Conditioned Pain Modulation Evoked by a Mechanical Craniofacial Stimulus Is Not Influenced by Noxious Stimulation of the Temporomandibular Joint

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Aims: *To investigate the influence of noxious stimulation of the temporomandibular joint (TMJ) on conditioned pain modulation (CPM) and the possible influence of gender on such CPM effects in the craniofacial region of humans.* ***Methods:*** *Twenty healthy men and 20 healthy women participated in two sessions. Conditioning stimulation (CS) was standardized mechanical stimulation of pericranial muscles at a pain level of 5 on a 0 to 10 visual analog scale (VAS). Intra-articular electrical stimuli were applied to the left TMJ with an intensity around VAS = 5 (painful session). No electrical stimulation was applied in the control session. Pressure pain threshold (PPT) and pressure pain tolerance threshold (PPTol) were used as responses to pressure (test) stimuli and were assessed in the right masseter muscle and left forearm before and during TMJ stimulation in addition to the CS (during, immediately after, and 10 minutes after CS). PPT and PPTol were analyzed by multilevel analysis of variance.* ***Results:*** *The parameters were not dependent on gender, assessment site, or session, but were dependent on time (PPT, PPTol:* $P < .001$*) with session-time interactions (PPT:* $P < .001$*, PPTol:* $P = .002$*). CS triggered increases in PPT and PPTol (hypoalgesia) in both sessions and without significant differences between sessions or assessment sites during CS (painful session: 49.2 ± 3.7%, control session: 46.0 ± 3.4% for PPT and painful session: 17.7 ± 3.2%, control session: 21.4 ± 3.5% for PPTol).* ***Conclusion:*** *Acute noxious stimulation of the TMJ does not alter the magnitude of CPM effects on masseter muscle pain in either gender. It is suggested that deficiencies in CPM in persistent pain conditions are most likely more related to the duration of clinical pain than the pain per se.* J OROFAC PAIN 2012;26:105–116

Key words: conditioned pain modulation (CPM), experimental craniofacial pain, gender differences, human volunteers, trigeminal system

Temporomandibular disorders is an umbrella term that covers various problems including pain in the masticatory muscles, the temporomandibular joint (TMJ), and associated tissues.[1] The clinical presentation of these problems is more prevalent in women than men.[2] One interesting concept related to persistent pain conditions such as temporomandibular disorders is that they may reflect a dysfunctional or impaired state of endogenous pain-modulatory pathways.[3] One such pain-inhibitory process is "diffuse noxious inhibitory controls" (DNIC).[4,5] DNIC are phenomena whereby the activities of nociceptive neurons in the spinal dorsal horn[4] or trigeminal brainstem nuclei[6,7] are selectively inhibited by noxious conditioning

stimulation (CS) applied outside their excitatory receptive fields.[8] This neurophysiological phenomenon was initially described in animal species[4–11] and subsequently in humans.[11–14] It has recently been suggested that the DNIC-like effects in humans should be termed "Conditioned Pain Modulation" (CPM).[15] Although CPM is defined as a phenomenon through which a CS modulates the effects of a test stimulus, the following study focuses on CPM elicited by painful CS.

Some studies have reported significant gender differences in the magnitude of CPM,[16,17] but other studies have failed to identify robust gender differences.[18,19] Dysfunction of endogenous pain-modulatory systems and/or related facilitatory mechanisms may be evaluated by CPM paradigms and could play an important role in the development and maintenance of persistent musculoskeletal pain conditions.[1,20] In fact, there is some evidence suggesting that dysfunctional modulatory mechanisms are implicated in craniofacial muscle pain conditions such as myofascial temporomandibular disorder pain,[3,21,22] migraine,[23,24] and chronic tension-type headache.[23,25,26]

A recent experimental study in healthy volunteers demonstrated that two concomitant CSs (muscle pain and cold pressor pain) in the spinal region produced less CPM effects compared with either of the CSs given alone, and that men showed greater CPM effects than women.[27] The application of two concomitant CSs could mimic CPM in clinical pain conditions. However, no CPM study in the craniofacial region has been performed to date with concomitant CS inducing experimental pain. Therefore, it would be of interest to study if CPM effects can be impaired by experimentally induced musculoskeletal pain in the craniofacial region. The hypothesis tested was that experimentally induced musculoskeletal pain in the craniofacial region impairs CPM effects.

Recently, a mechanical craniofacial compressive device has been developed, which enables systematic testing of CPM effects.[28] This device produces pain similar to chronic tension-type headache, and is associated with robust CPM effects.[28,29] An experimental craniofacial pain model with repetitive electrical stimulation of the TMJ has also been developed.[30] This method is reliable and allows for the generation of a constant painful input to the TMJ without tissue damage.

The specific aims of the present study were to investigate the influence of noxious stimulation of the TMJ on CPM and the possible influence of gender on such CPM effects in the craniofacial region of humans.

Materials and Methods

Subjects

Twenty healthy men (mean ± SEM age: 25.4 ± 0.9 years, age range: 20 to 38 years) and 20 healthy women (mean ± SEM age: 24.9 ± 0.8 years, age range: 20 to 30 years) participated in the study. None of the subjects had any pain complaints or previous injuries that interfered with normal somatosensory functioning. Informed consent was obtained from all subjects before inclusion. The study followed the Helsinki Declaration and was approved by the local ethics committee (VN20090047).

Experimental TMJ Pain

Experimental TMJ pain was induced by repetitive electrical stimulation of the left TMJ.[30] After the skin over the insertion site was disinfected with alcoholic tissues, two unipolar needle electrodes (20 mm × 0.35 mm, 28G, Alpine Biomed 9013 R0272) were inserted into the left TMJ and placed about 2 to 3 mm apart while the subjects kept their mouths slightly open. The needle electrodes were targeted at the posterior part of the upper joint compartment. Repetitive electrical stimulation (0.5 ms duration, 5 Hz)[30] was used to provide a constant noxious stimulation of the TMJ and was applied for 20 minutes. The intensity of the repetitive electrical stimulation was adjusted individually to reach a painful level around 5 on a 0- to 10-cm visual analog scale (VAS-TMJ). If the pain scores increased, the electrical stimulus intensity was decreased and vice versa. No adjustment of the electrical stimulus intensity was performed during the application of the mechanical craniofacial compressive force (CS). During the electrical stimulation, the intensity of the repetitive electrical stimulation (mA) was noted in 5-minute intervals.

Mechanical CS

The compressive device inducing standardized mechanical craniofacial pain (the CS) (Fig 1)[28,29] was positioned over the vertex and fastened on the four probes (left, occiput, right, forehead, 10-mm radius) around the skull with two centrally joined c-clamps offset from each other by 90 degrees. A strain-gauge force transducer was attached on the four probes and the pressure adjusted over time by using the VAS feedback from the subject. The device was gradually and continuously tightened until the participants scored their instantaneous pain intensity at a target level (VAS-CS = 5) and it was

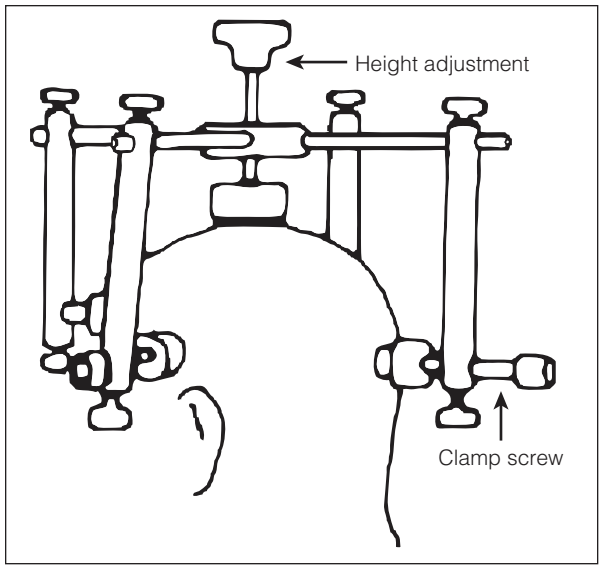

Fig 1 The compressive device for inducing experimental craniofacial pain. The device was set on the vertex. It was height-adjustable by a downwardly directed screw. Compression of the craniofacial region was achieved by tightening four horizontally opposed clamp screws with a force transducer.

maintained for about 7 minutes. The applied forces on the four probes were recorded in newtons (N) and mean values of the four probes were used for further analysis.

Pain Ratings

Subjects continuously rated the pain intensity of the TMJ stimulation on a 0 to 10 electronic VAS (VAS–TMJ: 0 = no pain, 10 = worst pain imaginable) by moving the indicator of the VAS recorder with their right or left hand. The ratings were continuously sampled and stored on a computer every 5 seconds from the start of the needle insertion until the pain ratings returned to zero in both sessions. The VAS-TMJ pain values were used for further analysis.

Subjects reported the pain intensity of the mechanical craniofacial compression by using their right or left hand to place a mark on a 0- to 10-cm paper VAS (VAS–CS: 0 = no pain, 10 = worst pain imaginable). After adjustment of the mechanical compressive device to a VAS–CS value of 5, additional VAS–CS values were obtained after the measurements of the effects of the test stimulation at the masseter and forearm (see below) and after the final removal of the device. The VAS–CS pain values were used for further analysis.

Subjects were asked to fill in the Danish version[31] (for Danish subjects) or English version[32] (except for Danish subjects) of the McGill Pain Questionnaire (MPQ) to obtain a qualitative description of the electrically induced pain of the TMJ in the painful session or related to the insertion of the needles in the control session and the mechanically (CS) induced pain of the craniofacial region. The pain rating indices (PRI) for the different dimensions of pain (sensory [PRI(S)], affective [PRI(A)], evaluative [PRI(E)], miscellaneous [PRI(M)], and total [PRI(T)]) were calculated and used for further analysis.

Test Stimulation

Pressure pain threshold (PPT) and pressure pain tolerance threshold (PPTol) were recorded at the right masseter muscle and the left forearm (flexor carpi radialis muscle) by the application of a pressure algometer (Somedic) as the test stimulus. Measurements were performed in the following sequence: PPT at masseter muscles, PPT at forearm, PPTol at masseter muscle, and PPTol at forearm. The PPT was defined as the amount of pressure (kPa) that the subjects first perceived as being painful, and the PPTol was defined as the most painful pressure (kPa) the subject could tolerate. The algometer probe (1 cm^2 area) was applied with a constant application rate of 30 kPa/s.[27] The subjects had the stop button of the algometer in their right hand and pushed the button to stop the pressure stimulation when the threshold was reached. The PPT measurements at each location were repeated three times with about 1 minute in between (for masseter muscle or forearm, respectively), and the average value was used for further analysis. PPTol was recorded only once at each time point and site to avoid excessive stimulation and sensitization phenomena.

Experimental Protocol

Subjects rested in a chair with an armrest. The two sessions (painful session with electrical TMJ stimulation and control session without TMJ electrical

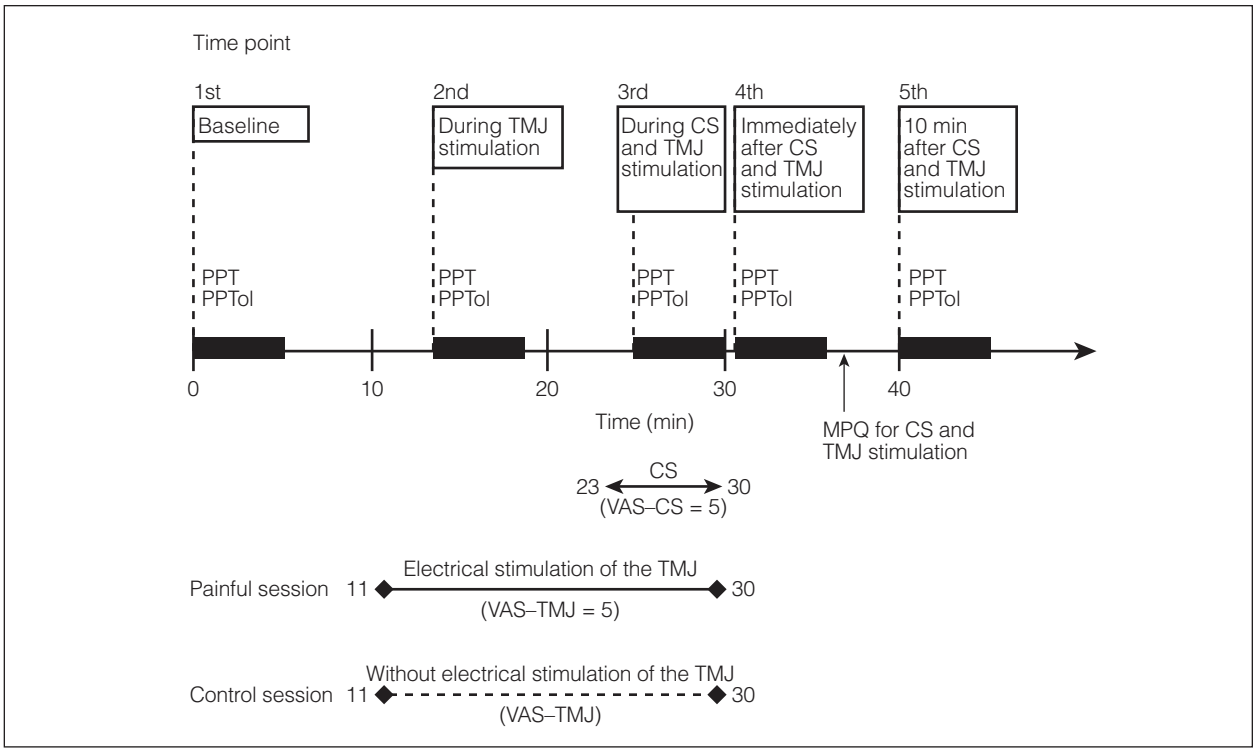

Fig 2 Overview of the study design. PPTs and PPTols at right masseter and left forearm were measured at five time points in each session. In the painful session, the repetitive electrical stimulation was applied to the left TMJ by using two unipolar needle electrodes with the VAS pain intensity = 5 from 11 to 30 minutes after the start of the study. In the control session, two unipolar needle electrodes were inserted into the left TMJ without electrical stimulation. The standardized mechanical compression to the craniofacial region (CS) was applied with the pain intensity of VAS = 5 for about 7 minutes from 23 to 30 minutes after the start of the study. The black bars show the period of PPT and PPTol recording: baseline (before), during TMJ stimulation, during CS and TMJ stimulation, immediately after the end of the CS and TMJ stimulation, and 10 minutes after the end of the CS and TMJ stimulation. Subjects rated the pain intensity of the repetitive electrical TMJ stimulation in the painful session or the pain intensity of the needle electrodes in the control session (VAS–TMJ). Subjects also rated the pain intensity of the CS (VAS–CS). Subjects were asked to fill in the MPQ for the TMJ stimulation and CS after the needle electrodes and mechanical craniofacial compressive device were removed from the craniofacial region.

stimulation) were randomized and separated by at least 1 week. No subjects had previous experience participating in an experiment with needle electrodes. In some of the subjects (n = 6) who were afraid of needle insertion, a control session was performed as the first session to accustom them to needle electrodes. Subjects rated the pain intensity of electrical TMJ stimulation (VAS–TMJ). In the painful session, experimental TMJ pain was induced by repetitive TMJ electrical stimulation from 11 to 30 minutes after the start of the study with an intensity adjusted to produce a VAS value of 5. In the control session, no electrical stimulation was applied. As CS, the mechanical craniofacial compressive device was applied from 23 to 30 minutes after the start of the study and adjusted to VAS = 5. Subjects were also asked to rate the pain intensity of the CS (VAS–CS). PPT and PPTol were determined in the right masseter muscle and left forearm. These test stimulus thresholds were recorded at five time points: first, before TMJ stimulation (baseline values, from 0 to 5 minutes after the start of the study); second, during TMJ stimulation (from 13 to 18 minutes); third, during CS and TMJ stimulation (from 25 to 30 minutes); fourth, immediately after the end of the CS and TMJ stimulation (within 60 seconds after the end of the PPT and PPTol recordings during CS and TMJ stimulation, from 31 to 36 minutes); and fifth, 10 minutes after the end of the CS and TMJ stimulation (from 40 to 45 minutes). The third test stimulus recording was performed after the VAS–CS reached 5 and remained stable for 2 minutes. The needle electrodes and the mechanical craniofacial compressive device were removed immediately after the third test stimulus recording. After the fourth test stimulus recording, subjects were asked to complete the MPQ for the TMJ stimulation and for the CS (Fig 2).

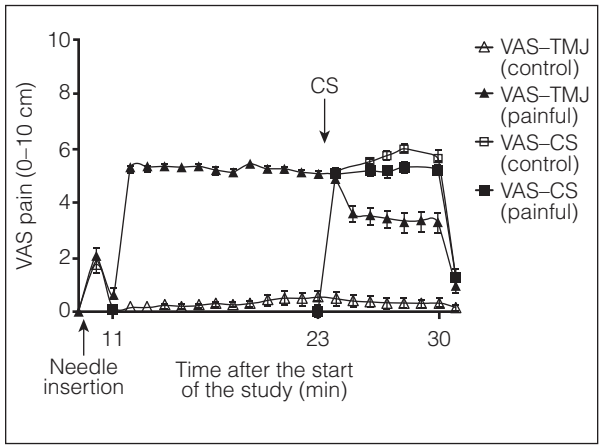

Fig 3 Continuous VAS ratings of the pain intensity of the repetitive TMJ electrical stimulation in the painful session (VAS–TMJ, painful), the pain intensity of the needle electrodes in the control session (VAS–TMJ, control), and the pain intensity of craniofacial compression (CS) (VAS–CS) in two sessions (control, painful) from all the subjects (n = 40) (mean ± SEM). The arrows show the needle insertion and the application of mechanical craniofacial pain (CS), respectively. Though the mere insertion of the needle electrodes elicited pain in some of the subjects in both sessions, pain decreased to a level around VAS = 1 in a few minutes. In the control session, the VAS–TMJ pain values after the needle insertion were low (under VAS = 1) and stable without gender difference. In the painful session, the VAS-TMJ pain values tended to decrease during CS. Though the tightening of the compressive device was stopped as soon as the pain intensity reached VAS–CS = 5, the pain intensity continued to increase gradually for the duration of the compression.

Statistical Analysis

The Kolmogorv-Smirnov test was applied to verify the normal distribution of data. As VAS values for the needle insertion could not pass the Kolmogorv-Smirnov test ($P < .05$), the Mann-Whitney U test was used.

The intensities of the repetitive electrical TMJ stimulation (mA) and VAS–TMJ during CS in the painful session were compared between genders by an unpaired t test.

The following tests were performed with repeated measures ANOVAs. The VAS–TMJ pain values, the VAS–CS pain values, the forces applied by the mechanical craniofacial compressive device, and the PRI for electrical TMJ stimulation and CS were analyzed by two-way ANOVA with gender (men, women) as between-group factor and session (control, painful) as the repeated factor.

To examine the effect of gender, assessment site, and session on test stimulus evaluation, absolute PPT and PPTol values at baseline were analyzed by three-way ANOVA: gender as between-group factor, and assessment site (masseter, forearm) and session (control, painful) as repeated factors. In order to account for baseline differences between assessment sites and genders, the PPT and PPTol values were normalized to the baseline values. Then to examine the following effects (gender, assessment site, session, and time) on CPM associated with CS, the normalized PPT and PPTol values were analyzed by a four-way ANOVA with gender as between-group factor and assessment site (masseter, forearm), session (control, painful), and time (baseline, during TMJ stimulation, during CS, immediately after the CS, 10 minutes after the end of the CS) as repeated measures. For post-hoc analyses, Bonferroni-corrected paired t tests were performed. All data are presented as mean values and standard errors of mean (mean ± SEM). The level of significance was set at $P < .05$.

Results

Experimental TMJ Pain

The intensity of the repetitive electrical TMJ stimulation required to evoke a pain intensity around VAS = 5 was 1.5 ± 0.2 mA in men and 1.5 ± 0.2 mA in women, with no differences between genders ($P = .985$).

Continuous VAS–TMJ pain ratings in the two sessions (control, painful) from all the subjects (n = 40) are shown in Fig 3. In both sessions, the mere insertion of the needle electrodes elicited pain in some of the subjects (VAS 1.9 ± 0.2 cm) without a session ($P = .303$) or gender difference ($P = .141$). However, pain did not last long and decreased to a level around VAS = 1 in a few minutes.

The VAS–TMJ pain values (before the application of CS) were significantly dependent on the session ($F = 1,356.403, P < .001$) but not on gender ($F = 1.227$, $P = .275$). As expected, the VAS–TMJ pain values

were significantly higher in the painful session (4.9 ± 0.1 cm) compared to the control session (0.3 ± 0.1 cm, $P < .001$). In the painful session, the VAS–TMJ pain values tended to decrease during CS (Fig 3). The VAS–TMJ pain values during CS were 3.4 ± 0.3 cm in men and 3.9 ± 0.5 cm in women ($P = .462$).

The [PRI(S)], [PRI(A)], [PRI(E)], [PRI(M)], and [PRI(T)] evoked by electrical TMJ stimulation were significantly dependent on the session ($F = 89.619$, $P < .001$; $F = 34.274$, $P < 0.001$; $F = 68.450$, $P < .001$; $F = 35.248$, $P < .001$; $F = 92.449$, $P < .001$) but not on gender ($F = .165$, $P = .687$; $F = .490$, $P = .488$; $F = .468$, $P = .498$; $F = 1.346$, $P = .253$; $F = .122$, $P = .729$, respectively). As expected, the [PRI(S)], [PRI(A)], [PRI(E)], [PRI(M)], and [PRI(T)] were significantly higher in the painful session (13.10 ± 1.13, 2.85 ± 0.42, 2.23 ± 0.25, 4.00 ± 0.56, 22.18 ± 1.86) compared to the control session (1.98 ± 0.62, 0.33 ± 0.16, 0.38 ± 0.13, 0.68 ± 0.22, 3.35 ± 0.90), respectively ($P < .001$).

Mechanical CS-Evoked Pain

There were no significant differences in the applied forces of the compressive device, neither between sessions ($F = .003$, $P = .957$) nor genders ($F = 1.369$, $P = .249$) (left: 14.8 ± 1.4 N, occiput: 16.4 ± 1.0 N, right: 16.4 ± 1.4 N, forehead: 16.5 ± 1.9 N, and mean: 16.0 ± 1.1 N).

VAS–CS pain ratings during mechanical craniofacial compression in the two sessions (control, painful) from all the subjects (n = 40) are shown in Fig 3. The positioning of the compressive device on the head did not elicit pain in any of the subjects and in the sessions before the application of the compression. The compression triggered craniofacial pain and it was reported as dull, bilateral, and strong headache similar to the quality of chronic tension-type headache.[33] Though the tightening of the compression was stopped as soon as the pain intensity reached a VAS value of 5, the pain intensity continued to increase gradually for the duration of the compression in accordance with previous studies.[29,30]

The VAS-CS pain values were significantly dependent on the session ($F = 7.721$, $P = .008$) but not on gender ($F = .331$, $P = .569$). The VAS–CS pain values were significantly higher in the control session (5.6 ± 0.1 cm) compared to the painful session (5.2 ± 0.1 cm) ($P = .008$) (Fig 3).

Regarding the PRI evoked by mechanical craniofacial compression, there was no significant difference in the [PRI(S)], [PRI(A)], [PRI(E)], [PRI(M)], and [PRI(T)], neither between genders ($F = .002$, $P = .966$; $F = .864$, $P = .359$; $F = .080$, $P = .779$; $F = .049$, $P = .826$; $F = .078$, $P = .782$) nor sessions ($F = .002$, $P = .962$; $F = .186$, $P = .668$; $F = 3.367$, $P = .074$; $F = 1.096$, $P = .302$; $F = .000$, $P = 1.000$) (13.60 ± 0.88, 2.90 ± 0.34, 1.96 ± 0.17, 4.59 ± 0.39, 23.05 ± 1.46, respectively).

Baseline Values of Test Stimulus

Baseline Values of PPT. The absolute PPT values at baseline were tested with ANOVAs and revealed an effect of gender ($F = 12.932$, $P < .001$), with significantly higher PPT in men (289.5 ± 16.7 kPa) than women (231.6 ± 13.6 kPa) ($P < .001$). Moreover there were significant differences between assessment sites ($F = 403.254$, $P < .001$, forearm: 369.9 ± 12.8 kPa, masseter: 151.1 ± 4.1 kPa), with a significant gender and assessment site interaction ($F = 9.579$, $P = .004$). Post-hoc tests showed significantly higher PPT at the male forearm (415.7 ± 16.7 kPa) compared to the male masseter (163.2 ± 5.5 kPa, $P < .001$), female forearm (324.1 ± 16.6 kPa, $P = .002$), and female masseter (139.1 ± 5.5 kPa, $P < .001$). There was no effect of session ($F = .057$, $P = .813$). A normalization of the PPT values to baseline recordings was performed to compare directly the effects of assessment site and gender on CS-induced threshold changes.

Baseline Values of PPTol. The effects of experimental factors on absolute PPTol values at baseline were tested with ANOVAs and indicated significant differences between genders ($F = 27.422$, $P < .001$), with significantly higher PPTol in men (651.6 ± 41.7 kPa) compared to women (395.7 ± 23.9 kPa) ($P < .001$). Again there were significant differences between assessment sites ($F = 248.162$, $P < .001$, forearm: 750.0 ± 35.4 kPa, masseter: 297.3 ± 13.3 kPa), with a significant gender and assessment site interaction ($F = 18.900$, $P < .001$). Post-hoc tests showed significantly higher PPTol at the male forearm (940.4 ± 49.1 kPa) compared to the male masseter (362.8 ± 19.4 kPa, $P < .001$), female forearm (559.6 ± 28.5 kPa, $P < .001$), and female masseter (231.8 ± 11.0 kPa, $P < .001$). There was no effect of session ($F = .018$, $P = .894$). Thus, the PPTol values were normalized to directly compare the effects of CS.

Effect of TMJ Noxious Stimulation and CS on Normalized Values of Test Stimulus

Normalized PPT Values. ANOVAs of normalized PPT values indicated no main effects of gender ($F = .676$, $P = .416$), assessment site ($F = 3.669$, $P = .063$), or session ($F = 1.478$, $P = .232$), but a significant time effect ($F = 110.437$, $P < .001$) with a significant assessment site-time interaction ($F = 8.201$, $P < .001$) and session-time interaction ($F = 4.939$,

$P < .001$). Post-hoc tests revealed that the normalized PPT increased at the time point during TMJ stimulation (13.3% ± 1.6%, $P < .001$), during CS (47.6% ± 2.5%, $P < .001$), immediately after the CS (27.0% ± 1.9%, $P < .001$), and 10 minutes after the CS (aftereffects) (10.4% ± 1.4%, $P < .001$) compared with baseline values.

At the masseter, there were significant changes in PPT increases at the time point during TMJ stimulation (13.3% ± 2.3%, $P < .001$), during the CS (41.8% ± 3.4%, $P < .001$), immediately after the CS (24.5% ± 2.3%, $P < .001$), and 10 minutes after the CS (10.8% ± 1.9%, $P < .001$) compared with baseline values. At the forearm, there were significant changes in PPT increases at the time point during TMJ stimulation (13.2% ± 2.3%, $P < .001$), during the CS (53.4% ± 3.6%, $P < .001$), immediately after the CS (29.6% ± 2.9%, $P < .001$), and 10 minutes after the CS (9.9% ± 2.1%, $P < .001$) compared with baseline values. The assessment site-time interaction showed that the values at the forearm were associated with significantly higher PPT increases (53.4% ± 3.6%) compared to the masseter (41.8% ± 3.4%) during the CS ($P = .001$), but there were no other significant differences at other time points.

In the control session, there were significant changes in PPT increases during the CS (46.0% ± 3.4%, $P < .001$), immediately after the CS (26.3% ± 2.7%, $P < .001$), and 10 minutes after the CS (12.7% ± 2.1%, $P < .001$) compared with baseline values. In the painful session, there were significant changes in PPT increases at the time point during TMJ stimulation (20.8% ± 2.4%, $P < .001$), during CS (49.2% ± 3.7%, $P < .001$), immediately after the CS (27.8% ± 2.6%, $P < .001$), and 10 minutes after the CS (8.1% ± 1.9%, $P = .007$) compared with baseline values (Fig 4a). In the painful session there were also significant differences in PPT increases between the time point during TMJ stimulation (20.8% ± 2.4%) and during CS (49.2% ± 3.7%, $P < .001$). The session-time interaction showed that the values in the painful session were associated with significantly higher PPT increases (20.8% ± 2.4%) compared to the control session (5.7% ± 1.9%) at the time point during TMJ stimulation ($P < .001$), but no other significant differences between sessions were observed at other time points (Figs 4a, 4c, and 4e).

Normalized PPTol Values. The normalized PPTol values were tested with ANOVAs and indicated no main effects of gender ($F = 1.408$, $P = .243$), assessment site ($F = .591$, $P = .447$), or session ($F = .241$, $P = .627$), but a significant time effect ($F = 21.585$, $P < .001$) with a significant session-time interaction ($F = 4.569$, $P = .002$). Post-hoc tests revealed that the normalized PPTol increased during the CS (19.5% ± 2.4%, $P < .001$) and immediately after the CS (7.6% ± 1.9%, $P = .003$) compared with baseline values.

In the control session, there were significant changes in PPTol increases during the CS (21.4% ± 3.5%, $P < .001$) compared with baseline values. In the painful session, there were significant changes in PPTol increases at the time point during TMJ stimulation (11.1% ± 2.5%, $P = .003$) and during CS (17.7% ± 3.2%, $P < .001$) compared with baseline values (Fig 4d). The session-time interaction showed that there were significantly higher increases in the painful session (11.1% ± 2.5%) compared to the control session (–0.2% ± 2.4%) at the time point during TMJ stimulation ($P = .001$), but no other significant differences between sessions were observed at other time points (Figs 4b, 4d, and 4f).

Overall, the normalized PPT and PPTol values were not dependent on gender, assessment site, or session, but dependent on time with session-time interactions. However, there were no significant differences in the magnitude of test stimulus increases between sessions during CS (painful session: 49.2% ± 3.7%, control session: 46.0% ± 3.4%, $P = .579$ for PPT and painful session: 17.7% ± 3.2%, control session: 21.4% ± 3.5%, $P = .445$ for PPTol) (Figs 4a and 4b).

Discussion

This study showed that acute experimental craniofacial pain evoked by repetitive electrical TMJ stimulation did not alter the magnitude of CPM evoked by painful mechanical stimulation of the craniofacial region. Moreover, there were no gender differences in the pain-modulatory effects with the current experimental pain model.

Methodological Considerations

There are various methodologies to evoke CPM.[34] It has recently been reported that the mechanical craniofacial pressure as applied in this study is able to trigger CPM consistently and induces pain like chronic tension-type headache.[28,29] Pressure pain thresholds (PPT and PPTol) are efficient to evaluate CPM responses.[20,27–29] Therefore, this model was chosen in the present study.

Electrical stimuli excite the full spectrum of peripheral nerve fibers.[35] Insertion of two needle electrodes into the posterior part of the TMJ avoided tonic contractions of the ipsilateral jaw muscles or blink responses. Electrical TMJ stimulation produced

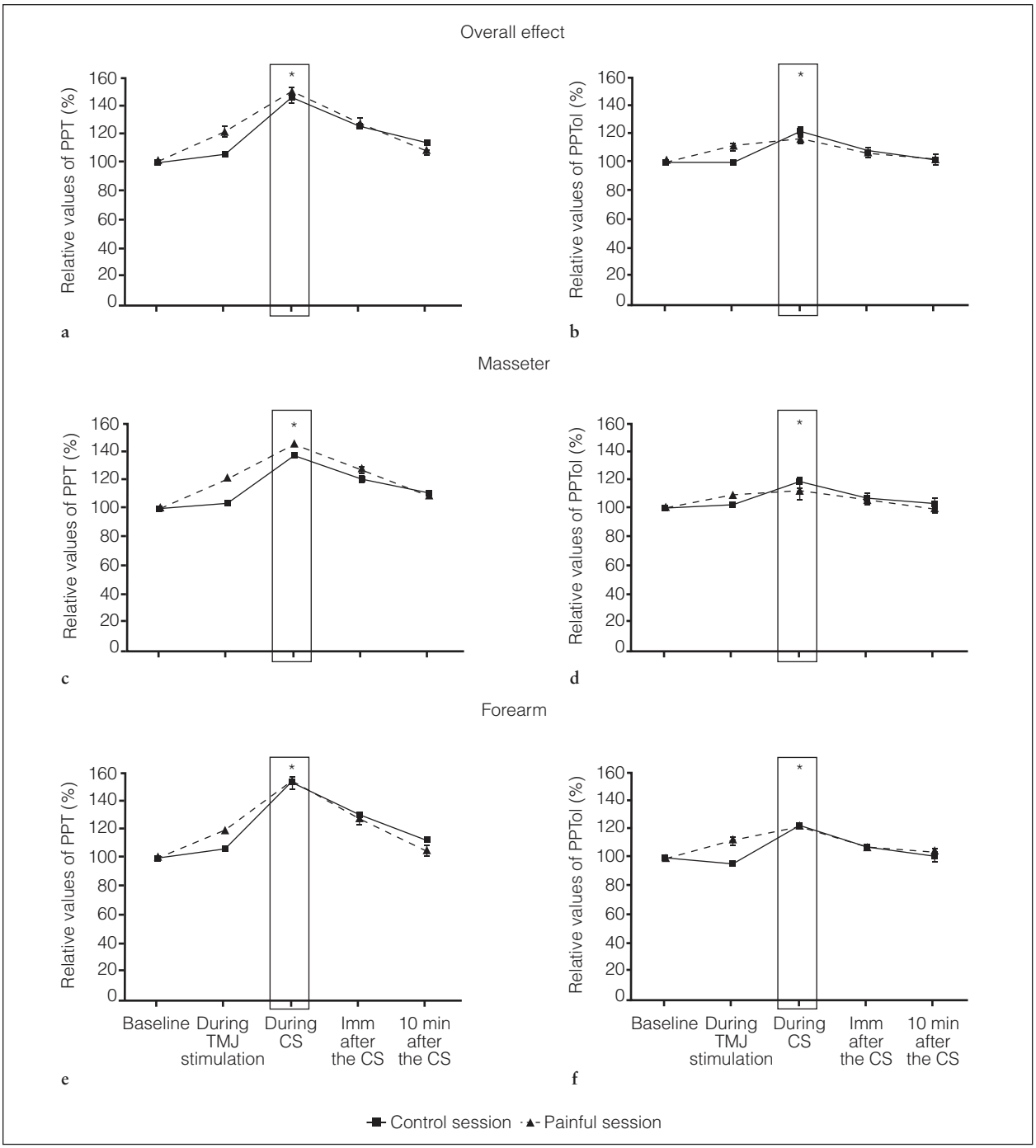

Fig 4 Relative values of the PPTs *(a, c, and e)* and PPTols *(b, d, and f)* (%, mean ± SEM) at five time points in the control and the painful sessions from all the subjects (n = 40). In *(a)* and *(b)*, the increases are the mean values for two assessment sites (masseter and forearm). In *(c)* and *(d)* and *(e)* and *(f)*, the increases are the values assessed at the masseter *(c and d)* and forearm *(e and f)*. *Indicates significant increases of normalized PPT and PPTol values compared with baseline values ($P < .001$) in both sessions (overall effect from four-way ANOVA and post-hoc tests). There were no significant differences between sessions in the increment of PPT and PPTol during CS. Imm = immediately.

a constant level of pain, which only required a minor adjustment of the stimulus intensities in accordance with a previous study,[29] and no muscle twitching was observed in most cases. The procedure followed the general principles for intra-articular injections, and the tip of the needle electrodes was most likely placed in the retroarticular pad, which consists of an upper and lower layer and the genum vasculosum with vessels, fat cells, and nerve endings.[1] The sensory innervation of this part is mainly through the auriculotemporal nerve, but the posterior deep temporal nerve and masseteric nerve also contribute.[1] There could be a possible confounding influence of the needle insertion due to an acupuncture-like effect. CPM may be involved in the analgesic mechanism of acupuncture[36,37] through bulbospinal pathways.[38] However, it has been indicated that prolonged repetitive electrical stimulation of the TMJ was associated with increased sensitivity in the areas adjacent to the TMJ.[30] Finally, the influence of the order of the experiment on the results needs to be noted. Although the control session was performed as the first session in some subjects (2 men and 4 women), the two sessions were randomized in all other subjects (18 men and 16 women). Therefore, the order of the sessions is unlikely to have influenced the results.

Influence of CS Characteristics on CPM

In this study, robust CPM effects of 46.0% for PPT and 21.4% for PPTol were found in the control session, which were also demonstrated in previous studies.[28,29] Various experimental pain modalities such as thermal cold,[27,39] heat,[14,40] electrical,[41] ischemic,[21,39,42] and chemical[43,44] have been applied to estimate CPM in healthy subjects.[34] The effects of CPM are known to differ depending on the magnitude and nature of the CS and stimulated nerve fibers.[10,13] A stronger CS results in greater CPM.[10–13,45–47]

This study showed after-effects of CPM associated with CS on the PPT, and these lasted 10 minutes. After-effects have been reported to last 6 to 9 minutes after the end of a thermal CS[48] and 15 minutes after an ischemic CS.[42] On the other hand, CPM may not outlast the duration of a thermal CS[14,49] because of the moderate intensity of the CS.[14] After-effects of CPM differ depending on the strength and modality of the CS[45,50] through centrally mediated mechanisms.[44,50,51]

Influence of TMJ Noxious Stimulation on CPM

In the present study with acute experimental TMJ pain, CPM evoked by the CS varied from 17.7% to 49.2% in the painful session and there was no significant difference compared to the control session without the experimental TMJ stimulation.

In persistent musculoskeletal pain conditions, the balance between supraspinal facilitation and inhibition of pain shifts towards an overall decrease in inhibition in the spinal region.[52,53] A similar phenomenon, a dysfunction of CPM in persistent pain patients, has been reported in the craniofacial region.[21–26] In temporomandibular disorder patients, CS is not associated with robust inhibitory responses resulting in CPM.[21,22] Furthermore, migraineurs and chronic tension-type headache patients showed facilitatory CPM (hyperalgesia, 31% and 40%, respectively) contrary to healthy subjects who showed inhibitory CPM (hypoalgesia, 30%).[26]

A recent human experimental pain study with PPTs around the knee joints found that CPM induced by two concomitant CSs was less efficient than when induced by a single CS (cold pressor test to the left hand or hypertonic saline injection into the left tibialis anterior).[27] These findings indicate that heterosegmental stimuli may interfere and disturb the balance between descending inhibition and facilitation, at least in the spinal system.[27] However, in the present study in the craniofacial region, there was no significant decrease in the CPM effect even though an experimental painful stimulus was applied to the TMJ.

One possible explanation could be the influence of the applied site of the CS and the assessment site of the test stimulus (segmental or extrasegmental). In the previous report,[27] the PPTs were measured extrasegmentally in relation to the cold pressor test but segmentally with respect to the experimental muscle pain. In the present study, both the CS and the experimental painful stimulus were applied segmentally (craniofacial region) and PPT was evaluated segmentally (masseter) and extrasegmentally (forearm). Concerning the experimental site modality, site-dependent decreases of the pain intensity at different spinal heterotopic sites have been reported.[54] Svensson et al[54] showed ipsilateral homotopic application of intramuscular hypertonic saline as the CS slightly increased the perceived pain intensity of electrical stimulation, indicating spatial summation. Graven-Nielsen et al[55] found no change in the pain intensity following homotopic painful pressure stimulation, whereas heterotopic CS caused CPM. Contrary to these studies, similar effects of CPM between heterotopic and homotopic sites have been reported.[56] Rosen et al[57] demonstrated tissue and site differences as well as stimulus-specific differences in CPM in the craniofacial region. These differences could also be due to the experimental design,

where the test stimulus was assessed, and where the concomitant experimental painful stimulus was applied, and finally also due to the modalities of the test stimulus and CS.[34]

Another possible explanation could be the influence of expectation. Subjects who expected an analgesic effect from the CS showed larger CPM, but the CPM could be completely blocked by antianalgesic expectancy.[58] The expectations can modulate the activation of endogenous pain inhibitory systems affecting spinal and cortical nociceptive responses. Thus, subjects unfamiliar with the experimental stimuli might expect a painful response to two concomitantly applied painful stimuli, which could lead to modification of the interaction of the two experimental painful stimuli.

Finally, the possibility of ceiling effects on CPM needs to be considered. However, in the painful session of the present study, the mechanical CS together with TMJ stimulation induced significantly larger CPM compared to TMJ stimulation per se, that is, there were no ceiling effects in the painful session. Hence, the lack of differences in CPM between the sessions is unlikely to result from ceiling effects.

In this study, the VAS–TMJ pain values tended to decrease during application of CS. This observation suggests that the CS triggered CPM on both the test stimulus and the TMJ stimulation. Moreover, the VAS–CS pain values in the painful session were significantly lower than in the control session even though there were no significant differences between sessions in the mean applied forces of the device and the pain modality of the MPQ evaluation, ie, it also implies that the painful TMJ stimulation triggered CPM on the effects of the CS. This phenomenon indicates a bidirectional mechanism of CPM that, to the authors' knowledge, has not been reported before. Overall, the result implies that two experimental painful stimuli may interact in a complex manner, perhaps analogous to persistent pain conditions where changes in the balance between descending inhibitory and facilitatory influences may occur.

Gender Effects on CPM

This study showed no gender differences in the perceived pain derived from the MPQ related to repetitive TMJ electrical stimulation or mechanical craniofacial compression. Furthermore, no gender differences in CPM were demonstrated, in agreement with previous studies,[18,19,47,56,59,60] although other studies have reported gender differences.[16,17,20,27,46,61]

Recent reviews of sex-dependent differences in pain responses have reported that there seems to be greater responses in women, particularly to stimuli of longer duration.[2,62,63] Hormonal influences have been implicated in a variety of predominantly female pain conditions.[63,64] Use of oral contraceptives is associated with significantly increased risk of temporomandibular disorders, and female reproductive hormones may play a role in craniofacial pain.[65] CPM effects can also be affected by hormonal factors in women (eg, a larger CPM in the ovulatory phase).[47,66] The hormonal cycle was not recorded in the present study, but it appears unlikely that this may have influenced the outcome of the results significantly because a paired design with the subjects as their own control was used.

Conclusions

This study has shown that acute TMJ noxious stimulation did not alter CPM evoked by tonic painful mechanical stimulation of the craniofacial region. Moreover, there were no gender differences in the CPM. It is suggested that deficiencies in CPM in persistent pain conditions may be more related to the duration of the clinical pain than the pain per se.

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