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Sex differences in endogenous pain modulation by distracting and painful conditioning stimulation

Raimi L. Quiton and Joel D. Greenspan

Program in Neuroscience, Department of Biomedical Sciences, Dental School, and Research Center for Neuroendocrine Influences on Pain, University of Maryland, Baltimore, Maryland, United States

Abstract

Sex differences in endogenous pain modulation were tested in healthy volunteers (32 men, 30 women). Painful contact heat stimuli were delivered to the right leg alone, and then in combination with various electrical conditioning stimuli delivered to the left forearm. Four conditioning protocols were applied to each subject in separate sessions: mild, nonpainful (control); distracting; stressful-yet-nonpainful; strongly painful. Thermal stimuli were rated on visual analog scales for pain intensity (INT) and unpleasantness (UNP). Distracting and painful conditioning stimuli significantly reduced heat pain INT and UNP ratings for both sexes, with significantly larger distraction effects on INT ratings for men than women ($p=0.004$). No sex differences in pain-evoked hypoalgesia were detected ($p>0.05$). The stress protocol did not consistently reduce heat pain ratings, possibly because the protocol was not sufficiently stressful to activate endogenous modulatory systems. Regression analysis revealed that the magnitude of pain-evoked hypoalgesia was predicted by the perceived distraction ($p=0.003$) and stress ($p=0.04$) produced by the painful conditioning stimulation, providing evidence that distraction and stress contribute to pain-evoked hypoalgesia. However, the contribution of stress to pain-evoked hypoalgesia differed by sex ($p=0.02$), with greater perceived stress associated with greater hypoalgesia in men and the opposite trend in women, suggesting sex differences in the mechanisms underlying pain-evoked hypoalgesia. This study provides indirect evidence that multiple neural mechanisms are involved in endogenous pain modulation and suggests that sex-specific aspects of these systems may contribute to greater pain sensitivity and higher prevalence of many chronic pain conditions among women.

Keywords

pain; sex; endogenous analgesia; distraction; stress; DNIC

INTRODUCTION

Application of strong pain to one part of the body inhibits pain in multiple remote body regions, a phenomenon known as diffuse noxious inhibitory controls, counterirritation, or pain-evoked hypoalgesia. Pain-evoked hypoalgesia is a psychophysical measure of the function of central endogenous pain modulation systems and has been used experimentally to test these systems in healthy (Willer et al. 1979; Price and McHaffie 1988), aged (Edwards et al. 2003a), and

Corresponding author: Raimi L. Quiton, Dept. of Biomedical Sciences, University of Maryland Dental School, 650 W. Baltimore Street, 7 South, Baltimore, MD 21201, phone: (410) 706-2027, fax: (410) 706-4172, e-mail: rquiton@umaryland.edu.

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chronic pain populations (Kashima et al. 1999; Staud et al. 2003). Pain-evoked hypoalgesic magnitude is inversely related to clinical pain frequency among healthy individuals (Edwards et al. 2003b), suggesting that reduced endogenous analgesic system function may be a risk factor for chronic pain.

Deficits in pain-evoked hypoalgesia are associated with several chronic pain conditions that disproportionately affect women, including temporomandibular disorder (Kashima et al. 1999), irritable bowel syndrome (Wilder-Smith et al. 2004), and fibromyalgia (Kosek and Hansson 1997; Lautenbacher and Rollman 1997; Staud et al. 2003; Julien et al. 2005). These findings suggest that women may have reduced endogenous analgesic system function compared to men, resulting in increased vulnerability to the development or maintenance of chronic pain states. Demonstration of sex differences in endogenous pain modulation among healthy subjects would provide support for this hypothesis. However, the few studies examining this issue have produced conflicting results, with some reporting reduced hypoalgesic magnitude (Staud et al. 2003; Serrao et al. 2004) or duration (Ge et al. 2004) in women, and others reporting no sex differences (France and Suchowicki 1999; Baad-Hansen et al. 2005; Pud et al. 2005).

Furthermore, little is known about the factors underlying pain-evoked hypoalgesia. Both distraction and stress can independently reduce pain (Fernandez and Turk 1989; Fields and Basbaum 1999) and are likely to contribute to pain-evoked hypoalgesia, as painful stimuli are not only painful but also distracting and stressful. Sex differences in both distraction- (Weisenberg et al. 1995; Bentsen et al. 1999; Unrod et al. 2004) and stress-evoked hypoalgesia (Koltyn et al. 2001; Rhudy and Meagher 2001; Sternberg et al. 2001; Girdler et al. 2005) have been reported. Thus, pain-evoked hypoalgesia may represent the combined effects of multiple sex-sensitive factors. Because the extent to which distraction and stress contribute to pain-evoked hypoalgesia in humans has not been systematically examined, it is unclear which factors underlie differences in endogenous analgesia reported for various populations.

The overall aim of this study was to more fully characterize sex differences in the function of endogenous pain modulatory systems in healthy human subjects. Electrical conditioning stimuli were used to inhibit sensory and affective components of pain evoked by thermal test stimuli applied to a heterotopic body site. Two hypotheses were tested: (1) Distracting, stressful, and painful conditioning stimuli independently produce significant hypoalgesia in both men and women, with sex differences in the magnitude and duration of the effects. (2) Pain-evoked hypoalgesia is partially mediated by the distracting and stressful nature of the painful conditioning stimuli, and the degree to which these factors are related to the hypoalgesic effects vary by sex.

MATERIALS AND METHODS

Subjects

Male (n=32) and female (n=30) subjects, constituting two groups of comparable age and racial makeup, participated in the study (Table 1). Data were collected for two additional female subjects but excluded from analysis due to the subjects' inability to perform the study tasks. All subjects were healthy, with no major medical, neurological, or chronic pain disorders. No subjects used prescription medication during the study period, with the exception of hormonal contraception (13 females) and hormone replacement therapy (1 female). Three female subjects were in a persistent follicular phase due to continuous use of hormonal contraception or hormone replacement therapy without breaks for menstruation. All other female subjects were tested during the follicular phase of their menstrual cycle (days 3 through 11) to reduce group heterogeneity in gonadal hormone levels, which may influence pain perception (Riley et al. 1999). Testing during the follicular phase was selected because sex differences between males

and follicular phase females have been observed for numerous pain measures, including pain threshold, temporal summation of pain, and endogenous analgesia (Goolkasian 1980; Sarlani et al. 2004; Serrao et al. 2004; Kowalczyk et al 2006). Informed consent was obtained from all subjects prior to experimentation. The protocol for this study was approved by the University of Maryland Institutional Review Board for the Protection of Human Subjects.

Experimental Design

Subjects participated in one training session and four test sessions, each conducted on separate days. During each test session, painful contact heat test stimuli were delivered to the right leg alone, then in combination with various conditioning stimulation protocols that engaged endogenous pain modulatory systems in different ways (Figure 1A). Conditioning protocols consisted of transcutaneous electrical stimuli delivered to the left median nerve and were designed to be distracting but not stressful (DISTRACTION), stressful but not painful (STRESS), or strongly painful (PAIN); mild, nonpainful stimuli were used as a control condition (CONTROL). Subjects experienced a different conditioning protocol in each test session, with the order counterbalanced within each sex. The magnitude of pain modulation evoked by each conditioning protocol was assessed by comparing heat pain ratings before and during application of the electrical conditioning stimulation.

Test Stimuli: Painful Contact Heat

Painful heat stimuli were delivered to the medial surface of the right lower leg using a Peltier thermal probe with a 2.6 cm² contact surface (TSA-II, Medoc Ltd., Israel). The probe was held in place during testing with a Velcro strap. Subjects rated heat pain on a computerized VAS, which consisted of a vertical scale labeled “no pain” at the bottom and “most intense pain imaginable” at the top (DAPSYS, Brian Turnquist, Johns Hopkins University, www.dapsys.net). VAS ratings were converted to numerical values ranging from 0 to 100. Two temperatures of painful heat were delivered to each subject: (1) a subject-specific temperature perceived as strongly painful (pain-hi), which was defined as the temperature the subject rated between 70 and 80 on a 100-point computerized visual analog scale (VAS) for pain intensity and (2) a subject-specific temperature perceived as moderately painful (pain-lo), which was defined as the temperature the subject rated between 40 and 50 on the VAS. Mean stimulus temperatures required to evoke these perceptions are listed in Table 2. Two of the 32 female subjects who completed the study failed to rate both selected temperatures within the target pain ranges in all test sessions; their data is not included in this report. Perceptually-equalized stimuli were used to ensure that changes in heat pain ratings produced by the conditioning protocols were not confounded by differences in baseline heat pain ratings. The two levels of painful heat stimuli were presented in a randomly-ordered sequence during testing to reduce expectation bias.

Conditioning Stimuli: Transcutaneous Electrical Nerve Stimulation

Electrical stimuli were delivered to the left forearm over the median nerve (0.2 msec square pulse waves, 4–30 Hz) using a programmable transcutaneous electrical nerve stimulation unit (Mettler Trio-Stim, Anaheim, CA) and 1.75-inch square reusable cloth electrodes (www.tensproducts.com). Subjects rated the perceived intensity of electrical stimuli on a paper “sensation intensity” VAS, which consisted of a 100-mm horizontal scale labeled “no sensation” at 0 mm, “mild pain” at 50 mm, and “most intense pain imaginable” at 100 mm. Subjects were instructed to use the lower portion of the scale (<50 mm) for nonpainful sensations and the upper portion of the scale for painful sensations. Subjects gained experience with the rating scale during a training session (subsequently described) in which pain threshold was determined by verbal report. To ensure that subjects used the scale properly, the experimenter verified that electrical stimuli delivered at intensities below pain threshold were

rated in the appropriate range on the scale (<50 mm) and that stimuli delivered at intensities above pain threshold were rated >50 mm.

For each conditioning protocol, subject-specific electric current amplitudes were selected to equalize perception within and across the groups and control for the potential influence of perceived conditioning stimulation intensity on the magnitude of the hypoalgesic effect (Le Bars 2002). For the CONTROL and DISTRACTION conditioning protocols, subject-specific electric current perceived as mild and nonpainful (sensation VAS rating 5–10) was applied at 4 Hz. During the DISTRACTION protocol, subjects were asked to pay attention to and make ratings of the electrical conditioning stimuli during periods when painful heat stimuli were applied. Thus, distraction is produced by this protocol primarily through an attentional manipulation, though the use of a stimulus modality that differed from the test stimulus may also be a source of distraction. For the STRESS conditioning protocol, subject-specific electric currents perceived as strong-yet-nonpainful (sensation VAS rating 35–45) were applied, with the frequency of the stimuli varied among 5 frequencies (4, 5, 10, 20 and 30 Hz) in an unpredictable manner to enhance the stressfulness of the protocol. Preliminary tests demonstrated that subjects rated variable frequency stimuli significantly more stressful than comparably intense stimuli of a single frequency (data not shown). For the PAIN conditioning protocol, subject-specific electric currents perceived as strongly painful (sensation VAS rating 70–80) were applied at 4 Hz. A stimulation frequency of 4 Hz was used for three reasons. First, incremental increases in current amplitude at this frequency produce a gradient of sensation from nonpainful tapping to pricking pain, with the peak current well below thresholds for tissue damage (Gibson 1968; Chesterton et al. 2002). This wide range of sensation enabled selection of stimulus intensities appropriate for each condition. Second, painful stimulation delivered at this frequency evokes significant heterotopic hypoalgesia (Chesterton et al. 2002), demonstrating that these stimuli are capable of activating central endogenous pain modulation systems. Third, nonpainful stimulation at this frequency produces no heterotopic hypoalgesic effects (Chesterton et al. 2003), indicating its appropriateness for use in the CONTROL and DISTRACTION conditions.

Experimental Protocol

Training Session—During the training session, subjects were screened for eligibility based on age and health criteria, underwent sensory tests to determine appropriate heat and electrical stimulation parameters for the test sessions, gained experience with the various rating scales, and participated in an abbreviated version of the test session protocol to minimize novelty and nonspecific stress effects during the test sessions.

Heat sensory testing involved application of two stimulus series: first, an ascending series of stimuli (temperatures ranging from 43 to 50.5°C), then a series of stimuli expected to be in the painful range (45 to 50.5°C, presented twice each in randomized order). Each temperature was presented for 15 seconds (including ramp up and down time using a 5°C/sec ramp rate), followed by a 30-second interstimulus period of nonpainful warmth (37°C). After each stimulus, subjects used a trackball to rate peak pain intensity on the computerized pain intensity VAS. VAS ratings were converted to numerical values ranging from 0 to 100. Individual subject ratings were used to determine subject-specific temperatures that evoked perceptions of strong pain (pain-hi) and moderate pain (pain-lo) (Table 2). Subjects were presented with a third series of randomized heat stimuli and rated unpleasantness of the stimuli using a computerized “unpleasantness” VAS. The unpleasantness VAS was labeled “not unpleasant” at the bottom and “most unpleasant sensation imaginable” at the top. Subjects were instructed to distinguish the pain intensity and unpleasantness scales using the “radio analogy” description (Price et al. 1983). Ratings obtained from these scales are appropriate for quantitative analyses because the pain intensity and unpleasantness VASs are validated ratio scale measures of the

sensory and affective dimensions, respectively, of both experimental and clinical pain (Price et al. 1983; Price et al. 1994). Furthermore, neuroimaging studies report that inter-individual differences or manipulation-evoked changes in VAS ratings of pain correlate with differences in pain-related activity in cortical regions that process nociceptive stimuli (Frankenstein et al. 2001; Coghill et al. 2003; Bingel et al. 2007), providing evidence that subjective reports are related to physiological responses to painful stimuli.

Electrical sensory testing consisted of an ascending series of 4 Hz stimuli that started at 0 mA and increased approximately 1 mA per second. Subjects were asked to verbally indicate sensation threshold, pain threshold, and pain tolerance. Based on the electrical current amplitudes corresponding to these measures, amplitudes were estimated for each conditioning protocol. Estimates were verified by applying the stimuli, obtaining ratings, and, if necessary, adjusting the electrical current until the subject rated the stimulus in the target range twice. Ascending series of 5 Hz, 10 Hz, 20 Hz, and 30 Hz stimuli were also applied to identify electrical currents associated with strong, nonpainful stimuli at these frequencies for use in the STRESS protocol.

Following determination of heat and electrical stimulation parameters, subjects participated in an abbreviated version of the test session in which they received simultaneous heat and electrical stimulation and were asked by the experimenter at various time points to rate the stimuli using the appropriate scale. Test sessions were then scheduled, and subjects were asked to avoid extreme deviations from their normal behavior (with respect to sleep, exercise, and consumption of food, caffeine, alcohol, and tobacco) in the 24 hours prior to each session.

Test Sessions—Each subject participated in four test sessions conducted on separate days. In each session, a different electrical conditioning protocol was used to modulate heat pain perception (Figure 1A), with conditioning protocol order counterbalanced across subjects. Each test session consisted of three phases (Baseline, Conditioning, Recovery) during which painful heat stimuli were applied to the right medial calf (Figure 1B). In each phase, 8 heat stimuli were delivered for 15 seconds each (including ramp up and down time at a 5°C/sec ramp rate) with a 30-second interstimulus period of nonpainful warmth (37°C). The 8 stimuli in a series consisted of the two subject-specific temperatures delivered four times each in a randomized order. After each stimulus, the computerized VAS was presented to the subject and the subject rated peak pain intensity or unpleasantness of the stimulus using the trackball. The order in which the two pain dimensions were rated was counterbalanced across subjects. Half the subjects rated pain intensity of the first four stimuli in each series and unpleasantness of the last four stimuli in the series; the order was reversed for the remaining subjects. For each subject, the same temperatures of painful heat were applied in all four test sessions.

In the Conditioning phase, electrical conditioning stimuli were applied to the left forearm for 4 minutes to allow time to engage endogenous pain modulatory systems, after which painful heat stimuli were delivered simultaneously with the conditioning stimuli (Figure 1B). The magnitude of hypoalgesia evoked by a particular conditioning protocol was measured as the change in pain ratings from the Baseline to Conditioning phase. The duration of hypoalgesia was evaluated by comparing change in pain ratings from the Baseline to Recovery phase across conditions (Figure 1B). In the 10-minute rest periods intervening between experimental phases, subjects completed a series of questionnaires (Figure 1B) then rested quietly without moving or talking for the remainder of the rest period.

Other measures obtained in each test session (Figure 1B) included blood pressure and room temperature (both at the beginning and end of each session). Room temperature was measured to verify that it remained within a narrow range (between 20 and 23°C). During the pre-test period, subjects completed a questionnaire to verify that they (1) were not experiencing any

spontaneous pain, (2) had not ingested any medications that could influence pain perception in the 12 hours prior to testing, and (3) had not deviated significantly from their normal behavior with respect to sleep, exercise, and consumption of food, caffeine, alcohol, and tobacco in the 24 hours prior to testing.

Test sessions were conducted between 12 and 7 pm, and at about the same time of day (within a 2-hour window) for each individual subject to control for circadian fluctuations in pain sensitivity (Strian et al. 1989). The interval between test sessions ranged from 1 to 8 days, with a mean of 2.4 days (SD 1.5). All test sessions were completed within a 14-day period with the exception of one female subject (17 days) and two male subjects (19 days and 25 days).

Distraction Scores

The magnitude of distraction produced by each conditioning protocol was assessed immediately after the Conditioning phase using a questionnaire adapted from Edwards et al. (2003a). The questionnaire consisted of two statements: “When I was feeling the heat and electrical stimulation at the same time, I found it difficult to pay attention to the heat stimulation” and “I found it easy to focus on the heat stimuli and give accurate ratings”. Each statement was followed by a 100-mm VAS with the descriptors “not at all” at 0 mm and “very much” at 100 mm. A composite distraction score was obtained by reverse-scoring the second statement, then averaging the scores for both statements.

Stress Reactivity Measures: Self-report, Skin Conductance, and Peripheral Pulse Rate

Self-report measures of perceived stress were obtained at 10 time points during each test session using the stress subscale of the Stress Symptoms Rating Scale (SSR) (Naliboff et al. 1991). The subscale consists of two 100-mm VASs anchored by adjective pairs (relaxed-tense, stressed-at ease). A composite measure of perceived stress was obtained for each time point by reverse-scoring the second rating and averaging it with the first rating. Self-report measures of stress were obtained before and after the Baseline phase; before, at five points during, and after the Conditioning phase; and after the Recovery phase.

Two physiological stress measures--skin conductance and peripheral pulse rate--were obtained continuously throughout the test session using the Psylab SAM system equipped with the SC5 skin conductance amplifier and PPA peripheral pulse amplifier (Contact Precision Instruments, Cambridge, MA). Skin conductance was measured by placing electrodes on the plantar surface of the left foot. Peripheral pulse rate was measured using a photoplethysmographic ear clip. Raw data were sampled at 40 Hz and reduced to 15-second bins for analysis. Due to equipment problems, pulse rate was not obtained for 2 female and 2 male subjects and skin conductance was not obtained for 1 female and 1 male subject.

Psychosocial Measures

Prior to the training session, subjects completed questionnaires to assess psychosocial variables that may influence pain responses. These questionnaires included (1) Spielberger’s Trait Anxiety Inventory, which quantifies generalized (trait) anxiety (Spielberger et al. 1970); (2) the trait version of the Positive and Negative Affect Scale (PANAS), which measures generalized positive affect and negative affect (Watson et al. 1988); (3) the Beck Depression Inventory, which measures attitudes and symptoms of depression (Beck et al. 1961); (4) the Kohn Reactivity Scale, which assesses a subject’s reactivity and arousability to external stimuli (Kohn 1985); (5) the Insomnia Questionnaire, which diagnoses the presence and quantifies the severity of clinical insomnia (Morin 1993), a variable of interest due to its higher prevalence among women (Zhang and Wing 2006) and its potential relationship to pain (Onen et al. 2001; Lautenbacher et al. 2006; Smith et al. 2007); and (6) the Coping Strategies Questionnaire, which evaluates the degree to which an individual utilizes various cognitive strategies for

coping with pain, including diverting attention, reinterpreting pain sensation, making coping self-statements, ignoring pain, praying/hoping, and catastrophizing (Rosenstiel and Keefe 1983). The questionnaires for one male subject were lost during delivery; as a result, psychosocial variables assessed in the questionnaires are reported for 30 female and 31 male subjects.

During each test session, state anxiety and state affect were assessed before and after the Conditioning phase (Figure 1B) using Spielberger's State Anxiety Inventory (Spielberger et al. 1970) and the state version of the PANAS (Watson et al. 1988).

Statistical Analysis

Significant conditioning effects and sex differences in the magnitude of these effects were tested for using a 3-way mixed-model analysis of variance (ANOVA), with sex as the between-subjects factor and protocol and pain level as within-subjects factors. The dependent measure used in the ANOVA was the change in heat pain ratings from the Baseline to Conditioning phase for each conditioning protocol, which reflects the magnitude of the conditioning effects. Posthoc comparisons were made with Newman-Keuls (N-K) tests. Pain intensity and unpleasantness ratings were evaluated separately. A similar statistical approach was used to evaluate the approximate duration of observed conditioning effects; the dependent measure used in this analysis was the change in heat pain ratings from the Baseline to Recovery phase.

Multiple measures (including distraction scores, stress reactivity, change in state anxiety and affect, and blood pressure) were evaluated using 2-way mixed-model ANOVAs, with sex as the between-subjects factor and protocol as the within-subjects factor. Posthoc comparisons consisted of t-tests, with Bonferroni adjustments to correct for multiple comparisons. Independent sample t-tests (two-tailed) were used to evaluate sex differences in normally-distributed demographic measures, lifestyle variables, stimulus temperatures, heat pain ratings, pain sensitivity, and psychosocial measures; non-normally-distributed measures were evaluated with the nonparametric Mann-Whitney U test.

Pearson correlations (or Spearman's for non-normally distributed variables) were carried out to examine the relationship between conditioning effect magnitude and various factors such as distraction, stress, and psychosocial variables in each of the two groups. Multiple linear regression models were used to test the hypotheses that (1) distraction and stress contribute to the modulatory effects of painful conditioning stimuli and (2) the contributions differ by sex. As an exploratory step, predictor variables (distraction and stress reactivity measures) were regressed on PAIN conditioning effect magnitude, separately for each sex. Based on the results of the exploratory regressions, a full regression model was run on conditioning effect magnitude data for the combined groups to test the hypothesis that distraction and stress differentially impact pain-evoked hypoalgesia in men and women. Significance was set at 0.05 for all statistical tests.

RESULTS

Subject Characteristics

Sex differences were not found for most demographic, lifestyle, and psychosocial variables (Table 1). Compared to men, women reported using more caffeine and alcohol, had significantly higher trait anxiety and emotional reactivity scores, and reported significantly greater use of two pain coping strategies: increasing activity (such as going for a walk or doing household chores) and increasing pain behavior (such as laying down, using a heating pad, or taking medication) (Table 1). Females had lower average systolic blood pressure across sessions than males (Table 1). Blood pressure did not vary across conditioning protocols for

either men or women ($p < 0.05$). None of the variables that differed between men and women were correlated significantly with the hypoalgesic effects measured in this study ($p > 0.05$).

Heat Stimulation Parameters

Mean pain intensity ratings for the 2 heat stimulus levels were within the target ranges for both female and male subjects, with relatively low standard deviations (Table 2), demonstrating that the protocol used to achieve equal pain perceptions across subjects was successful. Importantly, mean baseline ratings for each pain level did not differ significantly between the female and male groups (Table 3), providing evidence that sex differences in the hypoalgesic effects of the various conditioning protocols were not confounded by baseline differences in heat pain perception. In addition, baseline heat pain intensity and unpleasantness ratings did not differ significantly across the four conditioning protocols for either group or stimulus level ($p > 0.05$ for all statistical tests), demonstrating that the counterbalancing procedure was successful at accounting for potential confounds due to repeated testing. Mean ratings for pain-hi and pain-lo differed significantly (Table 3), indicating that the two temperatures used in the study represent distinct intensities of pain, with pain-lo evoking a perception in the lower half of the pain range and pain-hi evoking a perception in the upper half of the pain range. Although pain perception did not differ between the female and male groups, females required significantly lower temperatures than males to evoke both targeted levels of perceived pain (Tables 2 and 3). A nonsignificant trend for females to rate 48°C stimuli as more painful than males ($t = 1.8$, $p = 0.08$) was also observed (Table 2).

Electrical Stimulation Parameters

Mean sensation intensity ratings of electrical stimuli used for each conditioning protocol were in the target ranges for both male and female subjects (Table 2), demonstrating success of the protocol used to equalize perceived intensity of the conditioning stimuli. Men and women rated the stimuli for each protocol comparably (Tables 2 and 3); thus, sex differences in the hypoalgesic effects of the various conditioning protocols were not confounded by differences in perceived intensity of the conditioning stimuli. As expected, conditioning stimulation ratings differed significantly across the protocols (Table 3). Consistent with observations for the heat stimuli, females required significantly lower electrical current amplitudes than males to evoke the targeted levels of perceived conditioning stimulation intensity (Table 3).

Conditioning Effects on Heat Pain Intensity Ratings

The ANOVA used to evaluate the magnitude of pain modulation produced by each conditioning protocol (Table 4, Figure 2) detected significant interactions between sex and protocol ($F(3,180) = 2.6$, $p = 0.05$) and between protocol and pain level ($F(3,180) = 2.6$, $p = 0.05$). Posthoc analyses conducted to decompose the sex-by-protocol interaction revealed a significant sex difference in the magnitude of pain modulation produced by the DISTRACTION protocol (N-K, $p = 0.004$) but no sex difference for the STRESS (N-K, $p = 0.22$) or PAIN (N-K, $p = 0.53$) protocols. Posthoc analyses conducted to decompose the protocol-by-pain level interaction revealed that the DISTRACTION, STRESS, and PAIN protocols produced significantly greater reduction in pain-lo intensity ratings than the CONTROL protocol (N-K, $p < 0.001$ for all 3 protocols), indicating that all three non-control conditioning protocols significantly reduced perception of pain-lo stimuli. However, only the DISTRACTION and PAIN protocols reduced pain-hi ratings to a significantly greater degree than CONTROL (N-K, $p < 0.001$ for both protocols); the STRESS protocol had no significant effect on pain-hi ratings (N-K, $p = 0.15$). A measure of conditioning effect duration was calculated as the difference between Baseline and Recovery phase heat pain intensity ratings. The ANOVA used to evaluate the duration measures found no significant main effect of

protocol ($p=0.3$) or significant interactions (all $p>0.05$), indicating that none of the conditioning effects persisted for more than 10 minutes after cessation of the conditioning stimulation.

Conditioning Effects on Unpleasantness Ratings of Painful Heat

The ANOVA used to evaluate conditioning protocol effects on unpleasantness ratings (Table 4, Figure 3) found a significant interaction between protocol and pain level ($F(3,180)=4.7$, $p=0.004$). Posthoc analyses conducted to decompose the interaction revealed that the DISTRACTION protocol reduced pain-hi unpleasantness ratings to a significantly greater degree than pain-lo unpleasantness ratings (N-K, $p<0.001$). The DISTRACTION and PAIN conditioning protocols significantly reduced unpleasantness ratings of both levels of painful heat stimuli (N-K, pain-lo $p<0.001$ for DISTRACTION and $p=0.002$ for PAIN, pain-hi $p<0.001$ for both protocols), while the STRESS protocol did not reduce unpleasantness ratings for either pain level to a significantly greater degree than the CONTROL protocol (N-K, $p>0.05$ for both heat pain levels). No significant effects of sex on any aspect of unpleasantness ratings modulation were detected ($p>0.05$). As was observed for the pain intensity ratings, none of the conditioning effects on unpleasantness ratings of the heat stimuli persisted for more than 10 minutes after cessation of the conditioning stimulation ($p>0.05$; data not shown).

Distraction Produced by the Conditioning Protocols

Distraction scores were evaluated using a 2-way mixed-model ANOVA (sex, protocol), which detected a significant main effect of protocol but no significant main effect of sex or protocol by sex interaction (Tables 5 and 6, Figure 4). These results provide evidence that sex differences in conditioning effects are not related to differences in perceived distraction, as distraction scores for each protocol were comparable for men and women. Posthoc t-tests (Bonferroni-adjusted) found that distraction scores for the CONTROL protocol were significantly lower than for all the non-control conditioning protocols (DISTRACTION $t=-9.7$, $p<0.001$; STRESS $t=-6.0$, $p<0.001$; PAIN $t=-9.1$, $p<0.001$). These findings confirm the design assumptions that the non-control protocols would be perceived as significantly more distracting than the CONTROL protocol and that distraction is an inherent component of both stressful and painful conditioning stimuli. While significantly more distracting than CONTROL, the STRESS protocol was significantly less distracting than the DISTRACTION protocol ($t=3.8$, $p<0.001$).

Stress Reactivity to the Conditioning Protocols

Stress reactivity to each conditioning protocol was calculated as the difference between the mean value of the stress measure during the Conditioning phase and the mean value of the stress measure during the pre- and post-Conditioning rest periods (Figure 1B). Stress reactivity was calculated for all three stress measures collected (self-reported stress, skin conductance, peripheral pulse rate; Table 5, Figure 5). Each measure of stress reactivity was analyzed separately using a 2-way mixed-model ANOVA (sex, protocol). In addition, the rest period values used to calculate stress reactivity were analyzed using separate 2-way mixed-model ANOVAs (sex, protocol); no significant main effects or interactions were found (all $p>0.05$), demonstrating that the stress reactivity scores are not confounded by differences in baseline stress values across groups or conditioning protocols.

Self-reported Stress—Analysis of changes in perceived stress evoked by each conditioning protocol found a significant main effect of protocol but no significant main effect of sex or protocol-by-sex interaction (Tables 5 and 6, Figure 5A). Posthoc t-tests (Bonferroni-adjusted) found that the STRESS and PAIN protocols were significantly more stressful than the CONTROL and DISTRACTION protocols (STRESS $t=-5.9$, $p<0.001$; PAIN $t=-11$, $p<0.001$ vs. CONTROL) (Figure 5A). The CONTROL and DISTRACTION protocols produced minimal, comparable changes in perceived stress (DISTRACTION $t=1.3$, $p=0.2$ vs.

CONTROL). These findings confirm the design assumptions that the DISTRACTION protocol would not be stressful and that stress is an inherent component of painful conditioning stimuli. However, the PAIN protocol was perceived as significantly more stressful than the STRESS protocol ($t=-8.1$, $p<0.001$).

Skin conductance changes—Results from the analysis of changes in skin conductance evoked by each conditioning protocol parallel the findings for changes in perceived stress, with a significant main effect of protocol but no significant main effect of sex or protocol-by-sex interaction (Tables 5 and 6, Figure 5B). STRESS and PAIN produced significantly greater increases in skin conductance than the CONTROL and DISTRACTION protocols (STRESS $t=-5.7$, $p<0.001$; PAIN $t=-9.2$, $p<0.001$ vs. CONTROL), with the CONTROL and DISTRACTION protocols producing minimal, comparable changes in skin conductance (DISTRACTION $t=-2.6$, $p=0.01$ vs. CONTROL; not significant with Bonferroni adjustment). As with the self-report measure of stress, the PAIN protocol produced significantly greater increases in skin conductance than the STRESS protocol ($t=5.632$, $p<0.001$).

Peripheral pulse rate changes—Analysis of changes in peripheral pulse rate evoked by each conditioning protocol revealed no significant main effects of protocol or sex and no significant protocol-by-sex interaction (Tables 5 and 6, Figure 5C). Thus, none of the non-control conditioning protocols produced changes in peripheral pulse rate beyond those produced by the CONTROL protocol. Together with preliminary data (not shown) that found no changes in blood pressure in response to the various conditioning protocols, these results suggest that a 10-minute application of transcutaneous electrical nerve stimulation does not influence cardiovascular measures, even when delivered at strongly painful intensities.

State Anxiety and Mood Changes Produced by the Conditioning Protocols

Changes in state anxiety, positive affect, and negative affect evoked by each conditioning protocol (pre- minus post-Conditioning phase scores, Figure 1B) were evaluated using separate 2-way mixed-model ANOVAs (sex, protocol; Tables 5 and 6). The only significant finding from this analysis was a greater change in state anxiety during the PAIN conditioning protocol than the CONTROL protocol ($t=-3.3$, $p=0.004$). No other significant effects of protocol or sex on anxiety or mood variables were detected. Baseline (pre-Conditioning phase) scores for state anxiety, positive affect, and negative affect also did not differ significantly between groups or across conditioning protocols ($p>0.05$ for all effects).

Correlation Analyses

The magnitude of the PAIN conditioning effect on pain-hi ratings was significantly negatively correlated with distraction scores in men (Figure 6A), with individuals who rated the painful conditioning stimulation as more distracting showing greater reductions in pain-hi ratings. A non-significant trend in the same direction ($p=0.15$) was observed for women (Figure 6B). For pain-hi ratings, the magnitude of the PAIN conditioning effect was also significantly negatively correlated with changes in perceived stress in men (Figure 6C), with more stress-reactive individuals showing greater reductions in pain-hi ratings. A non-significant trend ($p=0.17$) for an association between these variables was observed in women, though in the opposite direction (Figure 6D), with less stress-reactive individuals showing greater reductions in pain-hi ratings. Other stress reactivity measures (skin conductance, pulse rate) did not correlate significantly with PAIN conditioning effects on pain-hi ratings for either sex ($p>0.05$). For the pain-lo ratings, the magnitude of the PAIN effect was not significantly correlated with distraction or any measure of stress reactivity in either men or women ($p>0.05$).

Multiple Linear Regression Analyses

Exploratory regression analyses conducted separately for each sex found that for pain-hi ratings, distraction and self-reported stress reactivity significantly predicted the PAIN conditioning effect magnitude in both men and women, with stress reactivity having an effect in opposite directions between the sexes (Table 7); these results parallel those from the correlation analyses. Skin conductance stress reactivity was not significantly predictive of conditioning effect magnitude and was therefore excluded from further regression analyses.

Based on the results of the exploratory regressions, a full regression model was run on the PAIN conditioning effect magnitude data for the combined groups to test the predictive value of distraction, self-reported stress reactivity, the distraction-by-sex interaction, and the stress-by-sex interaction. For pain-hi stimuli, this model explained a significant amount of the variance in the PAIN conditioning effects (Table 7) and confirmed the exploratory regression findings that distraction and self-reported stress reactivity are significant predictors of these measures. Furthermore, the model detected a significant stress-by-sex interaction (Table 7), demonstrating a significant sex difference in the contribution of stress to the modulatory effect of painful conditioning stimulation on pain-hi ratings. However, when the model was regressed on the magnitude of the PAIN conditioning effect on pain-lo ratings, it did not explain a significant amount of variance ($p=0.35$), and none of the modeled variables were significantly predictive of the effects ($p>0.2$), though the stress-by-sex interaction showed a non-significant trend that paralleled the significant effect found for the pain-hi ratings ($p=0.1$).

DISCUSSION

The present study found evidence for quantitative and qualitative sex differences in the function of endogenous pain modulatory systems in healthy subjects. The male and female groups in this study were homogeneous with respect to most demographic, lifestyle, psychosocial, and physiological variables assessed. Sex differences in some measures (such as blood pressure, anxiety, emotional reactivity, pain coping strategies, heat stimulus temperatures, and electrical stimulation current) were found and might contribute to differences in pain modulation. However, none of these variables were consistently correlated with the hypoalgesic effects, suggesting they are unlikely to play a major role in sex differences in endogenous pain modulation in this study.

Distraction-evoked hypoalgesia

In response to comparably distracting conditioning stimuli, the magnitude of distraction-evoked hypoalgesia was significantly greater in men than women for the sensory dimension of pain, with a nonsignificant trend in the same direction for affective responses. Consistent with these findings are reports that males show greater modulation of cold pain threshold and tolerance (Weisenberg et al. 1995; Unrod et al. 2004) and suprathreshold cold pain intensity and unpleasantness ratings (Bentsen et al. 1999) in response to visual-cognitive distractors such as film viewing. Keogh and Eccleston (2006) report that among adolescent chronic pain patients, males use distraction as a coping strategy to a significantly greater degree than females. Given that distraction reduces experimental pain to a greater degree in men than women and that males voluntarily use more distraction-based coping strategies than females, it is likely that distraction is a more efficacious pain reduction technique in men than women. Studies of sex differences in distraction effects on clinical pain are lacking but are needed to assess whether distraction-based pain-reduction techniques commonly used in clinical practice (Weisenberg 1999) are appropriate for women.

Additional research is also needed to elucidate mechanisms underlying sex differences in distraction-evoked hypoalgesia, which this psychophysical study cannot address. Distraction

is a cognitive manipulation hypothesized to produce hypoalgesia by introducing stimuli that compete with noxious stimuli for attentional resources (Fernandez and Turk 1989); thus, distraction may primarily modulate nociception at the level of the cerebral cortex. Consistent with this hypothesis, functional neuroimaging studies found that reduction of pain perception by distraction is associated with reduction of pain-related activity in multiple cortical regions involved in nociceptive processing (Bushnell et al. 1999; Petrovic et al. 2000; Frankenstein et al. 2001; Bantick et al. 2002). However, these studies do not definitively demonstrate that distraction alters pain perception by acting solely at cortical sites. Structures at several levels of the neuraxis are potentially involved in distraction-evoked modulation of pain-related neuronal activity, including the lateral orbitofrontal cortex (Petrovic et al. 2000; Bantick et al. 2002), periaqueductal gray (Tracey et al. 2002), and descending circuits that inhibit nociceptive processing at the level of the spinal cord (Hayes et al. 1981; Bushnell et al. 1984). Future work is needed to elucidate the specific mechanisms involved in distraction-evoked hypoalgesia and to determine whether sex differences in this effect result from differential activation of a common modulatory circuit or whether sex-specific modulatory mechanisms exist.

Pain-evoked hypoalgesia

Painful conditioning stimuli reduced heat pain perception to a comparable degree in both men and women, a finding consistent with previous studies that used acutely painful test stimuli (France and Suchowiecki 1999; Ge et al. 2004; Pud et al. 2005). The present study used a large sample and controlled for various potentially confounding variables that were not addressed in earlier studies (such as menstrual cycle phase, psychosocial factors, and the magnitude of pain produced by the conditioning stimuli) and therefore provides strong support for the published findings. In contrast, painful conditioning stimulation has been reported to reduce temporally-summed pain to a greater degree in men than women (Staud et al. 2003; Serrao et al. 2004), suggesting sex differences in pain-evoked hypoalgesia may emerge for some specific aspects of pain.

Sex differences in the duration of pain-evoked hypoalgesia were reported by Ge et al. (2004), with men demonstrating significantly longer-lasting hypoalgesic effects than women in response to intramuscular hypertonic saline injection as the painful conditioning stimulation. In this study, pain-evoked hypoalgesia did not persist in either sex more than 10 minutes after cessation of the conditioning stimuli, consistent with earlier work that examined the question of hypoalgesic duration (Price and McHaffie 1988; Willer et al. 1990). Inconsistencies between this study and that of Ge et al. may reflect engagement of different modulatory mechanisms by the different conditioning protocols. For example, hypoalgesic duration could vary markedly based on whether opioid- or nonopioid-mediated mechanisms are engaged; evidence for both has been found in humans (Willer et al. 1990; Edwards et al. 2004).

A novel finding of this study is that distraction and stress produced by painful conditioning stimulation were significant predictors of the magnitude of the modulatory effect on strongly painful heat in both men and women, supporting the hypothesis that distraction and stress are mediators of pain-evoked hypoalgesia. These factors did not account entirely for the variance in pain-evoked hypoalgesia, suggesting that the painfulness of the conditioning stimuli, independent of distraction and stress, also contributed to the hypoalgesic effects. Animal studies suggest that distinct neural mechanisms underlie antinociceptive effects of stressful (Basbaum and Fields 1984) and noxious (Le Bars 2002) conditioning stimulation, and, as discussed earlier, neuroimaging evidence suggests a primarily cortical modulatory mechanism for distraction effects on pain. Thus, pain-evoked hypoalgesia is a complex phenomenon that likely reflects the combined effects of activation of multiple endogenous pain inhibitory systems, an important consideration for investigators who use it to assess endogenous analgesic system function. Functional neuroimaging studies of pain-evoked hypoalgesia, particularly

those that capture neural activity at multiple levels of the nociceptive system, are lacking but are needed to characterize mechanisms underlying this phenomenon in humans.

Furthermore, while the overall magnitude of pain-evoked hypoalgesia was comparable between men and women, the stressfulness of the painful conditioning stimulation contributed to the hypoalgesic effect differently by sex. Among men, the more stressful the painful conditioning stimuli were perceived, the greater the hypoalgesic effect; this relationship is consistent with most animal and human literature on stress-induced analgesia (Craft 2003). Among women the opposite relationship was observed: the more stressful the painful conditioning stimuli were perceived, the smaller the hypoalgesic effect. This may reflect an important female-specific response to acutely painful stimuli in which women with greater stress reactivity to pain associated with illness and injury may be less able to activate endogenous pain inhibitory systems, putting them at greater risk of progressing from an acute to chronic pain state. Thus, though speculative, this study suggests that individual differences in stress reactivity combined with sex characteristics may be important risk factors for the development of chronic pain conditions. Clinical research, particularly long-term prospective studies (as recommended by Edwards (2005)), is needed to explore these potential risk factors. An important caveat is that distraction and stress were significantly predictive of pain-evoked hypoalgesia for strongly painful, but not less painful test stimuli. Other than the fact that pain-lo ratings were generally more variable than pain-hi ratings, there is no clear reason why distraction and stress would impact hypoalgesia differently for the two pain levels. One possibility is that endogenous analgesic systems may function differently depending upon the strength of the nociceptive input or the level of pain being modulated, though no reports have yet addressed this theory.

Stress-evoked hypoalgesia

Minimal stress-evoked hypoalgesia was observed in this study, possibly because the stressful conditioning stimuli were not sufficiently stressful to activate endogenous analgesic systems. Only pain-lo intensity ratings were significantly modulated by the stressful protocol, and these effects may be attributable to the distracting aspects of the stimuli, which were significant (though to a lesser degree than the distracting or painful conditioning stimuli). Most previous studies of sex differences in stress-evoked hypoalgesia use cognitive or social stressors (Rhudy and Meager 2001; Sternberg et al. 2001; Girdler et al. 2005), and a goal of this study was to evoke stress with somatosensory stimuli similar to those used to evoke pain-evoked hypoalgesia to enhance comparability of the two forms of pain modulation. However, the technical challenges associated with selecting stimuli that are stressful without being painful were not surmounted, and this study does not provide data sufficient to evaluate sex differences in stress-evoked hypoalgesia. Future studies of this topic are needed, particularly in light of the sex differences in the contribution of stress to pain-evoked hypoalgesia reported in this study.

Conclusions

In summary, the present study provides evidence for sex differences in the function of endogenous pain modulatory systems in healthy human subjects. Specifically, sex differences in the magnitude of distraction-evoked hypoalgesia and the mechanisms underlying pain-evoked hypoalgesia were found. Furthermore, this study supports the theory that pain-evoked hypoalgesia is a complex phenomenon that reflects engagement of multiple neural systems, each with the potential to be differentially influenced by sex. Sex differences in the function of endogenous pain modulatory systems may be important contributors to greater pain sensitivity and higher prevalence of many chronic pain conditions among women (Fillingim and Maixner 1995; Berkley 1997; Fillingim 2000).

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References

- Baad-Hansen L, Poulsen HF, Jensen HM, Svensson P. Lack of sex differences in modulation of experimental intraoral pain by diffuse noxious inhibitory controls (DNIC). *Pain* 2005;116:359–365. [PubMed: 15979791]
- Bantick SJ, Wise RG, Ploghaus A, Clare S, Smith SM, Tracey I. Imaging how attention modulates pain in humans using functional MRI. *Brain* 2002;125:310–319. [PubMed: 11844731]
- Basbaum AI, Fields HL. Endogenous pain control systems: Brainstem spinal pathways and endorphin circuitry. *Annu Rev Neurosci* 1984;7:309–338. [PubMed: 6143527]
- Beck AT, Ward CH, Mendelson M, Mock JE, Erbaugh JK. An inventory for measuring depression. *Arch Gen Psychiatry* 1961;4:561–571. [PubMed: 13688369]
- Bentsen B, Svensson P, Wenzel A. The effect of a new type of video glasses on the perceived intensity of pain and unpleasantness evoked by a cold pressor test. *Anesth Prog* 1999;46:113–117. [PubMed: 11692351]
- Berkley KJ. Sex differences in pain. *Behav Brain Sci* 1997;20:371–380. [PubMed: 10097000]
- Bingel U, Schoell E, Herken W, Buchel C, May A. Habituation to painful stimulation involves the antinociceptive system. *Pain*. 2007. in press, corrected proof online (<http://www.sciencedirect.com/science/article/B6T0K-4MX54SN-1/2/4ff2a0f00caef8e53d92059d757a101c>)
- Bushnell MC, Duncan GH, Dubner R, He LF. Activity of trigeminothalamic neurons in medullary dorsal horn of awake monkeys trained in a thermal discrimination task. *J Neurophysiol* 1984;52:170–187. [PubMed: 6747675]
- Bushnell MC, Duncan GH, Hofbauer RK, Ha B, Chen JI, Carrier B. Pain perception: Is there a role for primary somatosensory cortex? *Proc Natl Acad Sci USA* 1999;96:7705–7709. [PubMed: 10393884]
- Chesterton LS, Barlas P, Foster NE, Lundeborg T, Wright CC, Baxter GD. Sensory stimulation (TENS): effects of parameter manipulation on mechanical pain thresholds in healthy human subjects. *Pain* 2002;99:253–262. [PubMed: 12237203]
- Chesterton LS, Foster NE, Wright CC, Baxter GD, Barlas P. Effects of TENS frequency, intensity and stimulation site parameter manipulation on pressure pain thresholds in healthy human subjects. *Pain* 2003;106:73–80. [PubMed: 14581113]
- Coghill RC, McHaffie JG, Yen Y-F. Neural correlates of interindividual differences in the subjective experience of pain. *Proc Natl Acad Sci USA* 2003;100:8538–8542. [PubMed: 12824463]
- Craft RM. Sex differences in drug- and non-drug-induced analgesia. *Life Sci* 2003;72:2675–2688. [PubMed: 12679185]
- Edwards RR. Individual differences in endogenous pain modulation as a risk factor for chronic pain. *Neurology* 2005;65:437–443. [PubMed: 16087910]
- Edwards RR, Fillingim RB, Ness TJ. Age-related differences in endogenous pain modulation: A comparison of diffuse noxious inhibitory controls in healthy older and younger adults. *Pain* 2003a;101:155–165. [PubMed: 12507710]
- Edwards RR, Ness TJ, Fillingim RB. Endogenous opioids, blood pressure, and diffuse noxious inhibitory controls: a preliminary study. *Percept Mot Skills* 2004;99:679–687. [PubMed: 15560360]
- Edwards RR, Ness TJ, Weigent DA, Fillingim RB. Individual differences in diffuse noxious inhibitory controls (DNIC): association with clinical variables. *Pain* 2003b;106:427–437. [PubMed: 14659526]
- Fernandez E, Turk DC. The utility of cognitive coping strategies for altering pain perception: A meta-analysis. *Pain* 1989;38:123–135. [PubMed: 2674861]
- Fields HL, Basbaum AI. Central nervous systems of pain modulation. In: Wall, PD, Melzack, R., editors. *Textbook of Pain*. 4. Edinburgh:Churchill Livingstone: 1999. p. 309-329.
- Fillingim RB. Sex, gender, and pain: A biopsychosocial framework. In: Fillingim, RB., editor. *Sex, Gender, and Pain*. Seattle: IASP Press; 2000. p. 1-6.

- Fillington RB, Maixner W. Gender differences in the responses to noxious stimuli. *Pain Forum* 1995;4:209–221.
- France CR, Suchowiecki S. A comparison of diffuse noxious inhibitory controls in men and women. *Pain* 1999;81:77–84. [PubMed: 10353495]
- Frankenstein UN, Richter W, McIntyre MC, Remy F. Distraction modulates anterior cingulate gyrus activations during the cold pressor test. *Neuroimage* 2001;14:827–836. [PubMed: 11554801]
- Ge HY, Madeleine P, Arendt-Nielsen L. Sex differences in temporal characteristics of descending inhibitory control: an evaluation using repeated bilateral experimentation of muscle pain. *Pain* 2004;110:72–78. [PubMed: 15275754]
- Gibson, RH. Electrical stimulation of pain and touch. In: Kenshalo, DL., editor. *The Skin Senses*. Springfield IL: CC Thomas; 1968. p. 223–258.
- Girdler SS, Maixner W, Naftel HA, Steward PW, Moretz RL, Light KC. Cigarette smoking, stress-induced analgesia, and pain perception in men and women. *Pain* 2005;114:372–385. [PubMed: 15777863]
- Goolkasian P. Cyclic changes in pain perception: an ROC analysis. *Percept Psychophys* 1980;27:499–504. [PubMed: 7393696]
- Hayes RL, Price DD, Ruda MA, Dubner R. Neuronal activity in medullary dorsal horn of awake monkeys trained in a thermal discrimination task. II. Behavioral modification of responses to thermal and mechanical stimuli. *J Neurophys* 1981;46:428–443.
- Julien N, Goffaux P, Arsenault P, Marchand S. Widespread pain in fibromyalgia is related to a deficit of endogenous pain inhibition. *Pain* 2005;114:295–302. [PubMed: 15733656]
- Kashima K, Rahman OI, Sakoda S, Shiba R. Increased pain sensitivity of the upper extremities of TMD patients with myalgia to experimentally-evoked noxious stimulation: possibility of worsened endogenous opioid systems. *Cranio* 1999;17:241–246. [PubMed: 10650395]
- Keogh E, Eccleston C. Sex differences in adolescent chronic pain and pain-related coping. *Pain* 2006;123:275–284. [PubMed: 16644131]
- Kohn, PM. Sensation-seeking, augmenting-reducing, and strength of the nervous system. In: Spence, JT.; Izard, CE., editors. *Motivation, emotion, and personality*. Amsterdam: Elsevier; 1985.
- Koltyn KF, Trine MR, Stegner AJ, Tobar DA. Effect of isometric exercise on pain perception and blood pressure in men and women. *Med Sci Sports Exerc* 2001;33:282–290. [PubMed: 11224819]
- Kosek E, Hansson P. Modulatory influence on somatosensory perception from vibration and heterotopic noxious conditioning stimulation (HNCS) in fibromyalgia patients and healthy subjects. *Pain* 1997;70:41–51. [PubMed: 9106808]
- Kowalczyk WJ, Evans SM, Bisaga AM, Sullivan MA, Comer SD. Sex differences and hormonal influences on response to cold pressor pain in humans. *J Pain* 2006;7:151–160. [PubMed: 16516820]
- Lautenbacher S, Kundermann B, Krieg JC. Sleep deprivation and pain perception. *Sleep Med Rev* 2006;10:357–369. [PubMed: 16386930]
- Lautenbacher S, Rollman GB. Possible deficiencies of pain modulation in fibromyalgia. *Clin J Pain* 1997;13:189–196. [PubMed: 9303250]
- Le Bars D. The whole body receptive field of dorsal horn multireceptive neurones. *Brain Res Rev* 2002;40:29–44. [PubMed: 12589904]
- Morin, CM. *Insomnia: Psychological Assessment and Management*. New York: Guilford Press; 1993.
- Naliboff BD, Benton D, Solomon GF, Morley JE, Fahey JL, Bloom ET, Makinodan T, Gilmore SL. Immunological changes in young and old adults during brief laboratory stress. *Psychosom Med* 1991;53:121–132. [PubMed: 2031066]
- Onen SH, Alloui A, Gross A, Eschallier A, Dubray C. The effects of total sleep deprivation, selective sleep interruption and sleep recovery on pain tolerance thresholds in healthy subjects. *J Sleep Res* 2001;10:35–42. [PubMed: 11285053]
- Petrovic P, Petersson KM, Ghatan PH, Stone-Elander S, Ingvar M. Pain-related cerebral activation is altered by a distracting cognitive task. *Pain* 2000;85:19–30. [PubMed: 10692599]
- Price DD, Bush FM, Long S, Harkins SW. A comparison of pain measurement characteristics of mechanical and visual analogue and simple numerical rating scales. *Pain* 1994;56:217–226. [PubMed: 8008411]

- Price DD, McGrath PA, Rafii A, Buckingham B. The Validation of Visual Analog Scales as Ratio Scale Measures for Chronic and Experimental Pain. *Pain* 1983;17:45–56. [PubMed: 6226917]
- Price DD, McHaffie JG. Effects of heterotopic conditioning stimuli on first and second pain: a psychophysical evaluation in humans. *Pain* 1988;34:245–252. [PubMed: 3186271]
- Pud D, Sprecher E, Yarnitsky D. Homotopic and heterotopic effects of endogenous analgesia in healthy volunteers. *Neurosci Lett* 2005;380:209–213. [PubMed: 15862887]
- Rhudy JL, Meagher MW. Noise stress and human pain thresholds: Divergent effects in men and women. *J Pain* 2001;2:57–64. [PubMed: 14622786]
- Riley JL III, Robinson ME, Wise EA, Price DD. A meta-analytic review of pain perception across the menstrual cycle. *Pain* 1999;81:225–235. [PubMed: 10431710]
- Rosenstiel AK, Keefe FJ. The use of coping strategies in chronic low back pain patients: Relationship to patient characteristics and current adjustment. *Pain* 1983;17:33–44. [PubMed: 6226916]
- Sarlani E, Grace EG, Reynolds MA, Greenspan JD. Sex differences in temporal summation of pain and aftersensations following repetitive noxious mechanical stimulation. *Pain* 2004;109:115–123. [PubMed: 15082133]
- Serrao M, Rossi P, Sandrini G, Parisi L, Amabile GA, Nappi G, Pierelli F. Effects of diffuse noxious inhibitory controls on temporal summation of the RIII reflex in humans. *Pain* 2004;112:353–360. [PubMed: 15561391]
- Smith MT, Edwards RR, McCann U, Haythornthwaite J. The effects of sleep deprivation on pain inhibition and spontaneous pain in women. *Sleep* 2007;30(4):494–505. [PubMed: 17520794]
- Spielberger, CD.; Gorsuch, RL.; Lushene, R. The state-trait anxiety inventory manual. Palo Alto, CA: Consulting Psychology Press; 1970.
- Strian F, Lautenbacher S, Galfe G, Holzl R. Diurnal variations in pain perception and thermal sensitivity. *Pain* 1989;36:125–131. [PubMed: 2919090]
- Staud R, Robinson ME, Vierck CJ Jr, Price DD. Diffuse noxious inhibitory controls (DNIC) attenuate temporal summation of second pain in normal males but not in normal females or fibromyalgia patients. *Pain* 2003;101:167–174. [PubMed: 12507711]
- Sternberg WF, Bokar C, Kass L, Albayadjan A, Gracely RH. Sex-dependent components of the analgesia produced by athletic competition. *J Pain* 2001;2:65–74. [PubMed: 14622787]
- Tracey I, Ploghaus A, Gati JS, Clare S, Smith S, Menon RS, Matthews PM. Imaging attentional modulation of pain in the periaqueductal gray in humans. *J Neurosci* 2002;22:2748–2752. [PubMed: 11923440]
- Unrod M, Kassel JD, Robinson M. Effects of smoking, distraction, and gender on pain perception. *Behav Med* 2004;30:133–139. [PubMed: 15816316]
- Watson D, Clark LA, Tellegen A. Development and validation of brief measures of positive and negative affect: The PANAS Scales. *J Pers Soc Psychol* 1988;47:1063–1070. [PubMed: 3397865]
- Weisenberg M, Tepper I, Schwarzwald J. Humor as a cognitive technique for increasing pain tolerance. *Pain* 1995;63:207–212. [PubMed: 8628586]
- Weisenberg, M. Cognitive aspects of pain. In: Wall, PD.; Melzack, R., editors. *Textbook of Pain*. 4. Edinburgh:Churchill Livingstone: 1999. p. 345–359.
- Wilder-Smith CH, Schindler D, Lovblad K, Redmond SM, Nirkko A. Brain functional magnetic resonance imaging of rectal pain and activation of endogenous inhibitory mechanisms in irritable bowel syndrome patient subgroups and healthy controls. *Gut* 2004;53:1595–1601. [PubMed: 15479679]
- Willer JC, Boreau F, Albe-Fessard D. Supraspinal influences on nociceptive flexion reflex and pain sensation in man. *Brain Res* 1979;179:61–68. [PubMed: 509233]
- Willer JC, Le Bars D, De Broucker T. Diffuse noxious inhibitory controls in man: involvement of an opioidergic link. *Eur J Pharmacol* 1990;182:347–355. [PubMed: 2168836]
- Zhang B, Wing YK. Sex differences in insomnia: a meta-analysis. *Sleep* 2006;29:85–93. [PubMed: 16453985]

Figure 2. Mean magnitude of conditioning effects (+SE) on pain intensity ratings for (A) pain-hi and (B) pain-lo heat stimuli. *denotes significant differences in post-hoc comparisons at the $p < 0.05$ level.

Figure 3. Mean magnitude of conditioning effects (+SE) on unpleasantness ratings for (A) pain-hi and (B) pain-lo heat stimuli. *denotes significant differences in post-hoc comparisons at the $p < 0.05$ level.

Figure 4. Mean distraction scores (+SE) for each conditioning protocol. ***denotes significant differences in post-hoc comparisons at the $p < 0.001$ level.

Figure 5. Mean stress reactivity (+SE) evoked by the various conditioning protocols, as measured by change in (A) self-reported stress, (B) skin conductance, and (C) peripheral pulse rate. ***denotes significant differences in post-hoc comparisons at the $p < 0.001$ level.

Figure 6.

Correlation between the effects of painful conditioning stimulation on pain-hi ratings and distraction evoked by the painful conditioning stimulation for (A) males and (B) females. Correlation between the effects of painful conditioning stimulation effect on pain-hi ratings and self-reported stress changes evoked by the painful conditioning stimulation for (C) males and (D) females.

Table 1

Group characteristics

Variable	Female (n=30)	Male (n=32)	Statistic	p-value
Age	26.1 (2.4)	26.0 (2.7)	MWU Z = -0.4	0.7
Race	60%	59%		
White	13%	13%		
Asian	13%	13%		
Hispanic	7%	6%		
S. Asian Indian	3%	6%		
Black	3%	3%		
American Indian				
Exercise (sessions per week)	2.6 (2)	2.5 (2)	MWU Z = -0.01	0.99
Sleep (hours per night)	7.3 (0.9)	7.3 (0.7)	MWU Z = -0.6	0.5
Caffeine use (16 oz beverages/day)	1.3 (1)	0.84 (1)	MWU Z = -2.1	0.04 *
Alcohol use (drinks/week)	3.2 (4)	3 (6)	MWU Z = -2.1	0.04 *
Tobacco use (cigarettes/day)	0.2 (0.8)	0.9 (3)	MWU Z = -1.1	0.3
Systolic blood pressure	116 (10)	126 (17)	t=2.5	0.01 *
Diastolic blood pressure	70 (7)	71 (6)	t=-0.07	0.5
Trait anxiety	37 (7)	32 (6)	t=2.6	0.01 *
Trait positive affect	35.1 (6)	35 (5)	t=0.02	0.98
Trait negative affect	17 (5)	16 (6)	MWU Z=-0.9	0.4
Depression	3.8 (3.7)	3.1 (2.9)	MWU Z=-0.3	0.7
Emotional reactivity	79 (57)	63 (10)	MWU Z=-2.4	0.02 *
Insomnia	4.3 (4.2)	4.3 (3.2)	MWU Z=-0.6	0.5
Coping strategies				
Diverting attention	12 (8)	7.5 (7)	MWU Z=-2.3	0.02 *
Reinterpreting pain sensation	6.8 (8)	4.4 (6)	MWU Z=-1.3	0.2
Coping self-statements	20 (8)	16 (9)	MWU Z=-1.8	0.07
Ignoring pain	18 (8)	16 (8)	MWU Z=-1.0	0.3
Praying/hoping	6.2 (7)	6 (7)	MWU Z=-0.9	0.4
Catastrophizing	3.8 (4)	4.4 (7)	MWU Z=-0.7	0.5
Increasing activity	14 (7)	8.8 (7)	MWU Z=-2.9	0.004 *
Increasing pain behavior	17 (6)	11 (6)	MWU Z=-4.1	<0.001 *
Perceived ability to control pain	4.4 (1)	3.7 (1)	MWU Z=-2.1	0.04 *
Perceived ability to decrease pain	4.0 (1)	3.2 (1)	MWU Z=-2.1	0.04 *
Gender role expectations of pain				
Pain sensation (male> female)	51 (20)	49 (18)	t=0.33	0.75
Pain endurance (male>female)	50 (18)	58 (18)	t=-1.8	0.08
Willingness to report pain (male>female)	37 (18)	30 (19)	t=1.6	0.11
Perceived health score (GHQ)	25 (4)	27 (4)	t=2.6	0.01 *

Values are means (SD)

* Statistically significant difference

Table 2

Stimulation parameters

Variable	Female (n=30)	Male (n=32)
CONTACT HEAT STIMULI		
Temperature, pain-hi	48.5 (1.1)	49.1 (1.1)
Temperature, pain-lo	47.4 (1.1)	48.1 (1.1)
Pain intensity ratings, pain-hi	72.5 (10)	68.2 (10)
Pain intensity ratings, pain-lo	42.9 (15)	42.9 (14)
Pain intensity ratings of 48°C stimulus	62.2 (23)	52.2 (22)
ELECTRICAL STIMULATION PARAMETERS		
Electrical current amplitude: Control condition	9.3 (2)	12 (3)
Electrical current amplitude: Distraction condition	8.9 (2)	11 (3)
Electrical current amplitude: Stress condition	20 (8)	25 (10)
Electrical current amplitude: Pain condition	35 (12)	45 (15)
Sensation intensity ratings: Control condition	7.7 (2)	7.8 (3)
Sensation intensity ratings: Distraction condition	9.7 (2)	9.8 (3)
Sensation intensity ratings: Stress condition	35 (5)	35 (4)
Sensation intensity ratings: Pain condition	72 (3)	72 (5)

Values are means (SD)

Table 3

Stimulation Parameters: ANOVA Results

Variable	Factor	df	F	p	Post-hoc comparisons**
Heat pain temperatures	Pain level	1	980	<0.001*	Pain level: lo < high Sex: F < M
	Sex	1	5.4	0.02*	
	Interaction	1	2.2	0.15	
Heat pain intensity ratings	Pain level	1	200	<0.001*	Pain level: lo < high
	Sex	1	0.7	0.4	
	Interaction	1	1.2	0.3	
Electrical current amplitude	Protocol	3	300	<0.001*	Protocol: C/D < S (p<0.001) and S < P (p<0.001) Sex: F < M (C: p<0.001; D p=0.001; S p=0.04; P p=0.007)
	Sex	1	9.1	0.004*	
	Interaction	3	4.4	0.005*	
Electrical intensity ratings	Protocol	3	4,800	<0.001*	Protocol: C < D < S < P (p<0.001 for all comparisons)
	Sex	1	0.03	0.9	
	Interaction	3	0.1	0.95	

* Statistically significant difference

** C=control conditioning protocol; D=distracting protocol; S=stressful protocol; P=painful protocol

Table 4

Conditioning protocol effects on heat pain intensity and unpleasantness ratings

Magnitude of hypoalgesic effect*	Control	Distraction	Stress	Pain
Pain intensity ratings: pain-hi				
female (n=30)	2.1 (14)	-11 (18)**	-1.3 (14)	-12.4 (13)
male (n=32)	-4.6 (12)	-19 (16)**	-7.6 (17)	-10.5 (14)
Pain intensity ratings: pain-lo				
female (n=30)	1.8 (17)	-6.2 (23)**	-8.3 (21)	-9.3 (13)
male (n=32)	1.0 (16)	-16 (14)**	-8.5 (15)	-7.8 (17)
Unpleasantness ratings: pain-hi				
female (n=30)	-0.5 (15)	-18 (22)	-3.7 (18)	-11 (18)
male (n=32)	-2.5 (13)	-24 (22)	-7.8 (16)	-13 (19)
Unpleasantness ratings: pain-lo				
female (n=30)	-2.9 (17)	-8.5 (26)	-6.3 (20)	-8.5 (14)
male (n=32)	-5.6 (16)	-17 (21)	-8.0 (16)	-11 (14)

Values are means (SD)

* Magnitude calculated as difference between Baseline phase pain ratings and Conditioning phase pain ratings

** Statistically significant sex difference (p=0.004).

Table 5

Distraction scores, stress measures, and measures of state anxiety and affect associated with the conditioning protocols

	Control	Distraction	Stress	Pain
Distraction score				
female (n=30)	19 (20)	52 (26)	34 (22)	47 (24)
male (n=32)	19 (21)	50 (25)	40 (25)	45 (26)
Self-reported stress (change)¹				
female (n=30)	3.9 (7)	3.8 (7)	10.4 (8)	23 (14)
male (n=32)	2.9 (5)	1.4 (4)	8.8 (9)	18 (15)
Skin conductance (change)²				
female (n=29)	-0.02 (0.7)	0.11 (0.6)	0.59 (1)	1.8 (1)
male (n=31)	0.02 (0.7)	0.55 (0.8)	0.91 (1)	1.7 (2)
Pulse rate (change)²				
female (n=28)	-0.9 (3)	-0.9 (3)	-1.7 (3)	-0.06 (4)
male (n=30)	-0.9 (6)	0.1 (4)	0.7 (5)	0.8 (3)
State anxiety (change)³				
female (n=30)	0.73 (4)	-0.17 (4)	1.2 (4)	4.2 (6)
male (n=32)	0.44 (5)	0.88 (5)	2.9 (4)	3.6 (5)
State positive affect (change)³				
female (n=30)	-0.53 (4)	-0.4 (3)	-1.2 (3)	-1.2 (4)
male (n=32)	0.28 (2)	0.16 (4)	-1.5 (4)	0.06 (3)
State negative affect (change)³				
female (n=30)	0.43 (2)	0.08 (2)	-0.1 (2)	0.65 (2)
male (n=32)	-0.06 (2)	-0.06 (1)	0.9 (2)	0.84 (1)

Values are means (SD)

¹ Conditioning phase ratings (mean) minus mean ratings obtained during pre- and post-conditioning phase rest intervals

² Measures (mean) obtained during first four minutes of conditioning phase, minus mean measures obtained during pre-and post-conditioning rest intervals. Measures were not obtained from a few subjects due to equipment problems.

³ State measure obtained immediately following conditioning phase, minus measure obtained prior to conditioning phase

Table 6
ANOVA Results for distraction scores, stress measures, and measures of anxiety and affect associated with the conditioning protocols

Variable	Factor	df	F	p	Post-hoc comparisons**
Distraction score	Protocol	3	33	<0.001*	Protocol: C < S < D/P
	Sex	1	0	0.96	
	Interaction	3	0.5	0.7	
Self-reported stress (change)	Protocol	3	84	<0.001*	Protocol: C/D < S < P
	Sex	1	1.7	0.2	
	Interaction	3	0.6	0.5	
Skin conductance (change)	Protocol	3	44	<0.001*	Protocol: C/D < S < P
	Sex	1	1.1	0.3	
	Interaction	3	1.1	0.3	
Pulse rate (change)	Protocol	3	1.4	0.3	
	Sex	1	2.6	0.1	
	Interaction	3	1.1	0.3	
State anxiety	Protocol	3	9.6	<0.001*	Protocol: C < P
	Sex	1	0.4	0.5	
	Interaction	3	1.1	0.3	
State positive affect	Protocol	3	1.8	0.14	
	Sex	1	1.5	0.22	
	Interaction	3	0.5	0.65	
State negative affect	Protocol	3	2.6	0.052	
	Sex	1	0.14	0.7	
	Interaction	3	2.6	0.054	

* Statistically significant difference

** C=control conditioning protocol; D=distracting protocol; S=stressful protocol; P=painful protocol

Table 7

Results of multiple linear regressions predicting the magnitude of change in pain-hi intensity ratings evoked by painful conditioning stimulation

Exploratory regression: male			
Variable	Beta	t-statistic	p-value
Distraction score	-0.4	-2.5	0.017*
Self-reported stress change	-0.35	-2.2	0.038*
Skin conductance change	-0.15	-0.93	0.37
Final model: $F(3,27)=4.9$, $p=0.007$, $R^2=0.36$			
Exploratory regression: female			
Variable	Beta	t-statistic	p-value
Distraction score	-0.42	-2.2	0.038*
Self-reported stress change	0.39	2.1	0.046*
Skin conductance change	-0.04	-0.23	0.82
Final model: $F(3,25)=2.4$, $p=0.09$, $R^2=0.22$			
Full regression model			
Variable	Beta	t-statistic	p-value
Distraction score	-0.8	-3.1	0.003*
Distraction*sex interaction	0.53	1.9	0.06
Self-reported stress change	0.71	2.1	0.04*
Stress*sex interaction	-0.8	-2.4	0.02*
Final model: $F(4,57)=3.9$, $p=0.007$, $R^2=0.22$			