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Characterization of a novel capsaicin/heat ongoing pain model

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Abstract

Background: Human experimental pain models provide an important translational link between pre-clinical models and clinical pain. Using topical capsaicin and continuous heat application, the novel capsaicin/heat ongoing pain (CHOP) model induces long-lasting experimental pain of which the perceived intensity can be individually adjusted.
Methods: In the CHOP model, capsaicin or control cream is applied to a 10 × 10 cm skin area and a heating pad is applied over the area after cream removal. Two experiments in healthy participants were performed for model characterization. In Experiment 1, a constant temperature was applied for 60 min; in Experiment 2, temperature was adjusted to maintain a constant perceived intensity for 60 min.
Results: Experiment 1: across participants, constant temperature induced initial habituation followed by an increase in sensation back to baseline. Cluster analysis revealed that half the participants sensitized to the constant temperature, while the other half did not. The degree of sensitization was related to the baseline pain unpleasantness, relative to pain intensity. Experiment 2: constant perceived intensity was achieved in the painful and a non-painful control condition. The two conditions did not differ regarding possibly confounding variables, including blood pressure, heart rate, inflammation or physiological stress as measured by surrogate markers. Secondary allodynia and hyperalgesia were reported more following painful compared to control stimulation. Sensitizers as determined in Experiment 1 were also more pain sensitive in Experiment 2.
Conclusion: The CHOP model reproduces some aspects of clinical pain, such as longer duration, sensitization, secondary allodynia and hyperalgesia.
Significance: Here we demonstrate a novel pain model that can be applied for up to an hour without tissue damage. The CHOP model allows for investigation of primary and secondary hyperalgesia as well as top-down influences on sensitization, thereby providing an experimental model that can be used to assess clinically-oriented questions.

1. Introduction

Human experimental pain models play an important role in understanding physiological and psychological aspects of pain as well as providing a much needed bridge between pre-clinical animal research and clinical pain management (Staahl and Drewes, 2004; Rolke et al., 2006; Hollins et al., 2011). However, many do not model important features of

clinical pain, such as longer-lasting duration or hyperalgesia (Staahl and Drewes, 2004). Experimental pain stimuli frequently used in humans include thermal, electrical, or laser stimulation, which are applied for several seconds to a few minutes (Olesen et al., 2012). In addition, several stimuli are available that can be applied for longer durations. Both the cold pressor and the tourniquet ischemia test have been applied for up to 60 min (Hines and Brown, 1936; Smith et al., 1966; Mitchell et al., 2004). However, these pain stimuli induce cardiovascular reactions (Hines and Brown, 1936; Lovallo, 1975; Handwerker and Kobal, 1993), a potential confound because increased blood pressure, even in the normal range, is associated with decreased pain sensitivity (Zamir and Shuber, 1980; Bruehl et al., 1992). In addition, many models do not produce peripheral and central sensitization, a key factor in many clinical pain conditions (Koltzenburg et al., 1994; Schmelz, 2009).

Peripheral and central sensitization in clinical populations and healthy individuals can be assessed by measuring primary and secondary hyperalgesia and allodynia (Campbell et al., 1988; Gracely et al., 1992; Coronado et al., 2014). The heat/capsaicin model (Petersen and Rowbotham, 1999) is a short lasting model that produces stable levels of hyperalgesia and allodynia over time, but not of ongoing pain (Petersen and Rowbotham, 1999). In order to address this restriction, we developed the capsaicin/heat ongoing pain (CHOP) model, in which capsaicin and continuous application of heat produce ongoing painful sensations for up to 1-h. Thus, the CHOP model retains the advantages of the heat/capsaicin model (i.e. central and peripheral sensitization) and adds an ongoing painful experience. Here we describe the model in two experiments: (1) when a constant temperature is applied and (2) when constant perception is achieved by temperature regulation. To more fully characterize the model, we included measures of cardiovascular responses; mood; and biomarkers of stress (i.e. cortisol and alpha-amylase), endogenous pain modulation (i.e. beta-endorphin), and inflammation (i.e. interleukin-6 (IL-6), interleukin-10 (IL-10)) during the constant perception experiment.

2. General methods experiments 1 and 2

2.1 Participants

Healthy volunteers between 18 and 35-years of age were recruited via advertisements on an internal McGill University webpage. Exclusion criteria were: any pain conditions, frequent alcohol (more than 10 units/week) or recreational drug use, regular or frequent night shift work and any other medical conditions including neurological and psychiatric diseases that may influence pain perception.

The study was approved by the McGill University Institutional Review Board in accordance with the Declaration of Helsinki (2013). Written informed consent was obtained from all participants.

2.2 Capsaicin/heat ongoing pain (CHOP) model

The CHOP model is a modification of the heat/capsaicin model by Petersen and Rowbotham (1999). For the model, 0.075% capsaicin cream was applied in an approximately 2-mm thick layer to a 10 × 10 cm area on the calf. After 20 min, the cream was removed and a 12.7 × 20.3 cm flexible heating pad (TC-1000 Temperature Controller, CWE Inc., Ardmore, PA, USA) was applied to the area using Velcro® straps (one strap above and one below the cream application area). The temperature controller included an external temperature probe, which was placed between the participant's skin and the heating pad to monitor the temperature.

In Experiment 1, a constant temperature was applied while participants provided ratings of the perceived intensity and the unpleasantness/pleasantness (affective ratings). In Experiment 2, constant perceived intensity was achieved by adjusting the temperature based on participants' ratings. Experiment 2 consisted of one painful and one non-painful session.

2.3 Regulation phase

Before each experimental period, the temperature was titrated based on participants' intensity ratings to reach a predetermined target intensity. The temperature of the heating pad was initially set to 36 C. Regulation was then performed for 10 min with intensity and affective ratings recorded every 2 min. Temperature was increased or decreased in steps of 0.1–2 C until participants reached the target intensity. If the target was not reached within 10 min, regulation was continued for an additional 5 min with ratings taken every 1 min. At the end of this additional period, if the perceived intensity was not within the appropriate range of the scale (i.e. painful or non-painful) the experiment was discontinued, and the participant was excluded. The final intensity and affective ratings of the regulation phase were used as the baseline ratings in the analysis of the test phase and the last temperature of the regulation phase was used as baseline temperature.

2.4 Test phase

After regulation to the target sensation, a 60-min test phase began. During the test phase, intensity and affective ratings were taken every 4 min using an automated computer program. At 4-min intervals, a tone alerted the participants to rate using a number keypad, first on the intensity scale then on the affective scale. In between ratings, the computer screen was black. At each rating period, the applied temperature was also recorded because the temperature of the heating pad could fluctuate ±0.4 C from the target temperature.

Participants completed the McGill Pain Questionnaire (MPQ) every 8 min, after every second rating period. The MPQ included the sensory, affective, evaluative, and miscellaneous words used to calculate the 'total score' (max score = 78).

2.5 Numerical rating scales

Perceived intensity and affective ratings were obtained using standardized numerical rating scales (NRS) (Villemure et al., 2003; Wood et al., 2007; Loggia et al., 2008). On the intensity scale '0' represented 'no sensation' and '200' represented 'most intense pain tolerable', with '100' representing 'pain threshold', or the moment when the sensation was first perceived as painful. On the bipolar affective scale, '0' represented 'neutral', '−100' represented 'extremely unpleasant', and '100' represented 'extremely pleasant'. Standardized instructions based on Price et al. (1983) were used to explain the difference between intensity and affective ratings of painful stimuli.

2.6 Statistics

Statistical analyses were performed using PASW 17 (SPSS, IBM, New York, NY, USA). Data were assessed for outliers (defined as ±3 standard deviations from mean); if outliers were present, those values were excluded. All measures were tested for normality using the Kolmogrov–Smirnov test, which indicated that data were normally distributed.

Data were analyzed using two-sided *t*-tests or repeated measures or mixed measures analysis of variance (ANOVA), followed by *post-hoc t*-tests where appropriate. False discovery rate (FDR) was used to correct for the number of ANOVAs performed for each experiment (Benjamini and Hochberg, 1995). The FDR-adjusted significance levels are indicated as *q*-values; when the *p*-value of a test is smaller than the *q*-value, the result is considered significant following FDR correction for multiple comparisons. Effects sizes are indicated as generalized eta squared (η^2_G) (Bakeman, 2005) or Cohen's *d* (Cohen, 1988). Power analyses were performed using G-Power (version 3.1.9) and power is expressed as $1-\beta$.

2.7 Experiment 1: Constant temperature

2.7.1 Methods overview

Experiment 1 was performed with RCP as the experimenter. Each participant completed one session. Participants first completed the Pain Catastrophizing Scale (PCS), Pain Anxiety Symptoms Scale (PASS), and Fear of Pain Questionnaire (FPQ). Baseline blood pressure and heart rate were measured using an automated blood pressure cuff (Wagner, Greenwich, CT, USA) on the dominant arm. The CHOP model was applied as described above. Once the cream was removed, temperature was regulated until participants rated 130 ± 20 on the intensity scale. The final temperature reached in the regulation phase was subsequently applied during the 60-min test phase. During the test phase, blood pressure and heart rate were measured at 15, 30 and 45 min. After the 60-min test phase, the heating pad was removed, and blood pressure and heart rate were measured again. Fig. 1A displays a diagram of the session timeline.

2.7.2 Analysis

For one participant, the temperature probe fell out resulting in an invalid reading for one time point, which was replaced with the mean temperature for that participant. 'Time' served as within-subject factor for the repeated measures ANOVAS. Separate ANOVAs were conducted for intensity ratings and affective ratings ('time' 17 levels); the McGill Pain Questionnaire total score ('time' 7 levels'); and systolic blood pressure, diastolic blood pressure, and heart rate ('time' 5 levels).

Visual inspection of the data seemed to show different temporal patterns for intensity ratings: participants increased ratings, decreased ratings, or rated relatively constant over the test phase. A hierarchical cluster analysis was therefore performed to determine if different patterns were present and if yes, how many groups could be stratified based on the time course of intensity ratings. In order to perform the cluster analysis, a linear trend line was calculated for each participant's intensity ratings over time. The slope of this linear trend was used as the metric of the degree of sensitization or habituation

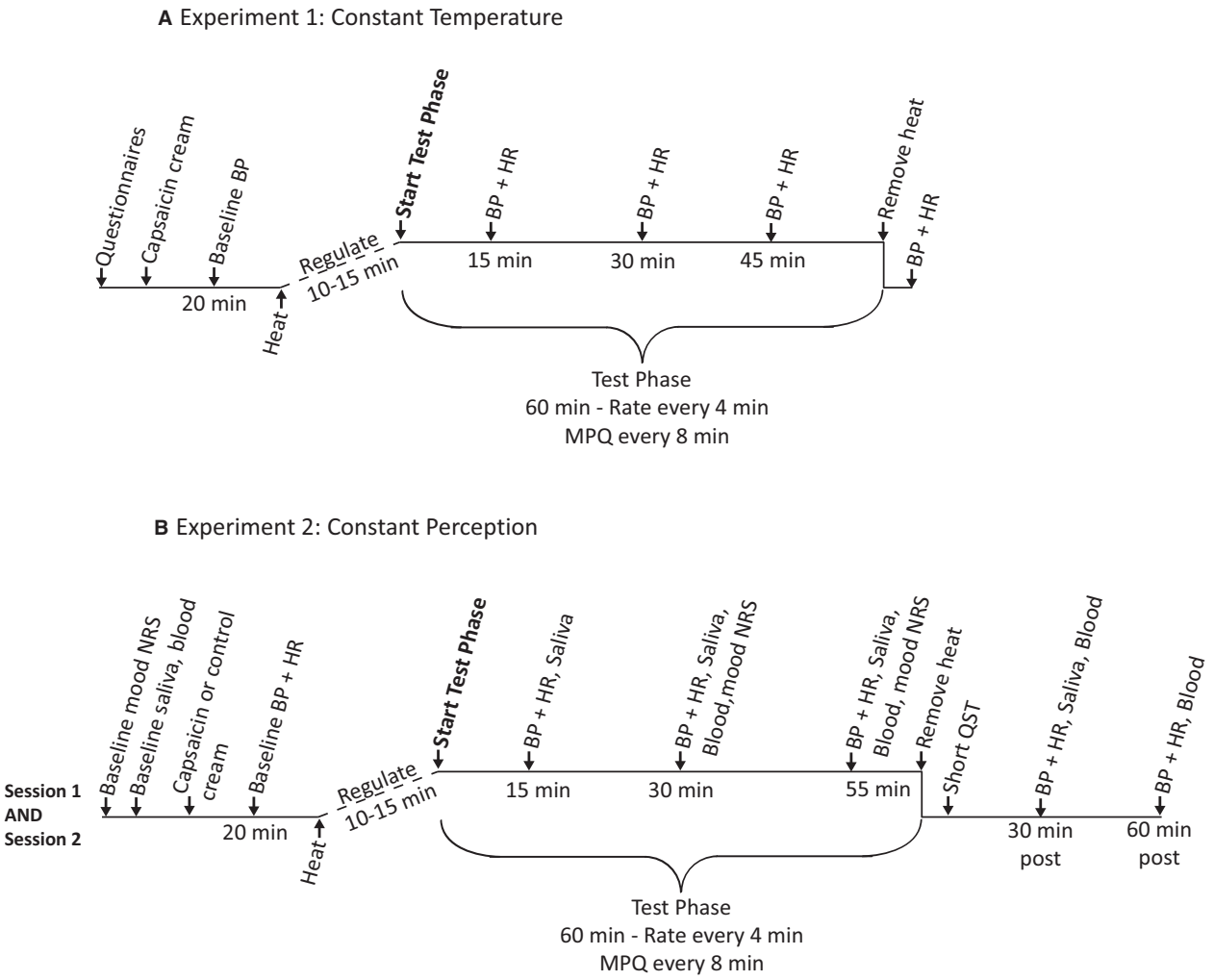

Figure 1 Timeline of experimental sessions. (A) This diagram shows the timeline of Experiment 1: 'Constant Temperature', when a constant temperature was applied for 60 min after sensitization with capsaicin cream. (B) This diagram shows the timeline of Experiment 2: 'Constant Perception', when capsaicin or control cream was applied and perceived intensity was maintained constant by adjusting the temperature. Order of capsaicin or control cream was counterbalanced across participants and the timeline was identical with both creams. In both diagrams, 'Regulate' indicates the 10–15 regulation period, while 'Test Phase' indicates the start of the 60-min experimental phase. BP, blood pressure; HR, heart rate; MPQ, McGill Pain Questionnaire; mood NRS, mood numerical rating scales; short QST, short quantitative sensory testing.

for each participant. A within groups average linkage agglomerative hierarchical cluster using squared Euclidean distance was then performed. Optimal clustering was determined based on two criteria: (1) the stage at which the agglomeration schedule coefficient increased by the greatest amount, and (2) where the largest gap in scaled distances occurred on the dendrogram (Supporting Information Fig. S1).

Following cluster analysis, data would be analyzed by the determined number of groups (if any). Age, questionnaire data, baseline temperature, baseline intensity, and baseline affective ratings would be analyzed using one-way ANOVAs with the between-subject factor 'group' (number of levels determined in cluster analysis). Intensity ratings; affective ratings; temperature; MPQ total score; systolic blood pressure; diastolic blood pressure; and heart rate would be analyzed using separate mixed measure ANOVAs with the within-subject factor 'time' and the between-subject factor 'group'. If any significant group differences were found for questionnaire data, baseline temperature, baseline intensity, or baseline affective ratings, that variable was correlated using Pearson's product moment with the degree of sensitization (slope of the linear trend of the test phase intensity ratings). Because affective ratings have been shown in

previous studies to be correlated with perceived intensity, baseline affective ratings were corrected for intensity by calculating the ratio of unpleasantness ratings to intensity ratings. For this, unpleasantness ratings were re-coded from 0 to 100 (previously 0 to −100) and painful intensity ratings from 0 to 100 (previously 100–200). The resulting ratios were correlated with the degree of sensitization to assess the relationship of relative affective ratings with the change in intensity ratings during the test phase.

2.8 Results

2.8.1 Participants

Twenty-eight healthy participants were recruited. One participant was excluded because the stimulation was not perceived as painful, two because the stimulation was perceived as intolerable, and one due to technical problems. The final sample of 24 participants of Experiment 1 included 13 females and 11 males [mean age in years (±SD): males = 26 (6.8) years, females = 21 (3.0), $p = 0.029$].

2.8.2 Test phase

The mean temperature applied across participants was 37.54 C (SD = 2.30 C). Intensity ratings showed a significant main effect of time ($F(1,23) = 10.637$, $p = 0.003$, $q = 0.025$, $\eta^2_G = 0.066$, $1-\beta = 0.87$; Fig. 2A), decreasing at early time points (12 min: $p < 0.1$; 8, 16 min: $p < 0.05$) compared to baseline ratings, while ratings at later time points increased back to baseline. Unpleasantness ratings showed a significant main effect of time similar to the intensity ratings ($F(1,23) = 16.209$, $p = 0.001$, $q = 0.017$, $\eta^2_G = 0.057$, $1-\beta = 0.95$; Fig. 2B).

In line with results on intensity and affective ratings, the MPQ total score showed a trend for a main effect of time ($F(1,22) = 5.438$, $p = 0.029$, $q = 0.033$, $\eta^2 = 0.19$, $1-\beta = 0.81$) such that scores decreased initially and increased at later time points compared to baseline. The most frequently reported words on the MPQ were burning, hot, annoying, continuous, and pricking. Several word groups were reported throughout the experiment; within these groups, words with greater intensity (words which receive a greater MPQ score) were reported more often later in the session (Supporting Information Fig. S2).

Systolic and diastolic blood pressure did not change over time (SBP: $F(1,22) = 0.1$, $p = 0.75$ $q = 0.042$, $\eta^2_G = 0.014$, $1-\beta = 0.061$; DBP: $F(1,22) = 0.98$, $p = 0.33$ $q = 0.05$, $\eta^2_G = 0.01$, $1-\beta = 0.16$). Heart rate showed a significant main effect of time ($F(1,22) = 41.760$, $p < 0.001$, $q = 0.0083$, $\eta^2_G = 0.0012$, $1-\beta = 0.99$): at all time points, heart rate was lower compared to the baseline measurement (p's < 0.01).

2.8.3 Cluster analysis

Hierarchical cluster analysis determined the ideal number of clusters in the data to be two (Supporting Information Fig. S1). The mean slope in Group 1 was 3.94 (SD = 1.33) and in Group 2 −0.05 (SD = 1.71). Therefore, Group 1 is referred to as 'sensitizers' and Group 2 as 'non-sensitizers' in the following. Sensitizers ($N = 11$) and non-sensitizers ($N = 13$) did not differ in age (mean age in years (±SD): sensitizers = 24 (6.5), non-sensitizers = 23 (4.7), $p = 0.58$) or gender distribution (sensitizers: 6 females, 5 males, non-sensitizers: 7 females, 6 males; chi-square statistic = 0.0012; p-value = 0.97).

There was a trend for a time by group interaction for temperature ($F(1,22) = 4.960$, $p = 0.036$, $q = 0.018$, $\eta^2_G = 0.00003$, $1-\beta = 0.98$). However, *post hoc* tests revealed no differences between groups for any time point (p's > 0.1). As a check of the clustering, intensity was assessed to verify separation of groups. There was a significant time by group interaction ($F(1,22) = 32.359$, $p < 0.001$, $q = 0.0045$, $\eta^2_G = 0.079$, $1-\beta = 0.99$; Fig. 2C), such that sensitizers significantly increased intensity ratings over time (p's < 0.05 from 40 to 60 min compared to baseline), while non-sensitizers did not significantly change intensity ratings over time. Sensitizers rated significantly more pain than non-sensitizers from 24 min onwards (p's < 0.05). Similarly, sensitizers increased unpleasantness ratings over time (p's < 0.05 from 48 to 60 min compared to baseline), while non-sensitizers did not change unpleasantness ratings (Time by group interaction: $F(1,22) = 14.604$, $p < 0.001$, $q = 0.009$, $\eta^2_G = 0.064$, $1-\beta = 0.99$; Fig. 2D), and sensitizers rated more unpleasantness than non-sensitizers at all time points (p's < 0.068).

In line with results on intensity and affective ratings, sensitizers increased MPQ scores over time (p's < 0.05), while non-sensitizers did not (time by group interaction: $F(1,22) = 9.385$, $p = 0.006$, $q = 0.014$, $\eta^2_G = 0.002$, $1-\beta = 1.00$). Systolic and diastolic blood pressure as well as heart rate did not differ between sensitizers and non-sensitizers nor did their time courses behave differently between the two groups (SBP: $F(1,21) = 0.471$, $p = 0.5$ $q = 0.027$, $\eta^2_G = 0.002$, $1-\beta = 0.1$; DBP: $F(1,21) = 0.466$, $p = 0.502$ $q = 0.05$, $\eta^2_G = 0.002$, $1-\beta = 0.1$).

To understand why one group of participants sensitized and one group did not, we examined potential

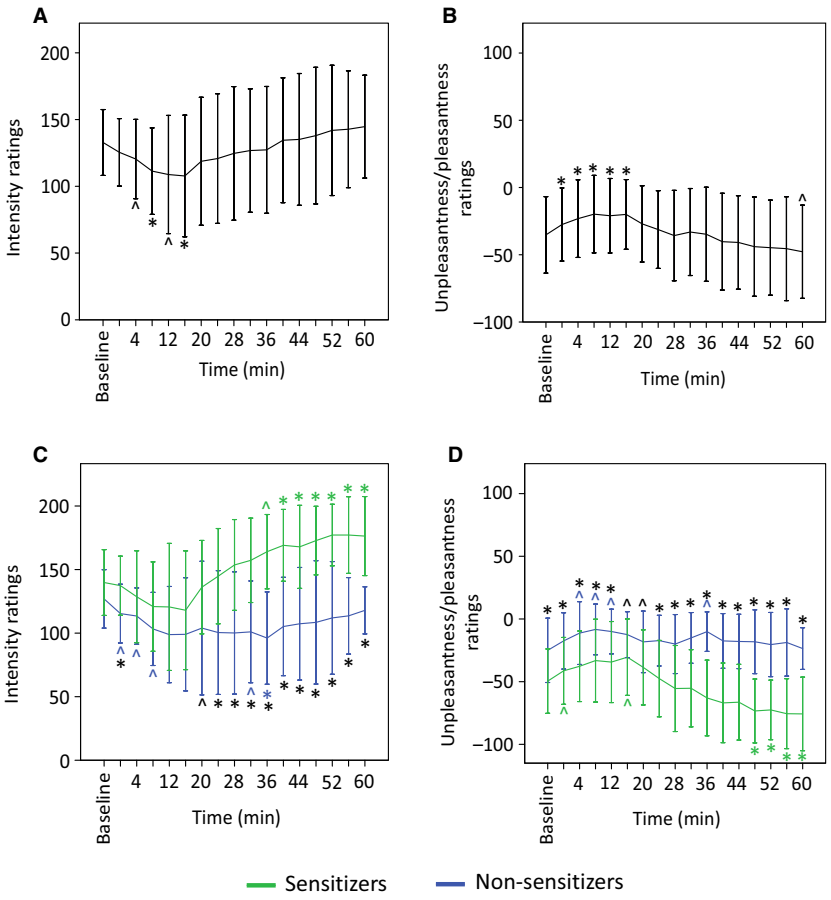

Figure 2 Results of Experiment 1: Constant Temperature. Whole sample results are shown for (A) intensity ratings and (B) affective ratings. Intensity and affective ratings initially decreased and then increased back to baseline levels (line). Results by group (sensitizers (green), non-sensitizers (blue)) are shown for (C) intensity ratings and (D) affective ratings. Sensitizers had significantly higher intensity and unpleasantness ratings than non-sensitizers. *$p < 0.05$, ^$p < 0.1$: in black, differences between groups; in green, differences from baseline for sensitizers (C and D); and in blue, differences from baseline for non-sensitizers (C and D).

group differences in pain catastrophizing (PCS), pain anxiety (PASS), and baseline intensity and affective ratings. Sensitizers tended to have higher PCS ($p = 0.069$, $d = 0.79$) and PASS ($p = 0.091$, $d = 0.72$) scores, and rated significantly more baseline unpleasantness ($p = 0.019$, $d = 1.03$). However, neither PCS and PASS scores nor baseline unpleasantness ratings correlated significantly with the degree of sensitization (p's > 0.2). However, the ratio of baseline affective ratings to baseline intensity ratings showed a strong trend to be correlated with the degree of sensitization (Fig. 3: $r = 0.40$, $p = 0.066$).

2.9 Experiment 2: Constant perception

2.9.1 Methods overview

Experiment 2 was performed with one female (CN) and one male (RT) experimenter, always subsequent to Experiment 1. Each experimenter tested equal numbers of male and female participants to avoid confounds of experimenter gender. Female and male participants were randomly assigned to either experimenter. There were no statistical differences between data collected from the different experimenters. Each participant completed two sessions 4–7 days apart; one with capsaicin cream and one with control cream (commercially available lotion). Order of conditions (capsaicin or control) was counterbalanced across participants. This experiment also included saliva and blood samples to measure biomarkers of stress (cortisol and alpha-amylase), inflammation (Il-6 and Il-10), and endogenous pain modulation (beta-endorphin).

The two sessions followed an identical outline. After arrival, participants completed several mood NRS described below. Fifteen minutes after arrival, a baseline saliva sample was taken. An intravenous

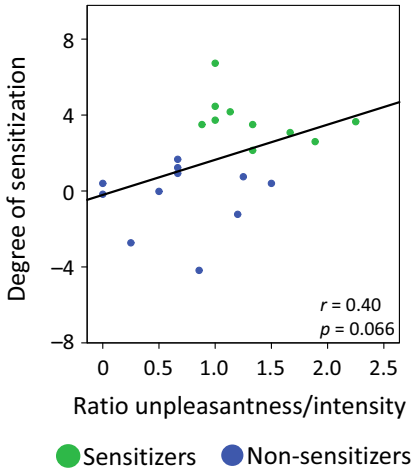

Figure 3 Degree of sensitization correlates with relative unpleasantness ratings. Initial unpleasantness (coded from 0 to 100) was divided by initial intensity ratings (coded from 0 to 100). The ratio of baseline unpleasantness to intensity ratings tended to correlate with degree of sensitization (slope of intensity ratings) during the constant temperature experiment ($r = 0.42$, $p = 0.06$), such that more relative unpleasantness (ratios > 1) related to more sensitization, while less relative unpleasantness (ratios < 1) related to less sensitization or even more habituation.

catheter was inserted in the non-dominant arm and a baseline blood sample was taken. Then, capsaicin cream or control cream was applied as per the CHOP model. Baseline blood pressure and heart rate measurements were taken 10 min after the cream was applied. Once the cream was removed, temperature was regulated as described above until participants rated 160 ± 20 on the intensity scale for the painful condition (capsaicin cream) and 30 ± 20 on the intensity scale for the non-painful condition (control cream).

The final perceived intensity reached in the regulation phase was used as the target intensity for the 60-min test phase. During the test phase, blood pressure and heart rate were measured and saliva samples taken at 15, 30 and 55 min. Blood samples were taken at 30 and 55 min. If saliva and blood samples were taken at the same time, saliva samples were taken first. Mood NRSs were completed at 30 and 55 min.

After the test phase, the heating pad was removed. Once any sensation from the CHOP had disappeared (time to zero sensation mean (±SD): non-painful condition mean = 2.19 min (±12.27 min); painful condition mean = 3.46 min (±12.32 min)), secondary allodynia and hyperalgesia were assessed using a short quantitative sensory testing protocol described below. A final saliva sample was taken 30 min after heating pad removal. Blood samples were taken 30 and 60 min after heating pad removal. Blood pressure and heart rate were measured again at 30 and 60 min after heating pad removal. Fig. 1B displays a diagram of the session timeline.

2.9.2 Mood numerical rating scales

Mood NRS were adapted from Villemure et al., 2003. Two bimodal numerical rating scales were used to assess 'Mood' and 'Anxiety'. For 'Mood', the scale ranged from 'extremely unpleasant' (−100), to 'neutral' (0), to 'extremely pleasant' (100). For 'Anxiety', the scale ranged from 'extremely anxious' (−100), to 'neutral' (0), to 'extremely calm' (100). Five numerical rating scales ranging from 'not at all' (0) to 'extremely' (100) were used for 'Anger', 'Fear', 'Sadness', 'Disgust', and 'Happiness'.

2.9.3 Saliva samples

Samples were collected using the passive drool method. Participants collected saliva in their mouths and drooled into a siliconized tube through a straw until 1.5 mL of saliva were collected. Samples were put on ice until centrifuged, aliquoted into 200–400 μL samples and stored at −80 °C until analyzed. Samples were analyzed in duplicate per manufacturer's specifications using an enzyme-linked immunosorbent assay (ELISA) (Salimetrics, State College, PA, USA) for cortisol and a kinetic reaction immunoassay kit (Salimetrics) for alpha-amylase. The means of the duplicate analyses were used as the respective outcome measures.

2.9.4 Blood samples

Venous blood sampling was performed by registered nurses CN and RT. Samples of 10 mL were collected at each time point in two 6-mL anticoagulant (EDTA) vacutainer tubes. After collection, blood samples were centrifuged within 30 min at 1300 *g* for 15 min at room temperature to separate plasma. Plasma was then aliquoted into 500 μL – 1 mL samples and stored at −80 C until analyzed. Samples were analyzed in duplicate using ELISAs per manufacturer's specifications for IL-6 (R&D Systems, Minneapolis, MN, USA) and for IL-10 (Life Technologies, Burlington, ON, Canada). Three samples from the painful and non-painful sessions (baseline, 30, and 55 min during test phase) were also analyzed in duplicate for beta-endorphin using a Milliplex human neuropeptide multiplex magnetic bead assay

per manufacturer's specifications (EMD Millipore, Etobicoke, ON, Canada). The means of the duplicates were used as the respective outcome measures.

2.9.5 Short quantitative sensory testing

The short quantitative sensory testing was adapted from the German Quantitative Sensory Testing Protocol (Rolke et al., 2006) to test for the presence of mechanical allodynia and mechanical hyperalgesia. Mechanical allodynia was assessed by applying strokes between 200 and 400 mN using a standardized brush (Senselab Brush-05, Somedic, Sweden). Mechanical hyperalgesia was assessed using a 256 mN von Frey filament (OptiHair, Marstock Nervtest, Germany). Mechanical hyperalgesia testing was always performed after allodynia testing to reduce sensitizing effects.

For each test, one test stimulus was first applied to the homologous site on the untreated leg, followed by 3 test stimuli in the center of the capsaicin or control cream area on the treated leg. Participants were asked to compare the stimuli and to remember how the stimuli on the treated leg felt. Next, stimuli were applied at 0.5-cm intervals starting 6 cm outside of the capsaicin or control cream application area, moving toward the affected area in each direction (top, bottom, left and right) until the edges of the area were reached. When participants indicated an increase in sensation, it was marked as the border of the allodynic or hyperalgesic area. If the participant did not indicate an increase in sensation, the border was marked at the edge of the cream area. If a decrease in sensation was indicated, the data of that participant was excluded for the assessment of allodynia or hyperalgesia (non-painful condition: $n = 7$ excluded for allodynia, $n = 4$ excluded for hyperalgesia, painful condition: $n = 2$ for allodynia, $n = 0$ for hyperalgesia). The distance from the edge of the area was measured and the sum of all four directions was used as the outcome measure.

2.10 Statistical analysis

One outlier was excluded at each time point for cortisol levels. No outliers were present in any of the other measures. Using separate repeated measure ANOVAs, intensity ratings, affective ratings, and temperature from the test phase were analyzed with within-subject factors 'condition' (2 levels; painful and non-painful) and 'time' (17 levels). Repeated measure ANOVAs with within-subject factors 'condition' (2 levels; painful and non-painful) and 'time' (7 levels) were performed for mood NRSs ('time' 3 levels); systolic blood pressure, diastolic blood pressure and heart rate ('time' 6 levels); cortisol, alpha-amylase, IL-6, and IL-10 ('time' 5 levels); and beta-endorphin ('time' 3 levels, baseline, 30 and 55 min during stimulation) levels. Hyperalgesia and allodynia data were compared between conditions using paired-sample *t*-tests. To assess any effects of session order, separate repeated measure ANOVAs were performed for all outcome measures with the within-subject factors 'session' (2 levels, 1 or 2) and 'time'.

To assess the relationships between biomarkers and pain sensitivity, exploratory analyses were performed using two-tailed Pearson's product correlations between biomarkers and pain measures (temperature and intensity). Because beta-endorphin and cortisol are both part of the hypothalamic pituitary-adrenal stress response (Akil et al., 1984; Drolet et al., 2001), correlations were also calculated between beta-endorphin and cortisol levels.

Data from Experiment 2 were also analyzed using the group assignment (sensitizers, non-sensitizers) determined in Experiment 1. Because two participants from Experiment 1 did not participate in Experiment 2, potential differences between sensitizers and non-sensitizers were reassessed for age, PCS, PASS and FPQ. Mixed effects ANOVAS were performed with the within subject factors of 'condition' and 'time' and the between subject factor of 'group' (2 levels) to analyze intensity ratings; affective ratings; temperature; MPQ total score; blood pressure; heart rate; mood NRSs; and biomarkers. Differences between conditions and group for allodynia and hyperalgesia were assessed using repeated measure ANOVAs.

3. Results

3.1 Participants

Of the 24 participants who completed Experiment 1, 18 contributed complete data to Experiment 2. Two participants dropped out for unknown reasons, and 4 participants were excluded because the capsaicin condition was rated as non-painful ($n = 1$) or the non-painful condition was rated as painful ($n = 3$). In addition, 2 participants who did not complete Experiment 1 because the sensation became intolerable or because of technical problems, completed Experiment 2. Thus, the final sample for Experiment 2 consisted of 20 participants (10 males, 10 females) (mean age in years (±SD): males = 25 (6.3), females = 22 (3.0), $p = 0.162$).

3.2 Test phase

As a manipulation check, intensity ratings were first assessed to ensure that the perceived intensity was constant during the 60-min test phase and that ratings were higher with capsaicin compared to the control cream. As intended, intensity did not change over time (effect of time: $F(1,19) = 0.033$, $p = 0.86$, $q = 0.05$, $\eta^2_G = 0.011$, $1-\beta = 0.98$) but did differ between conditions (effect of condition: $F(1,19) = 379.409$, $p < 0.001$, $q = 0.002$, $\eta^2_G = 0.890$, $1-\beta = 1.00$, Fig. 4A) with intensity ratings being significantly higher in the painful condition compared to the non-painful condition (p's < 0.001). Further, the painful condition was rated as significantly more unpleasant compared to the non-painful condition (main effect of condition: $F(1,19) = 79.001$, $p < 0.001$, $q = 0.004$, $\eta^2_G = 0.586$, $1-\beta = 1.00$, Fig. 4B), and the MPQ total score was higher in the painful compared to non-painful condition (main effect of condition: $F(1,19) = 46.373$, $p < 0.001$, $q = 0.008$, $\eta^2_G = 0.411$, $1-\beta = 1.00$). Temperatures needed to be upregulated in both conditions to produce constant perceptions (main effect of time: $F(1,19) = 8.144$, $p = 0.010$, $q = 0.015$, $\eta^2_G = 0.019$, $1-\beta = 0.35$, Fig. 4C). The most frequently reported words on the MPQ in the painful condition were burning, continuous, radiating, annoying, stinging, sharp, pricking and hot. In the non-painful condition, the most frequently reported words were hot, continuous, tingling and dull. Within word groupings, words were reported consistently during stimulation in the painful condition (Supporting Information Fig. S2), further indicating that a constant perception was maintained. Only low intensity words (those with an MPQ score of 1 or 2) were reported in the non-painful condition (Supporting Information Fig. S2).

'Overall mood' and 'happiness' ratings tended to be lower in the painful compared to non-painful condition (main effect of condition: 'overall mood' $F(1,19) = 3.591$, $p = 0.073$, $q = 0.025$, $\eta^2_G = 0.036$, $1-\beta = 0.44$; 'happiness' ($F(1,16) = 4.472$, $p = 0.050$, $q = 0.021$, $\eta^2_G = 0.02$, $1-\beta = 0.511$)). Greater areas of mechanical allodynia and hyperalgesia were reported in the short QST in the painful compared to the non-painful condition (allodynia: $t = -2.676$, df = 12, $p = 0.020$, $d = 0.93$; painful = 3.9-cm (±2.9-cm), non-painful = 1.6-cm (±2.3 cm); hyperalgesia: $t = -3.418$, df = 15, $p = 0.004$, $d = 0.97$; painful = 10.1-cm (±5.1-cm), non-painful = 4.6-cm (±4.9-cm)).

There was no difference in systolic blood pressure, diastolic blood pressure, or heart rate between painful and non-painful conditions (SBP: $F(1,18) = 2.375$, $p = 0.141$ $q = 0.019$, $\eta^2_G = 0.006$, $1-\beta = 0.309$; DBP: $F(1,18) = 0.33$, $p = 0.572$ $q = 0.05$, $\eta^2_G = 0.001$, $1-\beta = 0.085$; HR: $F(1,18) = 0.095$, $p = 0.762$ $q = 0.042$, $\eta^2_G = 0.001$, $1-\beta = 0.06$). Systolic blood pressure showed a trend for a main effect of time ($F(1,18) = 6.517$, $p = 0.02$, $q = 0.017$, $\eta^2_G = 0.013$, $1-\beta = 0.675$), however systolic blood pressure only increased significantly compared to baseline 60 min after the heating pad was removed ($p < 0.01$), regardless of condition. Heart rate significantly decreased over time, again regardless of condition (main effect of time: $F(1,18) = 32.061$, $p < 0.001$, $q = 0.006$, $\eta^2_G = 0.052$, $1-\beta = 1.00$). There were no main effects or interactions for diastolic blood pressure.

Alpha-amylase, IL-10, and beta-endorphin levels did not differ between painful or non-painful conditions, between session 1 or session 2, nor over the course of the session. IL-6 and cortisol levels neither differed between painful and non-painful conditions. However, IL-6 steadily increased over time (Fig. 5A), while cortisol levels increased and decreased (Fig. 5B) (main effect of time: IL-6 $F(1,19) = 23.971$, $p < 0.001$, $q = 0.012$, $\eta^2_G = 0.141$, $1-\beta = 0.996$; cortisol $(1,19) = 19.342$, $p < 0.001$, $q = 0.0096$, $\eta^2_G = 0.040$, $1-\beta = 0.99$). IL-6 showed a greater increase in session 2 compared to session 1 (Fig. 5C) (time by session interaction: $F(1,19) = 10.475$, $p = 0.004$, $q = 0.013$, $\eta^2_G = 0.040$, $1-\beta = 0.893$). Cortisol levels tended to be higher in session 1 compared to session 2 (Fig. 5D) (main effect of session: $F(1,19) = 4.26$, $p = 0.053$, $q = 0.023$, $\eta^2_G = 0.06$, $1-\beta = 0.991$).

Unlike cortisol and IL-6, there were no main effects or interactions of session order for any other measure, specifically intensity ratings; affective ratings; temperature; MPQ total score; the 7 mood NRSs; systolic blood pressure, diastolic blood pressure and heart rate; alpha-amylase; IL-10; or beta-endorphin.

Despite not being different between painful and non-painful conditions, beta-endorphin levels at baseline did relate to applied temperature during pain, such that higher baseline beta-endorphin (measured before capsaicin application) correlated with lower applied temperatures for 0 min to 16 min (Fig. 6A) (r's = −0.325 to −0.385, p's < 0.01). Further, lower applied temperature at these time points correlated with higher intensity ratings (Fig. 6B) (r's = −0.300 to −0.460, p's < 0.01), suggesting that higher baseline beta-endorphin is related to higher pain sensitivity (lower

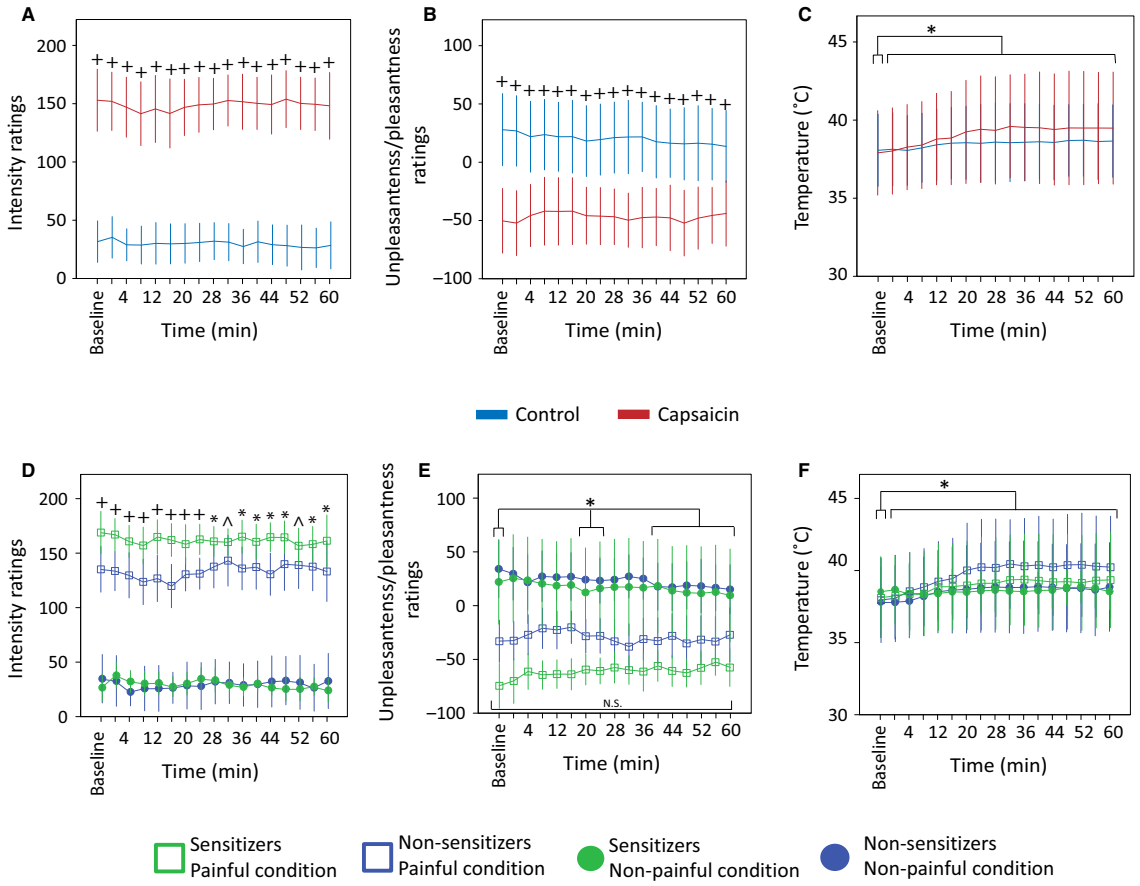

Figure 4 Results of Experiment 2: Constant Perception. Whole sample results are shown for (A) intensity ratings, (B) affective ratings, and (C) temperature. Intensity (A) and unpleasantness (B) ratings were significantly higher in the painful (red) compared to non-painful (light blue) condition. There was a significant main effect of time for temperature (C), but no interaction with painful or non-painful conditions. Results by group (sensitizers, non-sensitizers) are shown for (D) intensity ratings, (E) affective ratings, and (F) temperature. Sensitizers (green) rated significantly more intensity (D) during the painful condition (squares) compared to the non-sensitizers (blue); however, during the non-painful condition (circles), sensitizers (green) and non-sensitizers (blue) did not differ in intensity ratings. Sensitizers tended to rate more unpleasantness than non-sensitizers regardless of condition ($p = 0.052$). Although sensitizers did rate more unpleasantness in the painful condition (squares) than non-sensitizers (E), this difference was not significant ($p = 0.114$). Non-painful pleasantness (circles) ratings decreased over time, while painful unpleasantness (squares) ratings did not change over time. Temperatures (F) increased over time (as in C), but did not differ between sensitizers and non-sensitizers or between painful and non-painful conditions. $^{+}p < 0.001$, $*p < 0.05$, $^{\wedge}p < 0.1$: in black, differences between conditions (A, B, or C) or groups (D, E, or F); in red, differences from baseline for painful condition (A, B, or C); in light blue, differences from baseline for non-painful condition (A, B, or C); in green, differences from baseline for sensitizers (D, E, or F); and in blue, differences from baseline for non-sensitizers (D, E, or F).

applied temperatures in combination with higher intensity ratings). Baseline beta-endorphin did not correlate with temperature or intensity ratings during the non-painful session (r's < 0.2, p's > 0.4). Baseline beta-endorphin correlated negatively with the change in beta-endorphin from baseline to during pain, such that individuals with lower baseline beta-endorphin showed a greater increase in beta-endorphin during pain (30 min $r = -0.621$, $p = 0.004$; 55 min $r = -0.708$, $p < 0.001$; Fig. 6C). Baseline cortisol levels also correlated with pain sensitivity; higher baseline cortisol correlated with lower intensity ratings during pain at baseline throughout 8 min (Fig. 6D) (r's = −0.303 to −0.424, p's < 0.01). Cortisol and beta-endorphin levels did not correlate at any time point (r's < 0.2, p's > 0.4). However, baseline beta-endorphin levels correlated with the change in cortisol levels at 15 min and 30 min, such that participants with higher baseline beta-endorphin levels showed a greater increase in cortisol during pain (Fig. 6E,F) (15 min $r = 0.504$, $p = 0.028$; 30 min $r = 0.476$, $p = 0.039$). No relationships between pain measures and alpha-amylase, IL-6, or IL-10 were found.

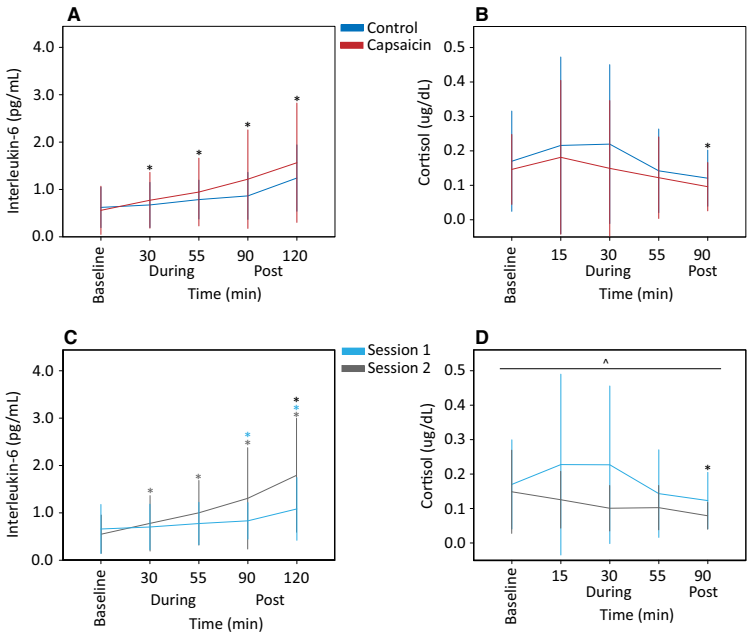

Figure 5 Session effects of cortisol and interleukin-6. Results of ANOVA by condition for interleukin-6 (IL-6) (A) and cortisol (B) show a main effect of time, such that IL-6 increases over time, while cortisol increases and then significantly decreases 30 min after heating pad was removed. There was no difference between control (blue) or capsaicin (red) conditions for either IL-6 or cortisol. Results for the ANOVA by session order (session 1 in light blue, session 2 in gray) are shown in (C) for IL-6 and (D) for cortisol. IL-6 showed a significant session by time interaction; IL-6 increased in both sessions, but the increase was significant at all time points only in session 2. Additionally, session 1 and session 2 significantly differed 60 min (120 min) after heating pad was removed. For cortisol, there was a significant main effect of time (as described for (B)) and a significant main effect of session, such that session 1 cortisol levels were significantly higher than session 2 levels. *$p < 0.05$, ^$p < 0.1$: in black, differences between conditions (A, B) or sessions (C, D); in light blue, differences from baseline for Session 1 (C, D); in gray, differences from baseline for Session 2 (C, D).

3.3 Do sensitizers and non-sensitizers differ in the constant perception experiment?

For participants who completed Experiment 1 and Experiment 2 ($n = 18$), data from Experiment 2 were also analyzed by groups (sensitizers and non-sensitizers) as determined by cluster analysis in Experiment 1. Sensitizers ($n = 8$) and non-sensitizers ($n = 10$) did not differ significantly in age (mean age in years (±SD): sensitizers = 22 (2.8), non-sensitizers = 26 (6.9), $p = 0.1$) or gender distribution (sensitizers: 4 females, 4 males, non-sensitizers: 6 females, 4 males; chi-square statistic = 0.18, p-value = 0.67).

Sensitizers defined in Experiment 1 were found to be more pain sensitive in Experiment 2. Sensitizers tended to have higher intensity ratings in the painful condition while intensity ratings in the non-painful condition did not differ between sensitizers and non-sensitizers (group by condition interaction: $F(1,16) = 6.567$, $p = 0.021$, $q = 0.0025$, $\eta^2_G = 0.15$, $1-\beta = 0.672$) (Fig. 4D). In both conditions, non-sensitizers tended to habituate in the beginning, but sensitizers did not change ratings throughout the test phase (time by group interaction: $F(1,16) = 5.331$, $p = 0.035$, $q = 0.008$, $\eta^2_G = 0.21$, $1-\beta = 0.583$). In contrast to intensity ratings, no significant differences between sensitizers and non-sensitizers were found for temperatures, affective ratings, or MPQ measures (group by condition interaction: temperature: $F(1,16) = 0.159$, $p = 0.695$ $q = 0.008$, $\eta^2_G = 0.005$, $1-\beta = 0.066$; affective ratings: $F(1,16) = 2.788$, $p = 0.114$ $q = 0.01$, $\eta^2_G = 0.06$, $1-\beta = 0.348$; MPQ: $F(1,16) = 1.944$, $p = 0.182$ $q = 0.013$, $\eta^2_G = 0.034$, $1-\beta = 0.259$; Fig. 4E,F).

'Overall Mood' ($F(1,16) = 5.736$, $p = 0.029$, $q = 0.005$, $\eta^2_G = 0.19$, $1-\beta = 0.614$) as well as 'Happiness' ratings ($F(1,16) = 4.211$, $p = 0.061$, $q = 0.0125$, $\eta^2_G = 0.22$, $1-\beta = 0.476$) tended to be lower in sensitizers than in non-sensitizers. 'Overall Mood' and 'Happiness' did not differ between conditions, possibly because of the decreased power compared to the total sample, and did not show an interaction between group and condition. No differences between sensitizers and non-sensitizers were found

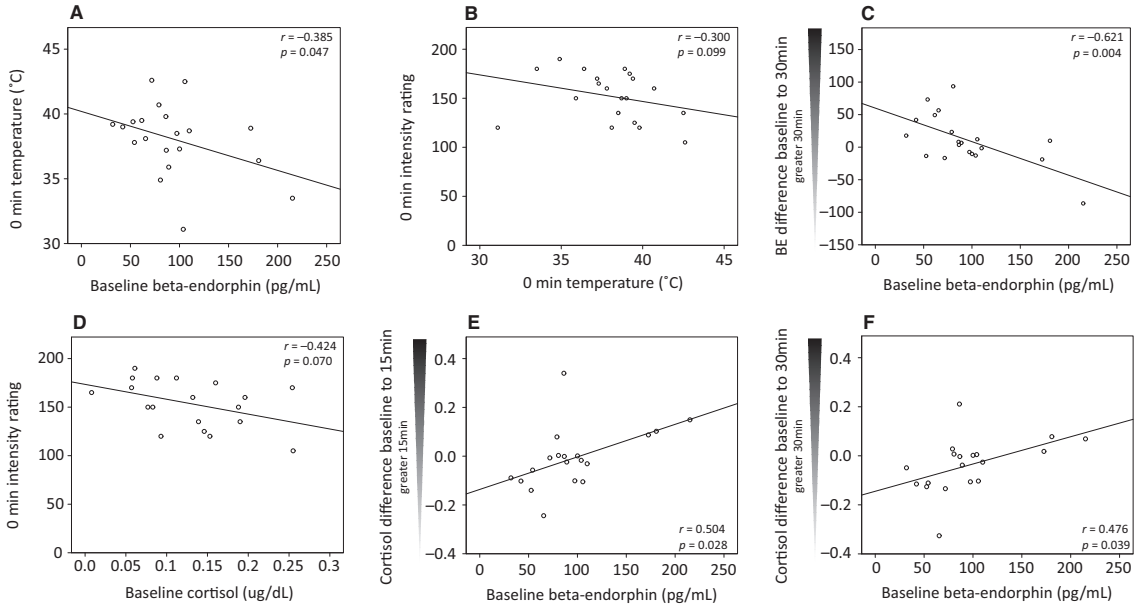

Figure 6 Beta-endorphin, cortisol, and pain interactions. The correlations between beta-endorphin, cortisol, and pain. Baseline beta-endorphin correlated with applied temperature from baseline to 16 min; (A) shows an example time point at 0 min where higher baseline beta-endorphin correlates with lower temperatures. Higher applied temperatures correlated with decreased intensity ratings as shown in (B), suggesting that higher baseline beta-endorphin correlated with increased pain sensitivity. (NOTE (B) shows the correlation of temperature and intensity at 0 min, but this relationship was consistent throughout the experiment.) Higher baseline opioid levels also correlated with a smaller change or even decrease in beta-endorphin levels during pain (C). Higher baseline cortisol levels correlated with lower intensity ratings (D), indicative of stress-induced analgesia. While beta-endorphin and cortisol levels were not correlated at any time points, higher baseline beta-endorphin correlated with a greater change in cortisol levels during pain at 15 min (E) and 30 min (F), suggesting activation of the hypothalamic-pituitary-adrenal stress response.

for the areas of allodynia or hyperalgesia, systolic blood pressure, diastolic blood pressure, heart rate, or any biomarker (alpha-amylase, cortisol, IL-10, IL-6, or beta-endorphin).

4. Discussion

Here we report the characterization of a novel pain model in humans that produces ongoing pain for at least 60 min. When a constant temperature was applied using the CHOP model, participants initially decreased and then increased their pain ratings back to baseline levels. Half of the participants showed significant sensitization, while the other half did not sensitize or even habituated. It was also possible with the CHOP model to produce constant intensity and affective ratings of painful and non-painful sensations. Potentially confounding cardiovascular, stress, and inflammatory responses did not differ between painful and non-painful conditions, indicating that the ongoing pain produced by the CHOP model was not affected by these measures. Lastly, the model produced secondary hyperalgesia and allodynia, as expected for capsaicin.

4.1 Constant temperature in the CHOP model causes habituation and sensitization

Across all participants, the application of a constant temperature caused an initial decrease followed by an increase in intensity and unpleasantness ratings. A similar pattern has been found before when applying short repetitive or constant stimuli (Granot et al., 2003; Naert et al., 2008; Hollins et al., 2011). Thus, the temporal pattern found across participants is consistent with previous studies. In the present study, the time span during which the temporal pattern occurs is longer, perhaps due to the lower intensity stimuli used.

With a constant, painful temperature, half the participants increased their pain ratings (sensitizers), while the other half did not change their ratings over time (non-sensitizers). Previous studies have also found sensitizers and non-sensitizers/habituaters to constant or repetitive noxious heat stimulation in similar proportions (Severin et al., 1985; Granot et al., 2003; Naert et al., 2008; Hollins et al., 2011; Stankewitz et al., 2013). Sensitizers and habituaters were observed with long duration stimuli (Granot et al., 2003; Hollins et al., 2011; Stankewitz et al.,

2013), but short duration stimuli only produced habituaters (Defrin et al., 2008; Teutsch et al., 2008). In addition, habituation decreases when the area of stimulation is increased (Defrin et al., 2008). Taken together, sensitization is more likely when noxious stimulation is applied for longer durations and over a larger area. Despite this clear relationship with the amount of noxious input, top-down mechanisms seem to modulate the development of sensitization. In the present study, sensitizers and non-sensitizers differed in their affective experience of pain. Previous reports showed that higher baseline pain sensitivity lead to more sensitization and less habituation (Defrin et al., 2008; Naert et al., 2008; Hollins et al., 2011). However, these studies only assessed pain intensity (and not pain affect) and thus, it is not clear whether sensory pain ratings indeed related to increased sensitization or whether it was an integration of sensory and affective perceptions. In the current study, baseline pain unpleasantness was significantly higher in sensitizers and, when expressed relative to intensity ratings, correlated with the degree of sensitization. Thus, our data suggest that it is actually the affective dimension, rather than the sensory dimension, that is more important in determining the degree of sensitization. The importance of the affective domain for the development of sensitization is further emphasized by the finding that pain catastrophizing and pain anxiety were higher in sensitizers compared to non-sensitizers. This suggests that these characteristics also contribute to group differentiation, perhaps via descending pain facilitation. It should be pointed out that pain catastrophizing and pain anxiety were always assessed at the beginning of Experiment 1, and therefore, these 'trait' measures might have been influenced by participants' state. Nevertheless, a higher degree of pain sensitivity of the 'sensitizers' as determined in Experiment 1 persisted in Experiment 2, indicating some stability of the effect.

4.2 The CHOP model can produce constant painful and non-painful sensory and affective ratings

By adjusting the applied temperature, constant perceived painful and non-painful sensations were produced for up to 60 min. Other human pain models can produce ongoing pain, but an advantage of the CHOP model is the ability to regulate the stimulus intensity using a non-invasive method. For instance, ongoing pain produced by high concentration capsaicin fluctuates over time (Koltzenburg et al., 1994; Anderson et al., 2002; Schmelz, 2009; Segerdahl et al., 2015; Lin et al., 2017) and intramuscular hypertonic saline, which can produce constant ongoing pain, is invasive (Zhang et al., 1993). The CHOP model exploits both capsaicin-induced sensitization and spatial summation to produce long duration pain without tissue damage. This model is possibly suited to test analgesics and to understand more complex psychological and physiological processes that accompany pain. For example, we have used this model to investigate the influence of ongoing pain on reward processing (Gandhi et al., 2013).

The ongoing pain produced by the CHOP model was not influenced by cardiovascular, stress, or systemic inflammatory factors. There was no change in blood pressure during pain; other models, such as the cold pressor and tourniquet ischemia tasks, are associated with blood pressure increases (Lovallo, 1975; Handwerker and Kobal, 1993), which can significantly affect pain sensitivity (Bruehl et al., 2010; Chalaye et al., 2013). Further, cortisol and IL-6 increased over time in both the painful and non-painful conditions, indicating that stress and inflammation do not specifically affect pain with this model. Previous studies of phasic painful stimuli have shown increased systemic cortisol and IL-6 in healthy participants (Lutgendorf et al., 2000, 2004; Edwards et al., 2008), but did not include a non-painful control condition. Therefore, it is unknown whether cortisol and IL-6 increase in response to phasic non-painful stimuli as well. In the present study, cortisol levels tended to be higher in session 1 compared to session 2 of Experiment 2, regardless of whether the session was painful or not. Uncertainty about session order may have caused higher stress in session 1 because participants were only aware that one session would be painful and one non-painful but they were unaware of the session order. Importantly, all participants had experienced the CHOP model previously, indicating that it is uncertainty and not anxiety driving this effect. IL-6 levels increased in both sessions, but the increase was greater in session 2 compared to session 1. HPA stress responses and inflammation are thought to have inhibitory effects on each other (Silverman and Sternberg, 2012), which might explain why the IL-6 increase was greater in session 2 when cortisol levels were lower.

Beta-endorphin levels did not differ between painful and non-painful sessions. However, baseline beta-endorphin correlated with pain sensitivity in the painful condition and did not correlate with non-painful sensations. Higher baseline beta-endorphin

correlated with lower applied temperature, which correlated with higher pain intensity. Thus, higher baseline beta-endorphin was indirectly related to increased pain sensitivity. A positive correlation between beta-endorphin and pain sensitivity is consistent with previous studies that also assessed longer duration pain (Leonard et al., 1993; Matejec et al., 2003; Bruehl et al., 2012). Baseline beta-endorphin levels may be indicative of basal opioid tone; low baseline beta-endorphin correlated with a greater change in beta-endorphin during pain. In other words, individuals with low basal opioid tone mounted a greater opioid release in response to pain than individuals with a high basal tone, perhaps explaining the decreased pain sensitivity in these participants.

Release of beta-endorphin is related to activation of the HPA axis (Papadimitriou and Priftis, 2009). Interestingly, higher baseline beta-endorphin correlated with a greater increase in cortisol during pain. Cortisol and beta-endorphin had opposing effects: higher baseline cortisol was related to lower pain-sensitivity, suggestive of stress-induced analgesia (Butler and Finn, 2009). Thus, divergent effects were noted for beta-endorphin and cortisol, both for the pain-induced release as well as effects on pain sensitivity.

4.3 Potential clinical relevance of the CHOP model

The CHOP model produced sensitization and secondary allodynia and hyperalgesia, important factors in clinical pain. Central sensitization is a common symptom in chronic pain conditions and is thought to be mediated by the same peripheral and central factors as experimentally-induced hyperalgesia and allodynia (LaMotte et al., 1991; Simone and Ochoa, 1991; Koltzenburg et al., 1994). Chronic pain patients typically show more sensitization to experimental pain stimuli than healthy controls (Koltzenburg et al., 1994; Kleinböhl et al., 1999; Smith et al., 2008). In our study, across participants, sensitization occurred during the constant temperature experiment, but some participants sensitized to a greater extent than others, which could be an indication of different phenotypes of individuals (Granot et al., 2003; Naert et al., 2008; Hollins et al., 2011; Stankewitz et al., 2013), perhaps with different propensities to develop clinical pain. Here, we showed that sensitization was related to affective pain sensitivity and potentially to pain catastrophizing and pain anxiety. Future studies could investigate the differences between sensitizers and non-sensitizers in more detail to identify other potential factors that influence the different sensitization patterns. Recent discussions in the pain community have demonstrated a need for human pain models that more accurately portray clinical pain (Buonocore et al., 2015; Lötsch et al., 2015a,b). The CHOP model allows for investigation of secondary hyperalgesia as well as top-down influences on sensitization, thereby providing an experimental model that can be used to assess clinically-oriented questions.

Author contributions

RC Price designed the experiment, collected and analyzed data, and drafted the manuscript. W Gandhi assisted with the experimental design. C Nadeau collected data. R Tarnavskiy collected data. A Qu assisted with data entry and analysis. E Fahey assisted with experimental design and pilot testing. L Stone assisted with experimental design and provided expertise on biomarker analyses. P Schweinhardt designed experiment and drafted the manuscript. All authors discussed the results and commented on the manuscript.

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Supporting Information

Additional Supporting Information may be found online in the supporting information tab for this article:

Figure S1. Cluster analysis dendrogram.

Figure S2. McGill Pain Questionnaire qualitative histograms.