($p > 0.05$) on any measures of sympathetic nervous system activity (systolic blood pressure and heart rate) either at baseline or after any of the pain testing protocols. In an addition there were no group differences ($p > 0.05$) in changes in systolic blood pressure and heart rate after any pain test.

Questionnaires	Controls	IBS
State anxiety**	25.6 (1.6)	32.2 (1.7)
Trait anxiety	29.4 (1.6)	33.6 (2.0)
Depression*	1.9 (1.1)	4.8 (.9)
Anger expression	42.9 (.75)	45.0 (.7)
Anger-in expression	26.4 (.56)	25.3 (.57)
Anger-out expression*	11.9 (.64)	13.6 (1.0)
Perceived stress	16.9 (1.5)	21.5 (1.8)
Catastrophizing**	1.4 (1.8)	2.0 (.17)

Note: *$p < 0.05$, ** $p < 0.01$.
Table 4.1 - Group mean (SEM) scores for all psychological questionnaires

General Pain Sensitivity Measures

There were no group differences ($p > 0.05$) in any of the general pain sensitivity outcome measures for either thermode temperature, during baseline, or during the CS protocols in APR or MPR (Table 4.2). However, there were significant group differences for all of the DNIC measures (changes in APR and MPR from baseline to during the CS protocol) using noxious water temperature (12^0 C) as the conditioning stimulus and the 50^0 C thermode temperature as the test stimulus, but not when the water temperature was non-noxious (32^0 C), or when the thermode temperature was less noxious (mean temperature 46^0C), based on individualized TOL temperatures from earlier testing. In addition, when significant group differences found in the psychological measures; state anxiety on the Spielberger State Trait Anxiety Index, the Beck Depression Inventory, Anger-out subscale of the Anger Expression Inventory, and the Catastrophizing subscale of the Coping Strategies Questionnaire, were entered as covariates in separate ANCOVAs for APR and MPR DNIC effects, only the Catastrophizing subscale of the CSQ had any effect on group differences, and only for one of the endogenous pain mechanism outcome measures (MPR, $p = 0.071$).

Protocols	Controls	IBS
Threshold	39.4 (.65)	39.6 (.69)
Tolerance	45.9 (.63)	45.5 (.64)
Maximum Pain Rating#	39.9 (4.7)	42.2 (4.3)
Maximum Pain Rating^	55.5 (5.7)	54.3 (5.4)
Average Pain Rating#	31.5 (4.1)	36.2 (3.9)
Average Pain Rating^	47.6 (5.3)	47.5 (4.9)
Water Pain Rating@	67.1 (4.9)	68.1 (5.2)
Water Pain Rating$	58.6 (4.9)	64.6 (5.2)
Water Pain Rating#	58.3 (4.3)	58.9 (4.9)
Water Pain Rating^	55.5 (4.8)	57.4 (5.2)

Note: None of the group comparisons for means of the General Pain Sensitivity Measures were significant. All Water Pain Ratings were during 12^0 Celsius conditioning stimuli. # = individualized thermode temperatures, ^ = thermode at 50^0 Celsius. @ = pain threshold test, $ = pain tolerance test.

Table 4.2 Group means (SEM) for all general pain sensitivity outcome measures.

There were no group differences ($p > 0.05$) on any of the general measures of heat pain including: pain threshold or pain tolerance temperatures, APR, or MPR. Nor were there any group differences ($p > 0.05$) in pain ratings due to water submersion of the right hand (mean H2O pain ratings, HC= 56.5, IBS = 57.5).

All APR, MPR, and H2O general pain sensitivity ratings were significantly correlated with each other ($p < 0.01$) and with THR and TOL scores ($p < 0.05$). All measures of DNIC are also significantly correlated with each other. However, none of the DNIC measures were associated with any of the general pain sensitivity measures.

Primary Outcome Measures of Central Pain Modulation

Temporal Summation

Rate of Rise – No significant group differences ($p > 0.05$) were found in temporal summation (Table 4.3) for either thermode temperature setting (50^0 C or individualized temperatures). Using individualized thermode temperature setting, there was a trend (ANOVA, $p = 0.051$) showing IBS subjects to have a steeper increase ($\beta = 0.68$) in pain ratings, however HC did not demonstrate any temporal summation during this protocol, but in fact, showed a reduction in pain ratings across stimuli ($\beta = -1.03$). Slopes for the 50^0 C thermode setting were $\beta = 1.68$ for the IBS group and $\beta = 1.06$ for HC (ANOVA, $p = 0.41$).

Delta – Similarly, no group differences ($p > 0.05$) were seen for either thermode temperature setting protocols when comparing increases in pain ratings from the initial stimuli to the highest pain rating in the train of phasic stimuli.

Protocols	Controls	IBS
Rate of Rise#	-1.03 (.71)	.68 (.52)
Rate of Rise^	1.06 (.6)	1.68 (.47)
Delta#	7.5 (1.8)	9.5 (2.1)
Delta^	13.7 (2.4)	13.7 (2.5)

Note: None of the group comparisons for a Temporal Summation Rate of Rise (slopes), Delta values, or the DNIC effect (values not shown) on Rate of Rise or Delta were significant.
= individualized thermode temperatures, ^ = thermode at 50^0 Celsius.
Table 4.3 - Group means (SEM) for Temporal Summation measures.

Diffuse Noxious Inhibitory Controls Effect on Temporal Summation – Using Repeated-measures ANOVA, no significant group differences ($p > 0.05$) were found in the DNIC effect for either of the temporal summation measures (RR and Delta) for either thermode temperature setting (50^0 C or individualized temperatures).

Diffuse Noxious Inhibitory Controls

Average Pain Rating - With the phasic peak thermode temperature test stimulus at 50^0 C for all subjects and the conditioning stimulus of 12^0 C water, a Repeated Measures ANOVA demonstrated that there was a significant main effect of Time (indicating a DNIC effect across all subjects) [$F = 30.4$ (1,46), $p < 0.001$, $\eta^2 = .4$. There was also a significant interaction effect (Figure 4.1) between Group X Time [phasic pain rating (APR) with and without the conditioning stimulus], indicating a deficit in the DNIC effect for IBS subjects [$F = 6.97$ (1, 46), $p = 0.011$, $\eta^2 = 0.13$]**.**

This interaction effect remained significant after controlling for changes in APR pain ratings from IBS patients who reporting having a second chronic pain condition [$F = 4.85$ (1, 41) ($p = 0.033$). None of the psychological measures that demonstrated significant group differences were associated with APR scores or changes in APR scores during DNIC testing.

Repeated-measures ANOVA found non-significant interaction effects ($p > 0.05$) in APR for the DNIC protocol using the individualized thermode temperature (mean thermode temperature for control group = 46.3^0 C and mean thermode temperature for the IBS group = 46.0^0 C) while using 12^0 C circulating water as the conditioning stimulus ($p = 0.35$). There were also non-significant interaction effects for both protocols (test stimulus with the thermode at 50^0 C and at individualized thermode temperatures) using water temperature at 32^0 C for the CS ($p = 0.6$ and $p = 0.8$, respectively). See Figure 4.2 for a comparison of DNIC effects during both CS temperatures with thermode at 50^0 C. Group differences in APR were

consistently larger for the control group in all but one instance (where they were equal). However, this does not reflect a DNIC effect, in that a non-noxious CS does not recruit DNIC mechanisms. This reduction in pain ratings during non-noxious conditioning stimuli reflects non-specific effects, including distraction.

After controlling for non-specific effects, the interaction effect demonstrated significantly greater DNIC effects (Figure 4.3) for HC on reduction in APR scores compared to IBS subjects [$F = 11.04$ (1, 46) $p = 0.002$) $\eta^2 = 0.19$. Group differences in the DNIC effect score, based on percent reductions in APR scores were the same ($p = 0.002$). This provides a measure of the decrease in average heat pain ratings during 12^0 C relative to 32^0 C CS. Mean pain ratings for all water emersion protocols were 61.2 (3.4) for 12^0 C, and less than 1.0 (0.46) for 32^0 C water emersion. There were no differences between groups for either water temperature pain rating.

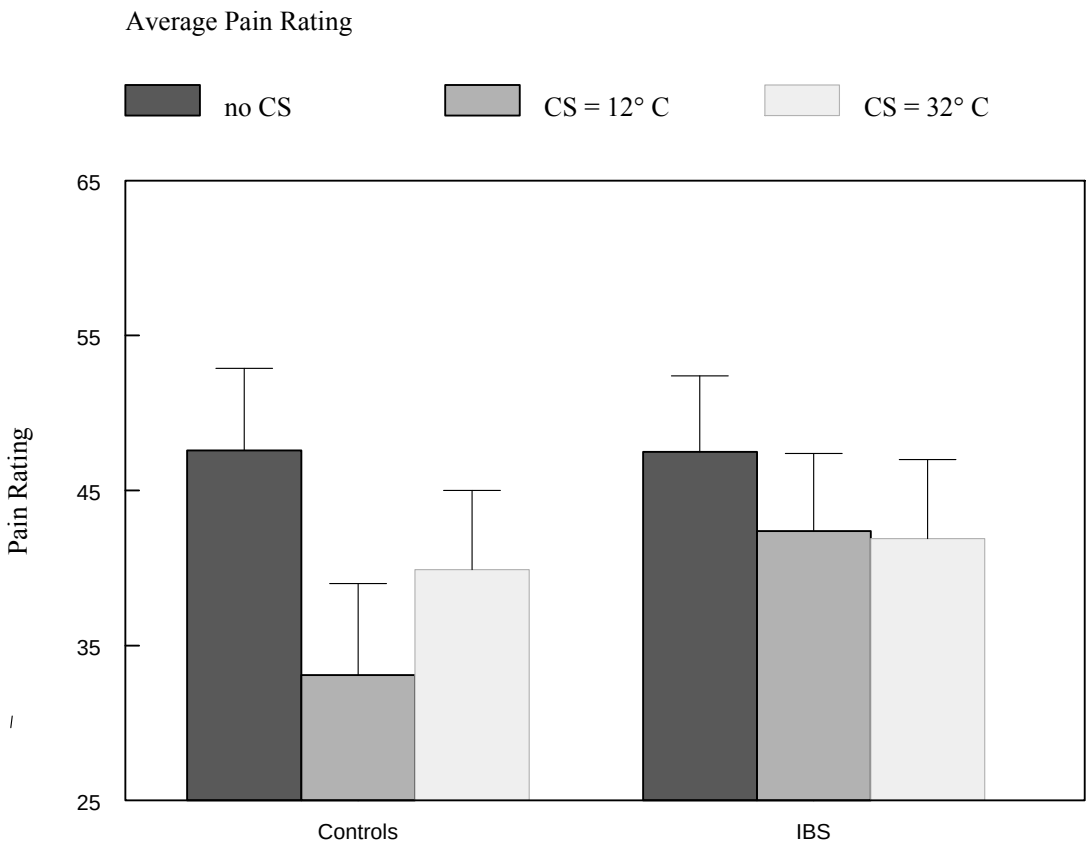

Figure 4.1 Average Pain Ratings of the phasic test stimulus (thermode at 50^0 C). Reductions in pain ratings (DNIC effect) from no CS to Average Pain Ratings during the tonic CS at 12^0 C, demonstrates compromised DNIC in IBS subjects compared to controls ($p = 0.011$). TS = test stimuli, CS = conditioning stimuli, IBS = Irritable Bowel Syndrome, C = Celsius.

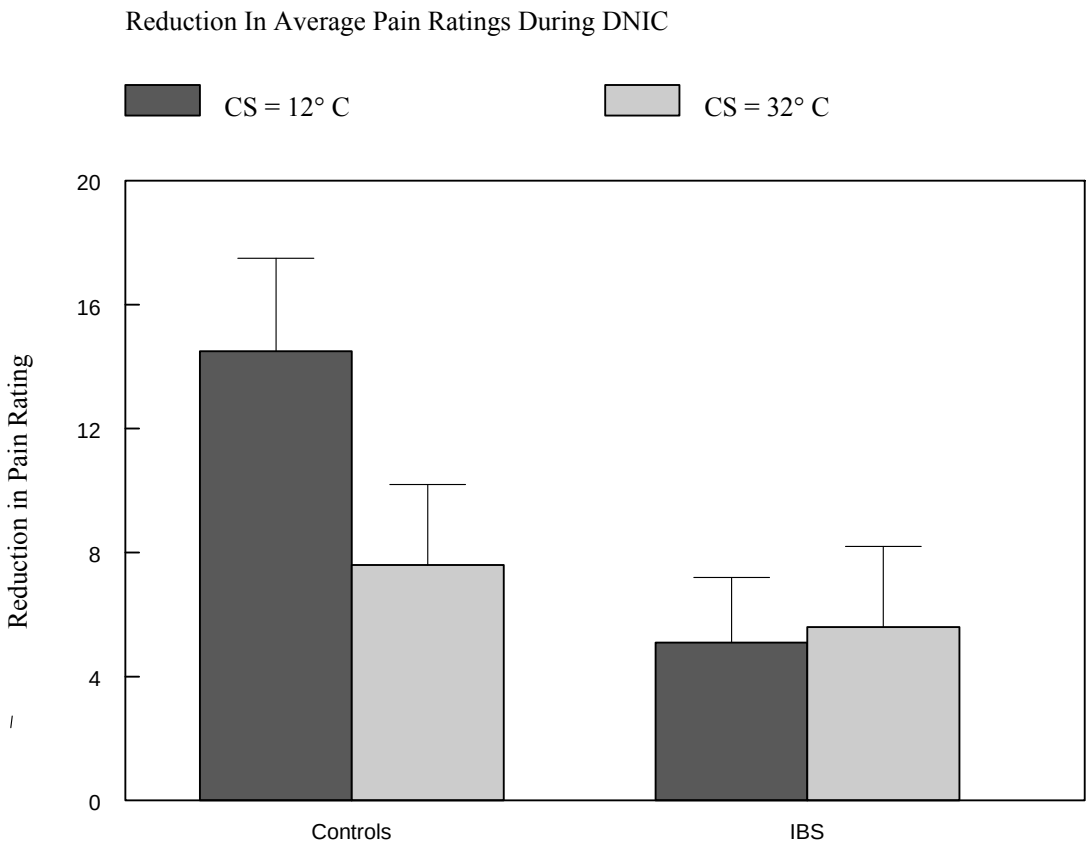

Figure 4.2 Reductions in Average pain Ratings of the phasic test stimulus (DNIC effect) during conditioning stimuli. Significant group differences in DNIC occurred only during 12^0 C conditioning stimuli, with the thermode at set 50^0 C (p = 0.011). C = Celsius

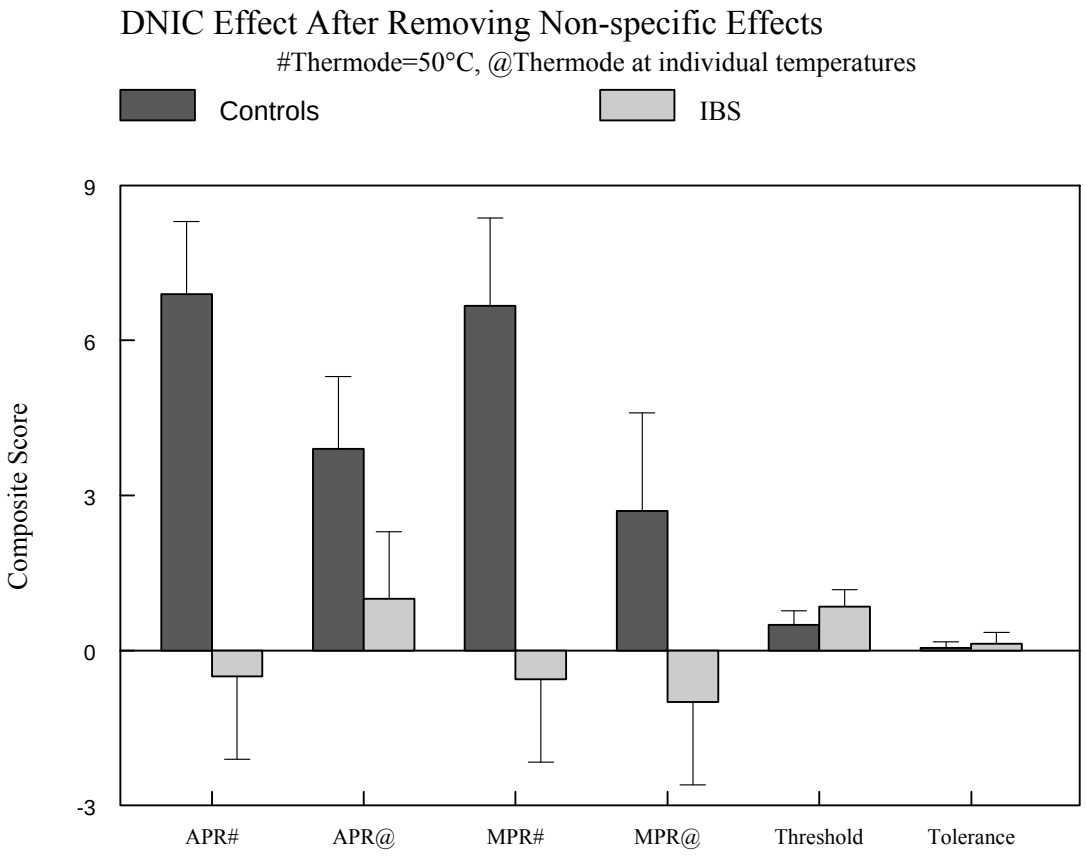

Figure 4.3 Group differences in the DNIC effect during noxious conditioning stimuli after removing non-specific effects from "counter-irritation" during neutral conditioning stimuli. APR = Average Pain rating, MPR = Maximum Pain Rating, IBS = Irritable Bowel Syndrome ($p < 0.01$ ANOVA).

Maximum Pain Rating - With the phasic peak thermode temperature test stimulus at 50^0 C for all subjects and the conditioning stimulus of 12^0 C circulating water, a Repeated Measures ANOVA demonstrated that there was a significant main effect of Time, indicating a DNIC effect across all subjects [$F = 35.8$ (1, 46), $p < 0.001$, $\eta^2 = .44$]. In addition, there was also a significant interaction effect (Figure 4.4) between Group X Time [phasic pain rating (MPR) with and without the conditioning stimulus], indicating a deficit in the DNIC effect for IBS subjects [$F = 7.4$ (1, 46), $p = 0.009$, $\eta^2 = 0.14$].

This effect remained significant after controlling for MPR from IBS patients who reported having a second chronic pain condition [$F = 5.0$ (1, 41) ($p = 0.032$). The Catastrophizing subscale of the Coping Strategies Questionnaire and the State subscale of the Spielberger State Trait Anxiety Inventory were negatively associated with the DNIC effect for MPR (r = - 0.37, $p = 0.01$ and r = - 0.37, $p = 0.01$, respectively). After including these psychological variables scores as covariates in a Repeated-measures ANCOVA, group differences in MPR demonstrated a trend in favor of the control group ($F = 2.24$ (1, 44), $p = 0.14$, $\eta^2 = 0.05$]. As scores on the Catastrophizing subscale of the Coping Strategies Questionnaire increased, the DNIC effect, as reflected by reductions in MPR during the 12^0 C circulating water CS, were reduced. In addition to the significant negative association with catastrophizing, DNIC effects on MPR were negatively correlated with IBS symptom severity (r = - 0.31, $p = 0.019$), and with severity of co-morbid conditions (r = - 0.31, $p = 0.03$, which suggests that elevations in catastrophizing and symptom severity may contribute to the DNIC effect for MPR, but not for APR.

Repeated-measures ANOCOVA found non-significant interaction effects ($p > 0.05$) in changes in MPR for the DNIC protocol using the individualized thermode temperature (mean thermode temperature for control group = 46.3^0 C and mean thermode temperature for the IBS group = 46.0^0 C) using 12^0 C circulating water as the conditioning stimulus ($p = 0.26$). There were also non-significant interaction effects for both protocols (test stimulus with the thermode at 50^0 C and at individualized thermode temperatures) using water temperature at 32^0 C for the CS. See Figure 4.5 for a comparison of DNIC effects during both CS temperatures with thermode at 50^0 C. Group differences in MPR tended to be larger for the control group in all but two instances (where they were equal). However, this does not indicate a DNIC effect, in that a non-noxious CS does not recruit DNIC mechanisms.

After controlling for non-specific effects (see Figure 4.3), the interaction effect continued to demonstrated significantly greater DNIC effects on reduction in MPR scores for HC compared to IBS subjects [$F = 9.45$ (1, 46) $p = 0.003$, $\eta^2 = 0.19$]. Group differences in the DNIC effect based on percent reductions in MPR scores were the same ($p = 0.003$). This provides a measure of the decrease in maximum heat pain ratings during 12^0 C relative to 32^0 C CS.

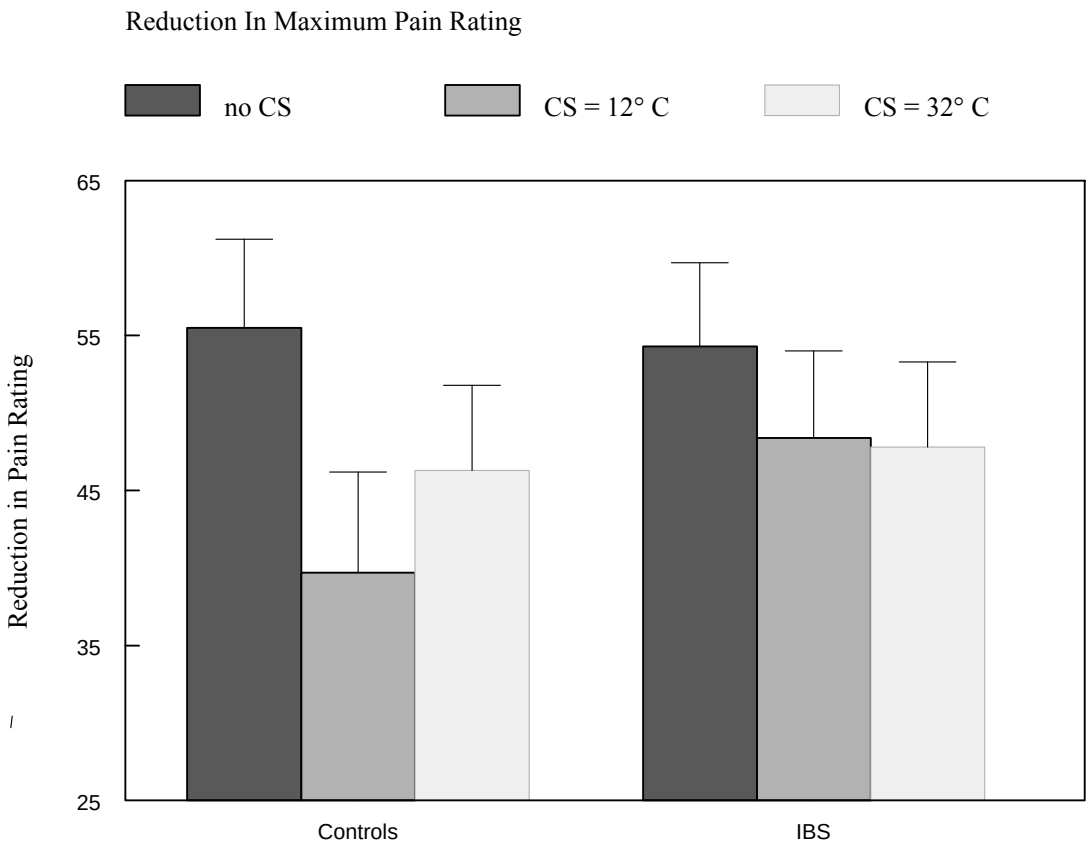

Figure 4.4 Maximum Pain Ratings of the phasic test stimulus (thermode at 50^0 C). Reductions in pain ratings (DNIC effect) from no CS to Maximum Pain Ratings during the tonic CS at 12^0 C, demonstrates compromised DNIC in IBS subjects compared to controls ($p = 0.009$). TS = test stimuli, CS = conditioning stimuli, IBS = Irritable Bowel Syndrome, C = Celsius.

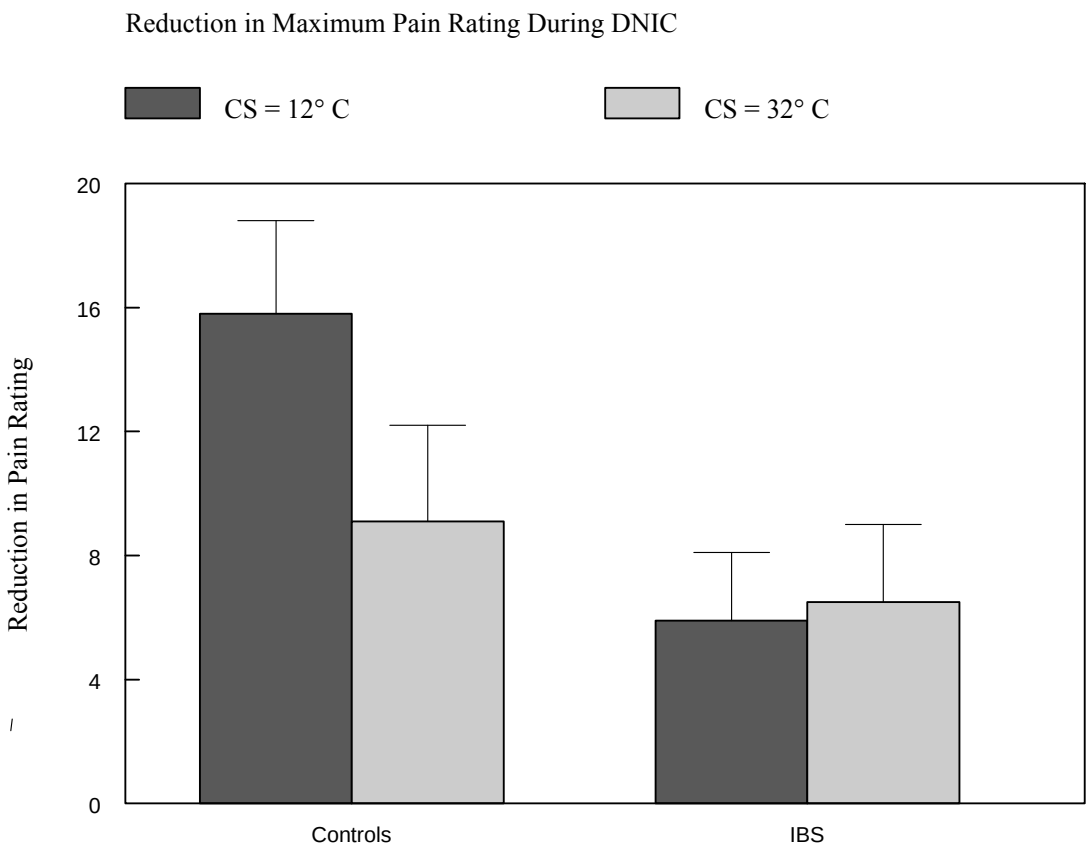

Figure 4.5 Reductions in Maximum Pain Ratings of the phasic test stimulus (DNIC effect) during conditioning stimuli. Significant group differences in DNIC occurred only during 12^0 C conditioning stimuli, with the thermode at set 50^0 C (p = 0.009).

Threshold

Although there were no group differences in the DNIC effect for pain Tolerance temperatures, a Repeated-measures ANOVA demonstrated that there was a significant main effect of Time (indicating a DNIC effect across all subjects) [F = 182.6 (1, 46), p < 0.001] during the 12^0 C conditioning stimulus (Figure 4.6) for Threshold. In addition, there was also a significant interaction effect) between Group X Time with and without the conditioning [F = 5.34 (1, 46), p = 0.025]. In addition, similar effects were seen for Threshold during the 32^0 CS with significant group differences [F = 12.34 (1, 46), p = 0.001]. See Figure 4.7 for a comparison of DNIC effects during both CS temperatures. These effects remained significant after controlling for Threshold temperatures from IBS patients who reporting having a second chronic pain condition [F = 5.0 (1, 41) (p = 0.032).

After controlling for non-specific effects (see Figure 4.3), there were virtually no changes in Threshold temperatures (1 to 2 percent) and no group differences. There were no interaction effects in the DNIC effect between groups for changes in TOL temperatures during either conditioning stimulus temperature. There were also no DNIC effects (less than 1 percent) for TOL after controlling for non-specific effects.

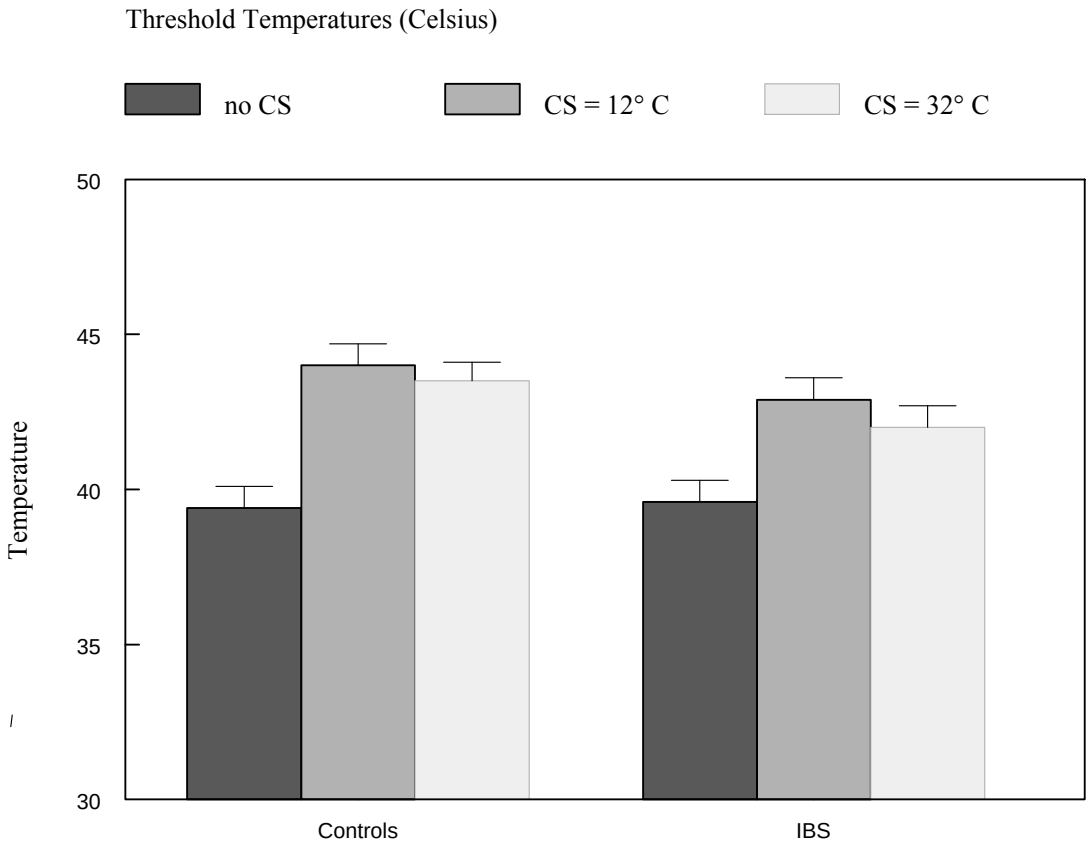

Figure 4.6 Threshold temperatures (slow-ramp ascending method of limits). Significant group differences in increased pain threshold temperatures occurred during both noxious and non-noxious conditioning stimuli ($p = 0.025$, and $p = 0.001$, respectively).

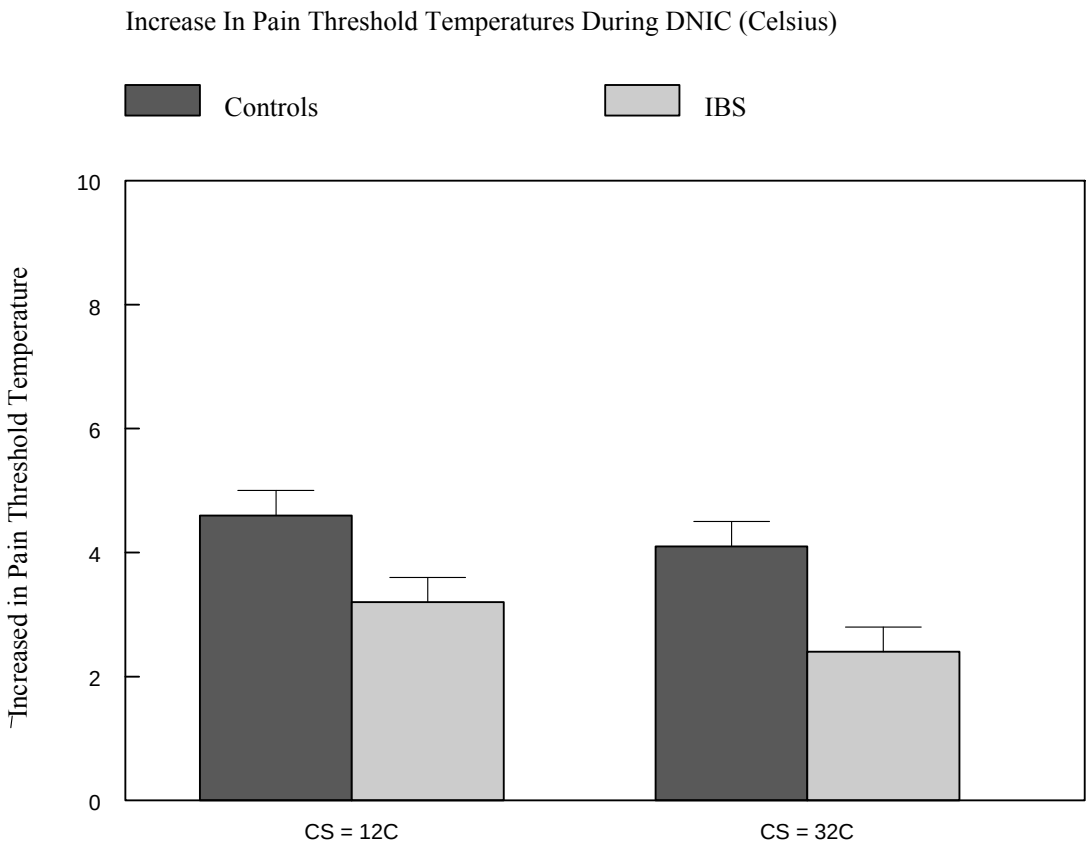

Figure 4.7 Increases in Pain Threshold temperatures (slow-ramp ascending method of limits) during conditioning stimuli (DNIC effect). Significant group differences in DNIC occurred during both noxious and non-noxious conditioning stimuli ($p = 0.025$, and $p = 0.001$, respectively).

Summary of Results

The primary outcome of the present investigation was that significant group differences in DNIC were found in APR, MPR and THR indicating deficits in DNIC for subjects with IBS compared to controls. These deficits remained unchanged after controlling for the effects of IBS subject who had a second chronic pain diagnosis, and after controlling for psychological factors that were found to be significantly different between groups. In addition, after controlling for non-specific effects, significant deficits in DNIC remained for the IBS group compared to controls on measures of changes in APR and MPR, but not THR.

There were significantly higher levels of psychological distress on measures on state anxiety, depression, anger-out expression style, and catastrophizing pain coping style in the IBS group compared to controls. After controlling for psychological measures of IBS subjects having other chronic pain conditions, group differences were unchanged for the state anxiety measure, depression, the catastrophizing coping strategy, and demonstrated a strong trend for the anger-out measure.

There were no group differences on any of the measures of temporal summation between groups. There also were no group differences on any of the measures of the DNIC effect on Temporal Summation. There were also no group differences on any of the general pain measures, or on measures of sympathetic nervous system arousal.

Chapter 4

Discussion

The results of the present investigation demonstrate deficits in endogenous analgesic mechanisms for patients with IBS. The recruitment of DNIC was significantly impaired in IBS subjects compared to controls, but unexpectedly, no group differences were observed in temporal summation or the DNIC effect on temporal summation. After controlling for IBS subjects diagnosed with a second chronic pain condition, the DNIC effects remained significantly compromised in IBS subjects. Significant group differences were also seen in anxiety, depression, anger expression style, and pain coping style. After controlling for psychological factors, which are known to be associated with both pain ratings, and DNIC, IBS subjects continued to show deficient DNIC. In addition, after controlling for non-specific effects on DNIC, IBS subjects continued to demonstrate dysregulation in DNIC compared to controls. This is an important innovation in attempting to isolate the physiological mechanisms of DNIC during psychophysical investigations.

Distraction

One of the strengths of this investigation was the design control of the effects of distraction on DNIC outcome measures. This was accomplished by including a protocol utilizing a non-painful conditioning stimulus in addition to a standard, painful conditioning stimulus protocol. This allowed for analysis of a controlled measure of the degree to which thermal pain ratings change, specifically as a result of heterotopic cold pain and excludes the non-

specific effects of the neutral water temperature session, which mirrored the noxious condition in every way except water temperature.

Painful conditioning stimuli are certainly a potential source of distraction (Plaghki et al., 1994), which has been shown to play a role in some studies on DNIC (Lautenbacher et al., 2002), but not in others (Pertovaara et al., 1982; Talbot et al., 1987). Riley and Levine (1988) showed that distraction from pain is most likely to occur when the conditioning stimulus and testing stimulus are very similar in perceptual quality (eg. heat vs. heat). The notion that perceptual similarity influences the degree of distraction may also explain why some types of clinical pain effect experimental pain procedures in some studies (Willer et al., 1990), but not others (Ekblom & Hansson, 1987; Sigurdsson & Maixner, 1994). It has been suggested that distraction may have contributed to the effects of earlier DNIC studies (Staud et al., 2003; Lautenbacher & Rollman, 1997). To address this important issue, Staud and colleagues (2003) designed an investigation to address the role of distraction in DNIC. Subjects were instructed to attend to either the painful TS or the painful CS (in counterbalance order) of the same quality (heat vs. heat) and then rate the pain from the other stimulus immediately after the end of the presentation of stimuli. They found that only female FMS subjects demonstrated a distraction effect beyond the DNIC effect found in these subjects. Although distraction does not completely explain DNIC (Reinert et al., 2000; Willer et al., 1989; Willer et al., 1999), Longe et al. (2001) demonstrated that attention has a major influence on modulation of pain. In this study, instructions to attend to non-painful vibratory CS significantly reduced pain ratings compared to attending to painful concurrent stimuli (Longe et al., 2001). Tracey (2004) suggested that brain regions that are involved in hypervigilance may connect to brainstem structures responsible for DNIC leading to dysregulation in DNIC. Further, Reinert et al. (2000) speculated that hypervigilance to TS

during DNIC protocols may disrupt DNIC, especially if the test stimulus is meaningful, such as rectal distension for IBS patients (Reinert et al., 2000), as seen in the Wilder-Smith IBS investigation of DNIC. The present investigation assessed the DNIC effect using somatic pain for both the test and conditioning stimuli, in part to avoid interference from peripheral sensitization that may be present in IBS patients. It is believed that by using somatic pain TS and CS, dysregulation in central pain mechanisms were isolated when compromised DNIC was identified in IBS patients.

Although there were no group differences during the non-painful conditioning protocol in the present investigation, there were substantial decreases in pain ratings shown by both IBS and control groups during 32^0 C conditioning stimuli. This reflects non-specific effects that have rarely been accounted for in previous investigations of DNIC. While several investigators have used similar counterbalanced conditioned stimuli protocols to compare DNIC effects in noxious and non-conditioning stimulus protocols (Sigurdsson & Maixner, 1994), only one other investigation has attempted to account for the role of non-specific effects in DNIC outcome measures (Edwards et al., 2003b). Edwards and colleagues (2003b) first identified the importance of isolating the DNIC effect from non-specific effects that are invariable part of all DNIC studies that utilize subjective reporting of pain ratings. Perhaps the most important findings in the present investigation are the robust group differences in DNIC that remained after controlling for distraction and psychological effects on pain ratings during counter-irritation protocols. Interestingly, this was true for protocols using phasic noxious test stimuli, but not when slow ramp heat was used as the test stimulus for changes in pain threshold. This may suggest that non-specific effects may have contributed to DNIC effects on pain threshold using slow-ramp TS that were reported in the majority of previous

studies on DNIC. It is recommended that future investigations examine the role of distraction and the degree to which it may contribute to abnormalities found in DNIC.

Lack of Association between Endogenous Analgesic Mechanisms and Simple Pain Measures

All of the simple pain measures were significantly correlated with each other and all of the DNIC measures were significantly correlated with each other, but none of the general pain measures were associated with any of the DNIC measures. Pielsticker et al. (2005) reported that chronic headache patients with deficient DNIC did not have lower pain thresholds, and concluded that the stimuli during THR testing was insufficient to recruit DNIC. Other investigators have failed to recruit a DNIC effect on pain threshold during counter-irritation as well (De Tomasso, 2003; Flor 2004). The present investigation found that non-painful conditioning stimuli led to reduced pain ratings. Lautenbacher and colleagues also found that non-noxious stimuli generated DNIC (Lautenbacher & Rollman, 1997; Lautenbacher et al., 2002). They suggest that a sufficient number of nociceptors may be fired to recruit DNIC even when conditioning stimulus was not describes as painful. Lautenbacher (Lautenbacher et al., 2002) theorized that because ascending pain signals travel different spinal tracts than the descending inhibitory signals, they are likely to have different activation thresholds. This may explain the lack of concurrence between experiencing pain and DNIC induction (De et al., 1990). Alternatively, these results may reflect distraction or other non-specific effects that were not accounted for as was done in the present investigation.

Edwards et al. (2003b) also found that the magnitude of DNIC was not associated with any other pain measures (THR, TOL, supra-threshold thermal pain ratings, or magnitude of temporal summation measures) and concluded that endogenous pain systems are subserved by different mechanisms than are these less complex pain responses. The authors

suggested that DNIC may be a more clinically relevant laboratory pain process than THR or TOL or simple pain rating scores, because it directly measures central nervous system pain regulatory mechanisms. In an attempt to identify what factors were associated with DNIC, Edwards and colleagues performed a hierarchical regression to parse out variables that were associated with DNIC variability (Edwards et al., 2003b). They found that DNIC predicted 17% of the bodily pain subscale scores on the Short-Form 36 quality of life scale, but found no association between DNIC measures and cardiovascular function or hypothalamus-pituitary-adrenal activity. They further found that DNIC was not mediated by psychological mechanisms, including: self efficacy, perceived stress, negative mood, positive mood, or greater stress reactivity. Edwards suggested that this may indicate a specific pain modulatory system for DNIC that is independent from other inhibitory systems and that enhancing pain modulatory ability may yield successful treatment for chronic pain sufferers (Edwards et al., 2003b). The results of the present study are in agreement with Edwards (2003b). While many studies have identified differences in psychological factors or autonomic dysregulation for IBS patients, the fact that these variables do not appear to be associated with abnormalities in endogenous analgesic mechanisms suggests that psychological and autonomic factors may simply be co-morbid conditions, or possibly a consequence of long-term suffering form chronic pain condition such as IBS and not a causative factor as has often been suggested.

Temporal Summation

There were no significant group differences in temporal summation in the present investigation. Similarly, there were no group differences in the DNIC effect on temporal summation. Given the consistent reports of exaggerated temporal summation in other chronic pain conditions, this was an unexpected finding. This may have been due to a general lack of wind-up being elicited in the protocols that were employed. Small slope and Delta values

indicated that very little, if any temporal summation occurred. Hence it is unlikely that a DNIC effect on temporal summation, or perhaps even group differences in temporal summation could be detected due to floor effects. Reasons for recruiting minimal temporal summation may be due to conservative parameters selected for noxious stimuli or may be due to limitations of the instrumentation settings. Two phasic pain protocols were used. One set peak thermode temperatures at 50^0 C, whereas other investigations demonstrating enhanced temporal summation have used 53^0 C. The second protocol used individualized thermode temperatures based on slow-ramp heat pain tolerance temperatures. Slopes for the individualized thermode temperature were less than 1.0 (controls had a negative slope) and less than 2.0 for the more noxious thermode temperature protocol (50^0 C). Again, floor effects may limit the conclusions that can be drawn from the investigations regarding temporal summation in IBS. Further research is needed to elucidate whether IBS patients suffer from exaggerated temporal summation. Making certain that the noxious stimulus is sufficiently painful will help too ensure that temporal summation is recruited. This is required to be able to detect group differences, if in fact they exist. In addition, new equipment is now available that can provide much faster thermal pulses such that wind-up can be more consistently recruited.

Psychological Factors

Controlling for catastrophizing in the present study did not alter the results showing IBS deficits in DNIC for Average Pain Ratings, but group differences only showed a trend for group differences in DNIC scores for Maximum Pain Ratings. In addition to catastrophizing, the DNIC effect for Maximum Pain Ratings was also negatively correlated with IBS symptom severity, but not with other psychological measures, or sympathetic nervous system activity scores. Pain coping styles, especially catastrophizing, shape pain response to a

significant degree (Edwards et al., 2006b; Buenaver et al., 2007). Edwards found catastrophizing to be associated with elevated levels of temporal summation (Edwards et al., 2006b) and suicidal ideation (Edwards et al., 2006a). Based on these results and those of the present study, the use of maximum pain scores may not be as reliable as average pain scores. It appears as though the tendency to catastrophize, seen in IBS patients, may artificially drive up maximum pain scores more so than average pain scores.

Although anger-out was not significantly associated with DNIC measures, IBS patients scored significantly higher than HC. In addition, there was no difference in anger-in scores between groups. Bruehl (2006a) and others (Kerns et al., 1994; Materazzo et al., 2000) have reported that scores on the Anger-Out Index were associated with elevated pain sensitivity and greater chronic pain intensity. In addition, Bruehl (2006b; 2006c) has shown that deficits in opioid analgesia in chronic pain patients, as well as in HC, were associated with anger-out expression, which suggests that these impairments are not the result of chronic pain conditions. Anger-in was not associated with opioid dysfunction (Bruehl et al., 2006b; Bruehl et al., 2006c). The authors suggested that anger-out expression may create increases in interpersonal stress, and the release of more opioids, which depletes the system, or may even create tolerance to endogenous opioids. More recently, Bruehl (2006c) and others (Burns et al., 2004), have shown that an anger out style of anger expression is associated with pain sensitivity and that this association depends on endogenous opioidergic anti-nociceptive dysfunction, whether indexed by circulating endogenous opioids or by opioid blockade. They further suggest that use of opioids medications may compensate for deficits in endogenous opioidergic analgesic systems. The present study found a combination of elevations in anger-out expression as well as a deficiency in endogenous analgesic systems in IBS subjects. This is consistent with the findings of Bruehl and the suggestions that IBS

patients suffer form dysregulation in serotonergic systems. Because DNIC effects are known to be largely serotonergic (Chitour et al., 1982), abnormalities in serotonin reuptake transporter polymorphisms seen in IBS suggests that failing DNIC may partially explain the success of selective serotonin reuptake inhibitor (SSRI) seen in the pharmaceutical treatment for some IBS patients. Selective serotonin reuptake inhibitors are known to improve motility, which may aid in pain reduction for IBS patients with constipation. It is widely acknowledged that improvement in symptoms offered by SSRIs may relate to improvement in mood. It is also believed that SSRIs offer direct analgesic effects, though this has not been proven. Conversely, a recent study determined that the SSRI, paroxitine, did not produce analgesic effects in comparison to maprotiline, a norepinephrine reuptake inhibitor. This may suggest an indirect mechanism of action for serotonin through it's effect of norepinephrine. Although antidepressants are still prescribed for patients with IBS, newer the serotonergic agents, tegaseraod and cilansetron, which are gut specific appear to be replacing traditional SSRI in the treatment of IBS. These agents are prescribed for constipation and diarhea predominant IBS, respectively, with the mechanism for pain relief being associated with changes in motility. Future investigations should identify associations between successful pharmaceutical treatment and the likely mechanism of deficient DNIC

Summary

The present investigation of endogenous analgesic mechanisms in IBS is the first study to extensively control for alternative explanations for the findings of compromised DNIC. In addition to controlling for hormonal and psychological factors by exclusion criteria, sympathetic arousal during pain testing and psychological factors found on questionnaires were statistically controlled for in secondary analyses. Most importantly, the design control for non-specific effects allowed for isolating the analgesic mechanisms from distraction and

other non-specific effects. Group differences in phasic DNIC measures remained significant even after controlling for this array of non-specific effects that are known to influence DNIC scores. Thus, the group differences in DNIC can be attributed to dysregulation in endogenous analgesic mechanisms in patients with IBS with confidence.

Wilder-Smith et al. (2004) previously demonstrated deficits in DNIC in IBS subjects using a visceral test stimulus and somatic conditioning stimulus (Wilder-Smith et al., 2004). Unfortunately, it can not be determined if the reduction in median pain scores for HC was due to distraction or whether the lack of DNIC in IBS subjects was due to hypervigilance to the test stimulus of rectal distensions, as suggested by Reinert et al. (2000) and Tracey et al. (2004). We have extended the findings of the Wilder-Smith study and improves on the design by controlling for non-specific effects in several important ways: 1) non-specific effects were controlled for by counter-balancing two CS protocols. One utilized a noxious conditioning stimulus (12^0 C water submersion of the hand), while the other provided the identical stimuli (circulating water) in a non-noxious form (32^0 C), 2) the effects of IBS patients with additional chronic pain syndromes were excluded, without affecting the results of the study, even with the reduction in power (this was not addressed in the Wilder-Smith study), 3) any significant group differences in psychological measures were controlled for by adding those measures as covariates in the relevant analyses.

There has been an increased interest in attempting to identify physiological mechanisms that contribute to functional chronic pain conditions, like FMS, TMD, and IBS. Recent studies have identified a post-infectious subgroup of IBS patients found to suffer form abnormalities in immunological function following an infection in the gastrointestinal tract that is believed to contribute to the development of IBS. There have also been several studies that have recently identified polymorphisms in serotonin reuptake transporter genes

in IBS patients. Multiple studies, including, the present one, have also demonstrated significant elevations in psychological distress, often associated with pain sensitivity. This is the second investigation, following Wilder Smith et al. (Wilder-Smith et al., 2004) to identify dysregulation in endogenous analgesic mechanisms for IBS. However, this is the first study to adequately control for non-specific effects that are known to contribute to DNIC effects.

Limitations of this Study

Failure to identify abnormalities in temporal summation may have been due to the use of test stimuli that was not sufficiently painful, limitations of the equipment to provide sufficiently rapid oscillations in phasic pain stimuli, or both. Replication of this investigation using newly available instrument capable of much faster delivery of stimuli along with increasing the temperature to 53^0 C, as has been done elsewhere, may yield different results.

Although removing non-specific effects from the DNIC effect is an important finding, this was accomplished through statistical means. Isolating the mechanisms responsible for DNIC through direct observation of those mechanisms, such alterations in fMRI activity (Wilder-Smith et al., 2004), or the RII reflex (Sandrini et al., 2006) will improve upon our understanding of physiological mechanisms that contribute to functional pain disorders.

Future Research

Although the conclusions that can be drawn form the Wilder-Smith investigation of DNIC in IBS patients are limited, their observations of abnormalities in fMRI activity in pain processing areas of the brain during the DNIC procedure provides insight into the mechanisms that may be involved in DNIC for IBS patients. Just published results report that deficient DNIC effects were also identified for abnormalities in the RIII nociceptive reflex in patients with migraines (Sandrini et al., 2006). These strategies provide an important

advancement in the study of DNIC in chronic pain disorders, allowing for the isolation of the DNIC mechanism from non-specific effects.

Whether deficits in DNIC are a cause or consequence of chronic pain is not known, but the relationship between DNIC and pain scores in healthy subjects suggests that compromised DNIC is not likely to entirely be a consequence of having chronic pain (Edwards et al., 2003b). Pielsticker et al. (2005) reported that while deficient DNIC was only weakly correlated with headache intensity, it may predispose individuals to developing chromic headache, but not be relevant for sustaining headache later on. On the other hand, Kosek et al. (2000b) showed that osteoarthritis patients lacked DNIC (compared to HCs) when in pain, but recovered DNIC after surgery when they were pain free. This supports the idea that DNIC may be a consequence of chronic pain rather than a cause. Further long-term studies into whether disinhibition of pain in healthy individuals leads to IBS are needed.

Conclusion

In conclusion, the present study showed that IBS patients demonstrated compromised inhibitory regulation of phasic somatic pain stimuli. This disinhibition was independent of autonomic or psychological mechanisms, including distraction. It is hoped that these findings will contribute to the understanding of the role that central pain dysregulation plays in IBS and may lead to improvements in diagnosis, and ultimately, to novel therapies for patients with IBS.

Appendix

Demographic/medical history questionnaire. This questionnaire was developed for the purposes of screening patients referred for evaluation of irritable bowel syndrome, as well as fecal incontinence, pelvic floor dyssynergia, functional dyspepsia, and other functional and motility disorders of the gastrointestinal tract. It is used to provide a systematic medical history with respect to these complaints, and to characterize the bowel habits of the patients. It includes questions on prior health care utilization. Demographic data on gender, age, educational level, and ethnicity are also included. This questionnaire has been given to approximately 5000 gastroenterology medical clinic patients in three states (North Carolina, Washington state, and California). Tests of the validity of the questionnaire have been limited to the symptoms used to diagnose IBS by the Rome criteria: At each of two clinics, 25 patients who met Rome criteria for IBS based on the questionnaire and 25 patients who did not were selected at random and submitted to experienced clinicians who reviewed the charts blindly without reference to the questionnaire. Agreement with the questionnaire was similar for the two clinicians: sensitivity, 73% and 86%; specificity 56% and 64%(Levy et al., 2000a) .

The Irritable Bowel Syndrome Severity scale has been shown to be valid and reliable. In the original validation study of the scale (Francis et al., 1997), it was found to discriminate IBS patients from controls, and to discriminate between patients categorized as mild, moderate, and severe by independent clinical assessment. Test-retest reliability was judged to be excellent (Francis et al., 1997). Whitehead and colleagues adapted the IBSS slightly (Americanized the English) and administered it to 1603 patients with functional bowel disorders including 815 who met Rome II criteria for IBS. The IBSS correlated well (Spearman's rho=.66; $p<.0001$) with the Functional Bowel Disease Severity Index(Drossman

et al., 1995) and also correlated with the frequency of medical clinic visits for IBS in the past 6 months (r=.21; p<001). Internal consistency was adequate, with a standardized item Chronbach's alpha of .73. In their systematic review of outcome measures in IBS clinical trials, Bijkerk et al (2003) (Bijkerk et al., 2003) ranked the instrument as one of the two best outcome measures for IBS trials based on psychometric properties.

Physical Symptoms Questionnaire (Palsson et al., 2002) assessed levels of somatization. This questionnaire was developed to identify the somatic symptoms that are reported with increased frequency by patients with IBS. The RPSQ consists of 26 physical symptoms. Patients are asked to rate the frequency of occurrence of these symptoms in the last 30 days on a 5-point ordinal scale. A validation study was performed on 60 patients meeting Rome II criteria for IBS. The Cornell Medical Index (Abramson et al., 1965) was used to assess convergent validity. The internal consistency (Chronbach's alpha) for this questionnaire is .86, the split-half reliability is .84, and the test-retest reliability is .84 (p<.0001). The correlation with the Cornell Medical Index validity was r=.85. The RPSQ also correlates strongly with number of physician visits in the past year (r=.62).

The State-Trait Anxiety Inventory (Spielberger et al., 1983) has two versions both consisting of 20 adjectives/self-descriptions. Subjects rate the extent to which they feel that they match the statements. One version requires subjects to rate how they presently feel (state anxiety), and the other version asks subjects to report how they generally feel (trait anxiety). Both versions were used for both studies. For the trait anxiety scale, Spielberger et al. report that test-retest reliabilities range from .76 to .84 for 1 hour retesting, from .71 to .86 over 30 days, and from .65 to .77 over 60 days. Median Chronbach's alpha for trait anxiety was .90. Due to the anticipated transitory nature of state anxiety, test-retest reliabilities for the state anxiety measure are only in the low to moderate range. Median Chronbach's alpha for state anxiety

is .93. Factor analysis of the items confirms a two-factor (state and trait anxiety) solution for the instrument. Spielberger et al. report that the scales correlate highly with other measures of anxiety, that the state anxiety measure is sensitive to induced anxiety states such as those produced experimentally, and that trait anxiety shows good utility in identifying clinical populations with anxiety or anxiety related problems. Okun, Stein, Bauman, and Silver (1996) found that the trait measure of the instrument performed fairly well in measuring five of eight factors used to diagnosis generalized anxiety disorder in the DSM-IV.

Speilberger Anger Expression Index (Spielberger et al., 1995): This assessment was used to determine state and trait levels of anger, as this construct has been shown to be related to cardiovascular activity. The Anger Expression Index has well documented reliability and validity(Spielberger et al., 1995).

Perceived Stress Scale (Cohen et al., 1983) This assessment consists of 14 items which are evaluated on a 5-point Likert scale. The items on the PSS tap into the degree to which individuals feel that events in their lives are unpredictable and uncontrollable. Validity and test-retest reliabilities have been demonstrated for this scale will measure recent exposure and responses to stress(Cohen et al., 1983).

Coping Strategies Questionnaire (Rosenstiel & Keefe, 1983) was used to measure pain coping/pain responsivity. Cognitive and behavioral pain coping strategies were assessed by means of a questionnaire in a sample of 61 chronic low back pain patients. Data analysis indicated that the questionnaire was internally reliable. While patients reported using a variety of coping strategies, certain strategies were used frequently whereas others were rarely used. Three factors: (a) Cognitive Coping and Suppression, (b) Helplessness, and (c) Diverting Attention or Praying, accounted for a large proportion of variance in questionnaire responses. These 3 factors were found to be predictive of measures of behavioral and

emotional adjustment to chronic pain above and beyond what may be predicted on the basis of patient history variables (length of continuous pain, disability status, and number of pain surgeries) and the tendency of patients to somaticize. Each of the 3 coping factors was related to specific measures of adjustment to chronic pain (Rosenstiel & Keefe, 1983)

Catastrophizing subscale of the Coping Stratgies Questionnaire (Hirsh et al., 2006) will assess levels of catastrophizing as a coping style. Measurement and conceptual issues of pain catastrophizing have been raised in the literature. The issues of construct redundancy and measurement overlap have received particular attention, with suggestions that measures of pain catastrophizing are confounded with measures of negative mood, namely depression. The current study sought to investigate these issues in the coping strategies questionnaire-catastrophizing subscale, a widely used measure of pain catastrophizing. Chronic pain patients (n=152) were recruited from the University of Florida pain clinics and completed a battery of psychological measures. Regression analyses indicated that measures of depression, anxiety, and anger accounted for 69% and 19% of the variance in measures of pain catastrophizing and pain, respectively. Trait anger and the cognitive and fearful dimensions of depression and anxiety were uniquely associated with pain catastrophizing. After controlling for measures of negative mood, pain catastrophizing contributed minimally to the prediction of pain. This study suggests that the catastrophizing is highly related to measures of negative mood and raises doubts about its measurement of the construct of pain catastrophizing. Results also provide support for theoretical accounts of the relationships between pain catastrophizing, negative mood, and pain. Clinical implications, future research directions, and alternative measures of pain catastrophizing are discussed.

References

Aaron, L. A., Burke, M. M., & Buchwald, D. (2000). Overlapping conditions among patients with chronic fatigue syndrome, fibromyalgia, and temporomandibular disorder. *Arch.Intern.Med., 160,* 221-227.

Abramson, J. H., Terespolsky, L., Brook, J. G., & Kark, S. L. (1965). Cornell Medical Index as a health measure in epidemiological studies. A test of the validity of a health questionnaire. *British Journal of Preventive and Social Medicine, 19,* 103-110.

Accarino, A. M., Azpiroz, F., & Malagelada, J. R. (1995). Selective dysfunction of mechanosensitive intestinal afferents in irritable bowel syndrome. *Gastroenterology, 108,* 636-643.

Agreus, L., Svardsudd, K., Nyren, O., & Tibblin, G. (1995). Irritable bowel syndrome and dyspepsia in the general population: overlap and lack of stability over time. *Gastroenterology, 109,* 671-680.

Al-Chaer, E. D., Kawasaki, M., & Pasricha, P. J. (2000). A new model of chronic visceral hypersensitivity in adult rats induced by colon irritation during postnatal development. *Gastroenterology, 119,* 1276-1285.

American Gastroenterological Association (2001). *The Burdon of Gastrointestinal Disease*. American Gastroenterological Association, Bethesda, MD.

Aziz, Q., Thompson, D. G., Ng, V. W., Hamdy, S., Sarkar, S., Brammer, M. J. et al. (2000). Cortical processing of human somatic and visceral sensation. *J.Neurosci., 20,* 2657-2663.

Backonja, M., Arndt, G., Gombar, K. A., Check, B., & Zimmermann, M. (1994). Response of chronic neuropathic pain syndromes to ketamine: a preliminary study. *Pain, 56,* 51-57.

Beck, A. T. (1961). A systematic investigation of depression. *Compr.Psychiatry, 2,* 163-170.

Bennett, E. J., Tennant, C. C., Piesse, C., Badcock, C. A., & Kellow, J. E. (1998). Level of chronic life stress predicts clinical outcome in irritable bowel syndrome. *GUT, 43,* 256-261.

Bhalang, K., Sigurdsson, A., Slade, G. D., & Maixner, W. (2005). Associations among four modalities of experimental pain in women. *J.Pain, 6,* 604-611.

Bijkerk, C. J., de Wit, N. J., Muris, J. W., Jones, R. H., Knottnerus, J. A., & Hoes, A. W. (2003). Outcome measures in irritable bowel syndrome: comparison of psychometric and methodological characteristics. *Am.J Gastroenterol, 98,* 122-127.

Blomhoff, S., Jacobsen, M. B., Spetalen, S., Dahm, A., & Malt, U. F. (2000). Perceptual hyperreactivity to auditory stimuli in patients with irritable bowel syndrome. *Scand.J.Gastroenterol., 35,* 583-589.

Bouin, M., Delvaux, M., Blanc, C., Lagier, E., Delisle, M. B., Fioramonti, J. et al. (2001). Intrarectal injection of glycerol induces hypersensitivity to rectal distension in healthy subjects without modifying rectal compliance. *Eur.J.Gastroenterol.Hepatol., 13,* 573-580.

Bouin, M., Lupien, F., Riberdy, M., Boivin, M., Plourde, V., & Poitras, P. (2004). Intolerance to visceral distension in functional dyspepsia or irritable bowel syndrome: an organ specific defect or a pan intestinal dysregulation? *Neurogastroenterol.Motil., 16,* 311-314.

Bouin, M., Plourde, V., Boivin, M., Riberdy, M., Lupien, F., Laganiere, M. et al. (2002). Rectal distention testing in patients with irritable bowel syndrome: sensitivity, specificity, and predictive values of pain sensory thresholds. *Gastroenterology, 122,* 1771-1777.

Bruehl, S., Chung, O. Y., & Burns, J. W. (2006a). Anger expression and pain: an overview of findings and possible mechanisms. *J.Behav.Med., 29,* 593-606.

Bruehl, S., Chung, O. Y., & Burns, J. W. (2006b). Trait anger and blood pressure recovery following acute pain: evidence for opioid-mediated effects. *Int.J.Behav.Med., 13,* 138-146.

Bruehl, S., Chung, O. Y., Burns, J. W., & Biridepalli, S. (2003). The association between anger expression and chronic pain intensity: evidence for partial mediation by endogenous opioid dysfunction. *Pain, 106,* 317-324.

Bruehl, S., Chung, O. Y., Donahue, B. S., & Burns, J. W. (2006c). Anger regulation style, postoperative pain, and relationship to the A118G mu opioid receptor gene polymorphism: a preliminary study. *J.Behav.Med., 29,* 161-169.

Buenaver, L. F., Edwards, R. R., & Haythornthwaite, J. A. (2007). Pain-related catastrophizing and perceived social responses: Inter-relationships in the context of chronic pain. *Pain, 127,* 234-242.

Bueno, L. & Fioramonti, J. (2002). Visceral perception: inflammatory and non-inflammatory mediators. *Gut, 51 Suppl 1,* i19-i23.

Burns, J. W., Bruehl, S., & Caceres, C. (2004). Anger management style, blood pressure reactivity, and acute pain sensitivity: evidence for "Trait x Situation" models. *Ann.Behav.Med., 27,* 195-204.

Camilleri, M. (1999). Review article: clinical evidence to support current therapies of irritable bowel syndrome. *Alimentary Pharmacology & Therapeutics, 13 Suppl 2,* 48-53.

Cervero, F. & Laird, J. M. (2004). Understanding the signaling and transmission of visceral nociceptive events. *J.Neurobiol., 61,* 45-54.

Chang, L., Mayer, E. A., Johnson, T., FitzGerald, L. Z., & Naliboff, B. (2000). Differences in somatic perception in female patients with irritable bowel syndrome with and without fibromyalgia. *Pain, 84,* 297-307.

Chey, W. D., Beydoun, A., Roberts, D. J., Hasler, W. L., & Owyang, C. (1995). Octreotide reduces perception of rectal electrical stimulation by spinal afferent pathway inhibition. *Am.J.Physiol, 269,* G821-G826.

Chitour, D., Dickenson, A. H., & Le, B. D. (1982). Pharmacological evidence for the involvement of serotonergic mechanisms in diffuse noxious inhibitory controls (DNIC). *Brain Res., 236,* 329-337.

Chun, A., Desautels, S., Slivka, A., Mitrani, C., Starz, T., DiLorenzo, C. et al. (1999). Visceral algesia in irritable bowel syndrome, fibromyalgia, and sphincter of oddi dysfunction, type III. *Dig.Dis.Sci., 44,* 631-636.

Coffin, B., Bouhassira, D., Sabate, J. M., Barbe, L., & Jian, R. (2004). Alteration of the spinal modulation of nociceptive processing in patients with irritable bowel syndrome. *Gut, 53,* 1465-1470.

Cohen, S., Kamarck, T., & Mermelstein, R. (1983). A global measure of perceived stress. *J.Health Soc.Behav., 24,* 385-396.

Cook, I. J., van, E. A., & Collins, S. M. (1987). Patients with irritable bowel syndrome have greater pain tolerance than normal subjects. *Gastroenterology, 93,* 727-733.

Crowell, M. D., Harris, L., Jones, M. P., & Chang, L. (2005). New insights into the pathophysiology of irritable bowel syndrome: implications for future treatments. *Curr.Gastroenterol.Rep., 7,* 272-279.

De, B. T., Cesaro, P., Willer, J. C., & Le, B. D. (1990). Diffuse noxious inhibitory controls in man. Involvement of the spinoreticular tract. *Brain, 113 (Pt 4),* 1223-1234.

Delvaux, M., Beck, A., Jacob, J., Bouzamondo, H., Weber, F. T., & Frexinos, J. (2004). Effect of asimadoline, a kappa opioid agonist, on pain induced by colonic distension in patients with irritable bowel syndrome. *Aliment.Pharmacol.Ther., 20,* 237-246.

Drossman, D. A. (1999). Mind over matter in the postinfective irritable bowel. *GUT, 44,* 306-307.

Drossman, D. A., Corazziari, E., Talley, N. J., Thompson, W. G., & Whitehead, W. E. (2000). *Rome II. The Functional Gastrointestinal Disorders. Diagnosis, Pathophysiology and Treatment: A Multinational Consensus*. (2 ed.) McLean, VA: Degnon Associates.

Drossman, D. A., Li, Z., Andruzzi, E., Temple, R. D., Talley, N. J., Thompson, W. G. et al. (1993). U.S. householder survey of functional gastrointestinal disorders. Prevalence, sociodemography, and health impact. *Dig.Dis.Sci., 38,* 1569-1580.

Drossman, D. A., Li, Z. M., Toner, B. B., Diamant, N. E., Creed, F. H., Thompson, D. et al. (1995). Functional Bowel Disorders - A Multicenter Comparison of Health-Status and Development of Illness Severity Index. *Digestive Diseases and Sciences, 40,* 986-995.

Drossman, D. A., Whitehead, W. E., & Camilleri, M. (1997). Irritable bowel syndrome: A technical review for practice guideline development. *Gastroenterology, 112,* 2120-2137.

Edwards, R. R. (2005). Individual differences in endogenous pain modulation as a risk factor for chronic pain. *Neurology, 65,* 437-443.

Edwards, R. R., Fillingim, R. B., Maixner, W., Sigurdsson, A., & Haythornthwaite, J. (2004). Catastrophizing predicts changes in thermal pain responses after resolution of acute dental pain. *Journal of Pain, 5,* 164-170.

Edwards, R. R., Fillingim, R. B., & Ness, T. J. (2003a). Age-related differences in endogenous pain modulation: a comparison of diffuse noxious inhibitory controls in healthy older and younger adults. *Pain, 101,* 155-165.

Edwards, R. R., Ness, T. J., Weigent, D. A., & Fillingim, R. B. (2003b). Individual differences in diffuse noxious inhibitory controls (DNIC): association with clinical variables. *Pain, 106,* 427-437.

Edwards, R. R., Smith, M. T., Kudel, I., & Haythornthwaite, J. (2006a). Pain-related catastrophizing as a risk factor for suicidal ideation in chronic pain. *Pain, 126,* 272-279.

Edwards, R. R., Smith, M. T., Stonerock, G., & Haythornthwaite, J. A. (2006b). Pain-related catastrophizing in healthy women is associated with greater temporal summation of and reduced habituation to thermal pain. *Clin.J.Pain, 22,* 730-737.

Eide, K., Stubhaug, A., Oye, I., & Breivik, H. (1995). Continuous subcutaneous administration of the N-methyl-D-aspartic acid (NMDA) receptor antagonist ketamine in the treatment of post-herpetic neuralgia. *Pain, 61,* 221-228.

Ekblom, A. & Hansson, P. (1987). Thermal sensitivity is not changed by acute pain or afferent stimulation. *J.Neurol.Neurosurg.Psychiatry, 50,* 1216-1220.

France, C. R., France, J. L., al'Absi, M., Ring, C., & McIntyre, D. (2002a). Catastrophizing is related to pain ratings, but not nociceptive flexion reflex threshold. *Pain, 99,* 459-463.

France, C. R., Froese, S. A., & Stewart, J. C. (2002b). Altered central nervous system processing of noxious stimuli contributes to decreased nociceptive responding in individuals at risk for hypertension. *Pain, 98,* 101-108.

Francis, C. Y., Morris, J., & Whorwell, P. J. (1997). The irritable bowel severity scoring system: a simple method of monitoring irritable bowel syndrome and its progress. *Alimentary Pharmacology & Therapeutics, 11,* 395-402.

Garrison, D. W., Chandler, M. J., & Foreman, R. D. (1992). Viscerosomatic convergence onto feline spinal neurons from esophagus, heart and somatic fields: effects of inflammation. *Pain, 49,* 373-382.

Gebhart, G. F., Bielefeldt, K., & Ozaki, N. (2002). Gastric hyperalgesia and changes in voltage gated sodium channel function in the rat. *Gut, 51 Suppl 1,* i15-i18.

Giamberardino, M. A., Affaitati, G., Lerza, R., & Vecchiet, L. (2000). Pre-emptive analgesia in rats with artificial ureteric calculosis. Effects on visceral pain behavior in the post-operative period. *Brain Res., 878,* 148-154.

Gracely, R. H., Geisser, M. E., Giesecke, T., Grant, M. A., Petzke, F., Williams, D. A. et al. (2004). Pain catastrophizing and neural responses to pain among persons with fibromyalgia. *Brain, 127,* 835-843.

Granot, M., Friedman, M., Yarnitsky, D., & Zimmer, E. Z. (2002). Enhancement of the perception of systemic pain in women with vulvar vestibulitis. *BJOG., 109,* 863-866.

Guieu, R., Serratrice, G., & Pouget, J. (1994). Counter irritation test in primary fibromyalgia. *Clin.Rheumatol., 13,* 605-610.

Gwee, K. A., Collins, S. M., Read, N. W., Rajnakova, A., Deng, Y., Graham, J. C. et al. (2003). Increased rectal mucosal expression of interleukin 1beta in recently acquired post-infectious irritable bowel syndrome. *Gut, 52,* 523-526.

Gwee, K. A., Graham, J. C., McKendrick, M. W., Collins, S. M., Marshall, J. S., Walters, S. J. et al. (1996). Psychometric scores and persistence of irritable bowel after infectious diarrhoea. *Lancet, 347,* 150-153.

Gwee, K. A., Leong, Y. L., Graham, C., McKendrick, M. W., Collins, S. M., Walters, S. J. et al. (1999). The role of psychological and biological factors in postinfective gut dysfunction. *Gut, 44,* 400-406.

Herrero, J. F., Laird, J. M., & Lopez-Garcia, J. A. (2000). Wind-up of spinal cord neurones and pain sensation: much ado about something? *Prog.Neurobiol., 61,* 169-203.

Hirsh, A. T., George, S. Z., Riley, J. L., III, & Robinson, M. E. (2006). An evaluation of the measurement of pain catastrophizing by the coping strategies questionnaire. *Eur.J.Pain*.

Hobday, D. I., Hobson, A., Furlong, P. L., Thompson, D. G., & Aziz, Q. (2000). Comparison of cortical potentials evoked by mechanical and electrical stimulation of the rectum. *Neurogastroenterol.Motil., 12,* 547-554.

Houghton, L. A., Lea, R., Jackson, N., & Whorwell, P. J. (2002). The menstrual cycle affects rectal sensitivity in patients with irritable bowel syndrome but not healthy volunteers. *Gut, 50,* 471-474.

Hummel, T., Sengupta, J. N., Meller, S. T., & Gebhart, G. F. (1997). Responses of T2-4 spinal cord neurons to irritation of the lower airways in the rat. *Am.J.Physiol, 273,* R1147-R1157.

Johannesson, U., de Boussard, C. N., Brodda, J. G., & Bohm-Starke, N. (2006). Evidence of diffuse noxious inhibitory controls (DNIC) elicited by cold noxious stimulation in patients with provoked vestibulodynia. *Pain*.

Kashima, K., Rahman, O. I., Sakoda, S., & Shiba, R. (1999). Increased pain sensitivity of the upper extremities of TMD patients with myalgia to experimentally-evoked noxious stimulation: possibility of worsened endogenous opioid systems. *Cranio., 17,* 241-246.

Kawasaki, M. & Al-Chaer, E. D. (2003). Intradermal capsaicin inhibits lumbar dorsal horn neuronal responses to colorectal distention. *Neuroreport, 14,* 985-989.

Kerns, R. D., Rosenberg, R., & Jacob, M. C. (1994). Anger expression and chronic pain. *J.Behav.Med., 17,* 57-67.

Kim, H. J., Camilleri, M., Carlson, P. J., Cremonini, F., Ferber, I., Stephens, D. et al. (2004). Association of distinct alpha(2) adrenoceptor and serotonin transporter polymorphisms with constipation and somatic symptoms in functional gastrointestinal disorders. *Gut, 53,* 829-837.

Kirkup, A. J., Brunsden, A. M., & Grundy, D. (2001). Receptors and transmission in the brain-gut axis: potential for novel therapies. I. Receptors on visceral afferents. *Am.J.Physiol Gastrointest.Liver Physiol, 280,* G787-G794.

Kleinbohl, D., Holzl, R., Moltner, A., Rommel, C., Weber, C., & Osswald, P. M. (1999). Psychophysical measures of sensitization to tonic heat discriminate chronic pain patients. *Pain, 81,* 35-43.

Kosek, E. & Hansson, P. (1997). Modulatory influence on somatosensory perception from vibration and heterotopic noxious conditioning stimulation (HNCS) in fibromyalgia patients and healthy subjects. *Pain, 70,* 41-51.

Kosek, E. & Ordeberg, G. (2000a). Abnormalities of somatosensory perception in patients with painful osteoarthritis normalize following successful treatment. *Eur.J.Pain, 4,* 229-238.

Kosek, E. & Ordeberg, G. (2000b). Lack of pressure pain modulation by heterotopic noxious conditioning stimulation in patients with painful osteoarthritis before, but not following, surgical pain relief. *Pain, 88,* 69-78.

Kwan, C. L., Davis, K. D., Mikula, K., & Diamant, N. E. (2004). Abnormal rectal motor physiology in patients with irritable bowel syndrome. *Neurogastroenterol.Motil., 16,* 251-263.

Lautenbacher, S. & Rollman, G. B. (1997). Possible deficiencies of pain modulation in fibromyalgia. *Clinical Journal of Pain, 13,* 189-196.

Lautenbacher, S., Roscher, S., & Strian, F. (2002). Inhibitory effects do not depend on the subjective experience of pain during heterotopic noxious conditioning stimulation (HNCS): a contribution to the psychophysics of pain inhibition. *Eur.J.Pain, 6,* 365-374.

Le, B. D., Villanueva, L., Bouhassira, D., & Willer, J. C. (1992). Diffuse noxious inhibitory controls (DNIC) in animals and in man. *Patol.Fiziol.Eksp.Ter.,* 55-65.

Leffler, A. S., Hansson, P., & Kosek, E. (2002a). Somatosensory perception in a remote pain-free area and function of diffuse noxious inhibitory controls (DNIC) in patients suffering from long-term trapezius myalgia. *Eur.J.Pain, 6,* 149-159.

Leffler, A. S., Kosek, E., Lerndal, T., Nordmark, B., & Hansson, P. (2002b). Somatosensory perception and function of diffuse noxious inhibitory controls (DNIC) in patients suffering from rheumatoid arthritis. *Eur.J.Pain, 6,* 161-176.

Lembo, T. (2000). Neurotransmitter antagonism in management of irritable bowel syndrome. *Lancet, 355,* 1030-1031.

Lembo, T., Munakata, J., Mertz, H., Niazi, N., Kodner, A., Nikas, V. et al. (1994). Evidence for the hypersensitivity of lumbar splanchnic afferents in irritable bowel syndrome. *Gastroenterology, 107,* 1686-1696.

Levy, R. L., Whitehead, W. E., Feld, A. D., VonKorff, M., & Bjorling, E. (2000a). Agreement between clinician recorded diagnoses and Rome I and Rome II diagnostic criteria for irritable bowel syndrome. *Gastroenterology, 118,* A395.

Levy, R. L., Whitehead, W. E., Von Korff, M. R., & Feld, A. D. (2000b). Intergenerational transmission of gastrointestinal illness behavior. *American Journal of Gastroenterology, 95,* 451-456.

Li, J., Simone, D. A., & Larson, A. A. (1999). Windup leads to characteristics of central sensitization. *Pain, 79,* 75-82.

Longe, S. E., Wise, R., Bantick, S., Lloyd, D., Johansen-Berg, H., McGlone, F. et al. (2001). Counter-stimulatory effects on pain perception and processing are significantly altered by attention: an fMRI study. *Neuroreport, 12,* 2021-2025.

Maixner, W., Fillingim, R., Booker, D., & Sigurdsson, A. (1995). Sensitivity of patients with painful temporomandibular disorders to experimentally evoked pain. *Pain, 63,* 341-351.

Maixner, W., Fillingim, R., Kincaid, S., Sigurdsson, A., & Harris, M. B. (1997). Relationship between pain sensitivity and resting arterial blood pressure in patients with painful temporomandibular disorders. *Psychosomatic Medicine, 59,* 503-511.

Maixner, W., Fillingim, R., Sigurdsson, A., Kincaid, S., & Silva, S. (1998). Sensitivity of patients with painful temporomandibular disorders to experimentally evoked pain: evidence for altered temporal summation of pain. *Pain, 76,* 71-81.

Materazzo, F., Cathcart, S., & Pritchard, D. (2000). Anger, depression, and coping interactions in headache activity and adjustment: a controlled study. *J.Psychosom.Res., 49,* 69-75.

Mayer, E., Schuster, M., & Norton, N. Irritable Bowel Syndrome. 1-1-1999. Novartis Disease Education Series.

McRoberts, J. A., Coutinho, S. V., Marvizon, J. C., Grady, E. F., Tognetto, M., Sengupta, J. N. et al. (2001). Role of peripheral N-methyl-D-aspartate (NMDA) receptors in visceral nociception in rats. *Gastroenterology, 120,* 1737-1748.

Mendell, L. M. (1966). Physiological properties of unmyelinated fiber projection to the spinal cord. *Exp.Neurol., 16,* 316-332.

Mertz, H. (2003). Review article: visceral hypersensitivity. *Aliment.Pharmacol.Ther., 17,* 623-633.

Mertz, H., Morgan, V., Tanner, G., Pickens, D., Price, R., Shyr, Y. et al. (2000). Regional cerebral activation in irritable bowel syndrome and control subjects with painful and nonpainful rectal distention. *Gastroenterology, 118,* 842-848.

Mertz, H., Naliboff, B., Munakata, J., Niazi, N., & Mayer, E. A. (1995). Altered rectal perception is a biological marker of patients with irritable bowel syndrome. *Gastroenterology, 109,* 40-52.

Miranda, A., Peles, S., Rudolph, C., Shaker, R., & Sengupta, J. N. (2004). Altered visceral sensation in response to somatic pain in the rat. *Gastroenterology, 126,* 1082-1089.

Munakata, J., Naliboff, B., Harraf, F., Kodner, A., Lembo, T., Chang, L. et al. (1997a). Repetitive sigmoid stimulation induces rectal hyperalgesia in patients with irritable bowel syndrome. *Gastroenterology, 112,* 55-63.

Munakata, J., Naliboff, B., Harraf, F., Kodner, A., Lembo, T., Chang, L. et al. (1997b). Repetitive sigmoid stimulation induces rectal hyperalgesia in patients with irritable bowel syndrome. *Gastroenterology, 112,* 55-63.

Naliboff, B. D., Munakata, J., Fullerton, S., Gracely, R. H., Kodner, A., Harraf, F. et al. (1997). Evidence for two distinct perceptual alterations in irritable bowel syndrome. *Gut, 41,* 505-512.

O'Mahony, L., McCarthy, J., Kelly, P., Hurley, G., Luo, F., Chen, K. et al. (2005). Lactobacillus and bifidobacterium in irritable bowel syndrome: symptom responses and relationship to cytokine profiles. *Gastroenterology, 128,* 541-551.

Palmgreen, P., Stephenson, M. T., Everett, M. W., Baseheart, J. R., & Francies, R. (2002). Perceived message sensation value (PMSV) and the dimensions and validation of a PMSV scale. *Health Commun., 14,* 403-428.

Palsson, O. S., Jones, K. R., Turner, M. J., Drossman, D. A., & Whitehead, W. E. (2002). Development of specific questionnaires to assess somatization and comorbid diagnoses in irritable bowel syndrome (IBS). *Gastroenterology, 122,* A502.

Pata, C., Erdal, M. E., Derici, E., Yazar, A., Kanik, A., & Ulu, O. (2002). Serotonin transporter gene polymorphism in irritable bowel syndrome. *Am.J.Gastroenterol., 97,* 1780-1784.

Peles, S., Miranda, A., Shaker, R., & Sengupta, J. N. (2004). Acute nociceptive somatic stimulus sensitizes neurones in the spinal cord to colonic distension in the rat. *J.Physiol, 560,* 291-302.

Pertovaara, A., Kemppainen, P., Johansson, G., & Karonen, S. L. (1982). Ischemic pain nonsegmentally produces a predominant reduction of pain and thermal sensitivity in man: a selective role for endogenous opioids. *Brain Res., 251,* 83-92.

Peters, M. L., Schmidt, A. J., Van den Hout, M. A., Koopmans, R., & Sluijter, M. E. (1992). Chronic back pain, acute postoperative pain and the activation of diffuse noxious inhibitory controls (DNIC). *Pain, 50,* 177-187.

Pielsticker, A., Haag, G., Zaudig, M., & Lautenbacher, S. (2005). Impairment of pain inhibition in chronic tension-type headache. *Pain, 118,* 215-223.

Plaghki, L., Delisle, D., & Godfraind, J. M. (1994). Heterotopic nociceptive conditioning stimuli and mental task modulate differently the perception and physiological correlates of short CO2 laser stimuli. *Pain, 57,* 181-192.

Price, D. D. (1972). Characteristics of second pain and flexion reflexes indicative of prolonged central summation. *Exp.Neurol., 37,* 371-387.

Price, D. D. (1991). The use of experimental pain in evaluating the effects of dorsal column stimulation on clinical pain. *Pain, 45,* 225-226.

Price, D. D. (1999). *Psychological Mechanisms of Pain and Analgesia. International Association for the Study of Pain.* Sealtle,.: IASP Press.

Price, D. D. & Dubner, R. (1977). Mechanisms of first and second pain in the peripheral and central nervous systems. *J.Invest Dermatol., 69,* 167-171.

Price, D. D., Hu, J. W., Dubner, R., & Gracely, R. H. (1977). Peripheral suppression of first pain and central summation of second pain evoked by noxious heat pulses. *Pain, 3,* 57-68.

Price, D. D. & McHaffie, J. G. (1988). Effects of heterotopic conditioning stimuli on first and second pain: a psychophysical evaluation in humans. *Pain, 34,* 245-252.

Reinert, A., Treede, R., & Bromm, B. (2000). The pain inhibiting pain effect: an electrophysiological study in humans. *Brain Res., 862,* 103-110.

Ren, K. (1994). Wind-up and the NMDA receptor: from animal studies to humans. *Pain, 59,* 157-158.

Riley, J. F. & Levine, F. M. (1988). Counterstimulation and pain perception: effects of electrocutaneous vs. auditory stimulation upon cold pressor pain. *Pain, 35,* 259-264.

Ritchie, J. (1973). Pain from distension of the pelvic colon by inflating a balloon in the irritable colon syndrome. *Gut, 14,* 125-132.

Roby-Brami, A., Bussel, B., Willer, J. C., & Le, B. D. (1987). An electrophysiological investigation into the pain-relieving effects of heterotopic nociceptive stimuli. Probable involvement of a supraspinal loop. *Brain, 110 (Pt 6),* 1497-1508.

Rodrigues, A. C., Nicholas, V. G., Schmidt, S., & Mauderli, A. P. (2005). Hypersensitivity to cutaneous thermal nociceptive stimuli in irritable bowel syndrome. *Pain, 115,* 5-11.

Rosenstiel, A. K. & Keefe, F. J. (1983). The use of coping strategies in chronic low back pain patients: relationship to patient characteristics and current adjustment. *Pain, 17,* 33-44.

Rossel, P., Drewes, A. M., Petersen, P., Nielsen, J., & rendt-Nielsen, L. (1999). Pain produced by electric stimulation of the rectum in patients with irritable bowel syndrome: further evidence of visceral hyperalgesia. *Scand.J.Gastroenterol., 34,* 1001-1006.

Roza, C., Laird, J. M., & Cervero, F. (1998). Spinal mechanisms underlying persistent pain and referred hyperalgesia in rats with an experimental ureteric stone. *J.Neurophysiol., 79,* 1603-1612.

Saito, Y. A., Locke, G. R., Talley, N. J., Zinsmeister, A. R., Fett, S. L., & Melton, L. J., III (2000). A comparison of the Rome and Manning criteria for case identification in epidemiological investigations of irritable bowel syndrome. *Am.J.Gastroenterol., 95,* 2816-2824.

Sandrini, G., Rossi, P., Milanov, I., Serrao, M., Cecchini, A. P., & Nappi, G. (2006). Abnormal modulatory influence of diffuse noxious inhibitory controls in migraine and chronic tension-type headache patients. *Cephalalgia, 26,* 782-789.

Sarkar, S., Woolf, C. J., Hobson, A. R., Thompson, D. G., & Aziz, Q. (2006). Perceptual wind-up in the human oesophagus is enhanced by central sensitisation. *Gut, 55,* 920-925.

Sarlani, E., Grace, E. G., Reynolds, M. A., & Greenspan, J. D. (2004). Evidence for up-regulated central nociceptive processing in patients with masticatory myofascial pain. *J.Orofac.Pain, 18,* 41-55.

Serra, J., Azpiroz, F., & Malagelada, J. R. (1998). Intestinal gas dynamics and tolerance in humans. *Gastroenterology, 115,* 542-550.

Siddall, P. J. & Cousins, M. J. (2004). Persistent pain as a disease entity: implications for clinical management. *Anesth.Analg., 99,* 510-20, table.

Sigurdsson, A. & Maixner, W. (1994). Effects of experimental and clinical noxious counterirritants on pain perception. *Pain, 57,* 265-275.

Spielberger, C. D., Gorsuch, R. L., Lushene, R., Vagg, P. R., & Jacobs, G. A. (1983). *Manual for the State-Trait Anxiety Inventory*. Palo Alto: Consulting Psychologists Press.

Spielberger, C. D., Reheiser, E. C., & Sydeman, S. J. (1995). Measuring the experience, expression, and control of anger. *Issues Compr.Pediatr.Nurs., 18,* 207-232.

Spiller, R. C. (2003). Postinfectious irritable bowel syndrome. *Gastroenterology, 124,* 1662-1671.

Staud, R., Robinson, M. E., Vierck, C. J., Jr., & Price, D. D. (2003). Diffuse noxious inhibitory controls (DNIC) attenuate temporal summation of second pain in normal males but not in normal females or fibromyalgia patients. *Pain, 101,* 167-174.

Staud, R. & Rodriguez, M. E. (2006). Mechanisms of disease: pain in fibromyalgia syndrome. *Nat.Clin.Pract.Rheumatol., 2,* 90-98.

Staud, R., Vierck, C. J., Cannon, R. L., Mauderli, A. P., & Price, D. D. (2001). Abnormal sensitization and temporal summation of second pain (wind-up) in patients with fibromyalgia syndrome. *Pain, 91,* 165-175.

Steens, J., Van Der Schaar, P. J., Penning, C., Brussee, J., & Masclee, A. A. (2002). Compliance, tone and sensitivity of the rectum in different subtypes of irritable bowel syndrome. *Neurogastroenterol.Motil., 14,* 241-247.

Talbot, J. D., Duncan, G. H., Bushnell, M. C., & Boyer, M. (1987). Diffuse noxious inhibitory controls (DNICs): psychophysical evidence in man for intersegmental suppression of noxious heat perception by cold pressor pain. *Pain, 30,* 221-232.

Thompson, W. G., Heaton, K. W., Smyth, G. T., & Smyth, C. (1997). Irritable bowel syndrome: the view from general practice. *European Journal of Gastroenterology & Hepatology, 9,* 689-692.

Tracey, I. & Dunckley, P. (2004). Importance of anti- and pro-nociceptive mechanisms in human disease. *Gut, 53,* 1553-1555.

Vase, L., Robinson, M. E., Verne, G. N., & Price, D. D. (2003). The contributions of suggestion, desire, and expectation to placebo effects in irritable bowel syndrome patients. An empirical investigation. *Pain, 105,* 17-25.

Veale, D., Kavanagh, G., Fielding, J. F., & Fitzgerald, O. (1991). Primary fibromyalgia and the irritable bowel syndrome: different expressions of a common pathogenetic process. *Br.J.Rheumatol., 30,* 220-222.

Verne, G. N., Himes, N. C., Robinson, M. E., Gopinath, K. S., Briggs, R. W., Crosson, B. et al. (2003a). Central representation of visceral and cutaneous hypersensitivity in the irritable bowel syndrome. *Pain, 103,* 99-110.

Verne, G. N. & Price, D. D. (2002). Irritable bowel syndrome as a common precipitant of central sensitization. *Curr.Rheumatol.Rep., 4,* 322-328.

Verne, G. N., Price, D. D., & Robinson, M. E. (2000). Hypersensitivity to cutaneous (heat induced) pain in the Irritable Bowel Syndrome. *Gastroenterology, 118,* A845.

Verne, G. N., Robinson, M. E., & Price, D. D. (2001). Hypersensitivity to visceral and cutaneous pain in the irritable bowel syndrome. *Pain, 93,* 7-14.

Verne, G. N., Robinson, M. E., Vase, L., & Price, D. D. (2003c). Reversal of visceral and cutaneous hyperalgesia by local rectal anesthesia in irritable bowel syndrome (IBS) patients. *Pain, 105,* 223-230.

Verne, G. N., Robinson, M. E., Vase, L., & Price, D. D. (2003b). Reversal of visceral and cutaneous hyperalgesia by local rectal anesthesia in irritable bowel syndrome (IBS) patients. *Pain, 105,* 223-230.

Washington, L. L., Gibson, S. J., & Helme, R. D. (2000). Age-related differences in the endogenous analgesic response to repeated cold water immersion in human volunteers. *Pain, 89,* 89-96.

Whitehead W.E., Levy R, L., Von Korff M.V, Feld, A. D., Palsson O.S., Turner M.J. et al. (2004). Usual medical care for irritable bowel syndrome. *Alimentary Pharmacology & Therapeutics, 20,* 1305-1315.

Whitehead, W. E., Crowell, M. D., Robinson, J. C., Heller, B. R., & Schuster, M. M. (1992). Effects of Stressful Life Events on Bowel Symptoms - Subjects with Irritable-Bowel-Syndrome Compared with Subjects Without Bowel Dysfunction. *GUT, 33,* 825-830.

Whitehead, W. E., Engel, B. T., & Schuster, M. M. (1980). Irritable Bowel Syndrome - Physiological and Psychological Differences Between Diarrhea-Predominant and Constipation-Predominant Patients. *Digestive Diseases and Sciences, 25,* 404-413.

Whitehead, W. E., Holtkotter, B., Enck, P., Hoelzl, R., Holmes, K. D., Anthony, J. et al. (1990). Tolerance for rectosigmoid distention in irritable bowel syndrome. *Gastroenterology, 98,* 1187-1192.

Whitehead, W. E., Palsson, O., & Jones, K. R. (2002). Systematic review of the comorbidity of irritable bowel syndrome with other disorders: what are the causes and implications? *Gastroenterology, 122,* 1140-1156.

Whitehead, W. E. & Palsson, O. S. (1998). Is rectal pain sensitivity a biological marker for irritable bowel syndrome: psychological influences on pain perception. *Gastroenterology, 115,* 1263-1271.

Wilder-Smith, C. H., Schindler, D., Lovblad, K., Redmond, S. M., & Nirkko, A. (2004). Brain functional magnetic resonance imaging of rectal pain and activation of endogenous inhibitory mechanisms in irritable bowel syndrome patient subgroups and healthy controls. *Gut, 53,* 1595-1601.

Willer, J. C., Bouhassira, D., & Le, B. D. (1999). [Neurophysiological bases of the counterirritation phenomenon:diffuse control inhibitors induced by nociceptive stimulation]. *Neurophysiol.Clin., 29,* 379-400.

Willer, J. C., De, B. T., & Le, B. D. (1989). Encoding of nociceptive thermal stimuli by diffuse noxious inhibitory controls in humans. *J.Neurophysiol., 62,* 1028-1038.

Willer, J. C., Le, B. D., & De, B. T. (1990). Diffuse noxious inhibitory controls in man: involvement of an opioidergic link. *Eur.J.Pharmacol., 182,* 347-355.

Willert, R. P., Woolf, C. J., Hobson, A. R., Delaney, C., Thompson, D. G., & Aziz, Q. (2004). The development and maintenance of human visceral pain hypersensitivity is dependent on the N-methyl-D-aspartate receptor. *Gastroenterology, 126,* 683-692.

Willis, W. D. (1985). Nociceptive pathways: anatomy and physiology of nociceptive ascending pathways. *Philos.Trans.R.Soc.Lond B Biol.Sci., 308,* 253-270.

Woolf, C. J. (2000). Mechanisms of chronic pain - Proceedings of a symposium Vienna, Austria, August 23, 1999 - Introduction. *Clinical Journal of Pain, 16,* S113.

Woolf, C. J. (2004). Pain: Moving from symptom control toward mechanism-specific pharmacologic management. *Annals of Internal Medicine, 140,* 441-451.

Woolf, C. J. & Wall, P. D. (1986). Relative effectiveness of C primary afferent fibers of different origins in evoking a prolonged facilitation of the flexor reflex in the rat. *J.Neurosci., 6,* 1433-1442.

Xu, X. J., Hao, J. X., Aldskogius, H., Seiger, A., & Wiesenfeld-Hallin, Z. (1992). Chronic pain-related syndrome in rats after ischemic spinal cord lesion: a possible animal model for pain in patients with spinal cord injury. *Pain, 48,* 279-290.

Yeo, A., Boyd, P., Lumsden, S., Saunders, T., Handley, A., Stubbins, M. et al. (2004). Association between a functional polymorphism in the serotonin transporter gene and diarrhoea predominant irritable bowel syndrome in women. *Gut, 53,* 1452-1458.

Zighelboim, J., Talley, N. J., Phillips, S. F., Harmsen, W. S., & Zinsmeister, A. R. (1995). Visceral perception in irritable bowel syndrome. Rectal and gastric responses to distension and serotonin type 3 antagonism. *Dig.Dis.Sci., 40,* 819-827.