Simultaneous Noxious Stimulation of the Human Anterior Temporalis and Masseter Muscles. Part I: Effects on Jaw Movements

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Aims: To test the hypotheses that, in comparison to control (isotonic saline), simultaneous noxious stimulation (hypertonic saline) of the masseter and anterior temporalis muscles would result in (1) reductions in amplitude and velocity of jaw movements during standardized open/close jaw movements and during free and standardized chewing and (2) changes in amplitude and velocity of jaw movements that relate to higher levels of negative mood or pain-related thoughts.

Methods: Standardized open/close and free and standardized chewing were recorded in 15 asymptomatic participants in three blocks: block 1 (baseline), block 2 (during 5% hypertonic or 0.9% isotonic saline infusion into the right masseter and anterior temporalis muscles simultaneously), and block 3 (infusion sequence reversed). The Depression, Anxiety, and Stress Scale (DASS-21) and the Pain Catastrophizing Scale (PCS) were completed by the participants before the experiment, and the PCS was completed after the experiment. The amplitude and velocity of opening and closing movements for each task were compared between blocks (repeated-measures analysis of variance). Spearman rank correlation coefficient was used to explore correlations. Statistical significance was considered to be $P < .05$.

Results: In comparison to isotonic saline control, hypertonic saline resulted in significantly smaller opening and closing amplitudes and lower velocity during closing in free chewing, but no significant effects in the open/close task or standardized chewing. There were significant correlations between PCS scores and amplitude or velocity during isotonic saline and baseline, but not hypertonic saline. Conclusion: The pain-related reduction in amplitude and/or velocity of free chewing is consistent with the Pain Adaptation Model, but the absence of effects on the open/close task and standardized chewing is not. The few significant correlations between psychologic variables and jaw movement may reflect the low scores. J Oral Facial Pain Headache 2019;33:413–425. doi: 10.11607/ofph.2299

Keywords: facial pain, jaw, mastication, masticatory muscles, movement, pain measurement

Pain has effects on movements and muscle activity, but the mechanisms involved in the pain-motor interaction are not fully understood. Two theories have dominated the relevant literature: the Vicious Cycle Theory and the Pain Adaptation Model. Both theories implicate segmental circuitry alone in the stereotypical changes in muscle activity during pain. Although aspects of this segmental circuitry have been well characterized in animal experimental pain models, many datasets suggest that these earlier theories are too simplistic to account for the diversity of motor effects associated with experimental or clinical pain, as they cannot explain possible modifying influences (eg, psychologic distress associated with chronic pain states). These issues have led to more recent theories being proposed.

Valuable insights as to the effects of pain on movement and muscle activity have been provided by human and animal experimental pain models, although these experimental studies are generally considered to lack some of the psychologic distress usually associated with chronic pain states. In the jaw motor system, many human and animal experimental pain models have used noxious stimulation of the masseter muscle as...
a representative model of jaw muscle pain. In general, many of these studies report that pain results in reductions in the kinematics of jaw movements or the magnitudes of bite forces, in minimal or no effects, or in effects that may vary with the task performed or the phases of a task.

Patients with some forms of chronic orofacial pain (e.g., temporomandibular disorders [TMD] or fibromyalgia) can report pain and tenderness not only in the region of the masseter muscle but also in the temporalis muscle, and the Diagnostic Criteria for Temporomandibular Disorders (DC/TMD) uses these two muscles as the main muscles to be examined in deriving Axis I diagnoses. It has also been shown that pressure pain thresholds established over the masseter and temporalis muscles can be significantly lower in TMD patients in comparison to controls. There is, however, no information as to the effects of simultaneous experimental noxious stimulation at these two jaw muscle sites on jaw movement and jaw muscle activity. Earlier studies have shown that hypertonic saline injections (0.2 mL) into bilateral masseter muscles resulted in no effects on maximum displacements during chewing and no effects on the duration of opening and closing phases.

Given that some patients with TMD pain report pain not only in the masseter but also the temporalis muscle, an experimental model that results in noxious stimulation at more than one site may provide findings that may be more clinically relevant for some TMD patients. There is also reason to consider that it may not be possible to extrapolate the electromyographic (EMG) and jaw movement effects observed with single-site muscle injections to understand these effects with simultaneous noxious stimulation at two jaw muscle sites. First, noxious stimulation at two jaw muscle sites is likely to be associated with enhanced central sensitization processes given the extensive convergence of nociceptive afferents from multiple orofacial sites—including multiple muscle sites—onto neurons within the trigeminal brainstem sensory nuclear complex. Second, given the distributed representation of the face, jaw, and tongue muscles within the face primary motor cortex, simultaneous noxious stimulation at two jaw muscle sites would be likely to result in decreases in cortical excitability over a larger area of the face primary motor cortex than would be derived from noxious stimulation at a single jaw muscle site. As the face primary motor cortex plays a crucial role in the generation and/or control of both voluntary jaw movements and chewing movements, these possible and more extensive cortical effects would be likely to result in more pronounced effects on jaw movements and jaw muscle activity than observed with single-site muscle injections. Third, there is evidence from clinical and experimental pain studies that differences in pain location may influence the motor effects observed. Thus, noxious stimulation of the masseter muscle resulted in effects on jaw movement and jaw muscle EMG activity in some tasks that were different in comparison to the effects observed with noxious stimulation of the temporalis muscle.

Other factors to consider in the pain-motor interaction are psychologic factors. There is evidence that psychologic factors play a role in the initiation and progression not only of TMD but also of other chronic pain conditions, and associations have been demonstrated between some psychologic constructs, such as depression and pain catastrophizing, and some kinematic variables in clinical or experimental pain. Therefore, the aim of the present study was to test the hypotheses that, in comparison to control (isotonic saline), simultaneous noxious stimulation (hypertonic saline) of the masseter and anterior temporalis muscles would result in reductions in the amplitude and velocity of jaw movements during standardized open/close jaw movements and during free and standardized chewing and would result in changes in the amplitude and velocity of jaw movements that relate to higher levels of negative mood or pain-related thoughts.

Materials and Methods

A total of 15 asymptomatic participants (age 29 to 55 years, 14 men, 1 woman) volunteered. Exclusion criteria were: a TMD diagnosis by a calibrated examiner following the Research Diagnostic Criteria for Temporomandibular Disorders (RDC/TMD); a current history of an ongoing pain condition, serious systemic disease, analgesic, antidepressant, or other drug that might interfere with central nervous system functioning; and presence of dentures, current orthodontic treatment, or a large overbite and/or overjet. The experiment was performed over 3 to 4 hours in one session. Ethics approval was obtained from the Western Sydney Area Health Service Human Ethics Committee of Westmead Hospital and the Human Ethics Committee of the University of Sydney, and all participants gave written informed consent prior to enrolling in the study. Many of the approaches used for jaw tracking, EMG recording (second paper in this series), and experimental pain are similar to those detailed in previous publications by this group.

Figure 1 shows an overview of the experimental paradigm, which consisted of three blocks: block 1 (baseline, prior to infusion), block 2 (hypertonic saline or isotonic saline infusion), and block 3 (isotonic saline or hypertonic saline infusion). The sequence of infu-
tion was alternated between participants. The blocks involving infusion of hypertonic saline were termed the noxious stimulation blocks, and the blocks involving infusion of isotonic saline were termed the control blocks. In each of the three blocks, recordings of jaw movement (analyzed in the present paper) and jaw muscle activity (analyzed in companion paper) were made while jaw movement tasks were carried out.

**Measures of Mood and Pain-Related Cognitions**

Prior to experimental recordings, the Depression, Anxiety, and Stress Scales (DASS-21)\(^46\) and the Pain Catastrophizing Scale (PCS)\(^47\) were completed by all participants. The DASS-21 measures the levels of depression, anxiety, and stress experienced over the past week through the self-report of applicability of 21 statements, 7 for each dimension. Statements are summed to provide total scores for each scale. The PCS is a 13-item set of statements that provides a total score for thoughts and feelings about a painful experience, and the items are expressed in terms of magnification, rumination, and helplessness. The total score ranges from 0 to 52. The PCS was administered both before and after the experiment. For the PCS administered before, participants were asked to recall a past pain experience; for the PCS administered after, participants recalled the experimental pain just experienced.

**Jaw Movement Recording**

Jaw movement was recorded with an optoelectronic jaw tracking system (JAWS3D or JAWS2K; sampling rate 67 samples/second or 200 samples/second, respectively; resolution: ~0.1 mm). Both systems have lightweight target frames supporting light-emitting diodes (LEDs), which were secured to clutches on the maxillary and mandibular teeth so that jaw displacement could be monitored by charge-coupled devices (CCDs). The clutches were adapted to the labial surfaces of 3 to 4 right anterior teeth with self-curing acrylic resin (DuraLay, Reliance Dental), trimmed to avoid any occlusal interference, and attached with cyanoacrylate adhesive (SupaGlue) after thoroughly drying the teeth. The displacement of the mandibular mid-incisor point—ie, the point between the incisal edges of the mandibular central incisor teeth—was displayed on a video screen as a dot for visual feedback for standardizing some of the jaw tasks (see below). For subsequent off-line analysis, the jaw tracking systems recorded displacement (mm) of the mid-incisor point along three orthogonal axes: x axis (posterior-anterior), y axis (medial-lateral), and z axis (inferior-superior). In each participant, EMG activity was also recorded with surface electrodes from the right and left anterior temporalis and masseter muscles and the right digastric muscle. The EMG data will be reported in the next paper in this series.\(^46\)

**Jaw Tasks**

Participants sat upright without head support. A trial represents a single repetition of a task, and each trial commenced with the jaw at the postural or rest position. The jaw tasks that were repeated in each of the three blocks were as follows.

- **Postural Jaw Position.** Participants swallowed and relaxed their jaw with lips lightly touching and teeth slightly apart for 15 seconds (one trial).

- **Standardized Open/Close Jaw Movement.** At the beginning of a trial, participants started at the postural jaw position and then after about 2 seconds moved the mid-incisor point dot to match as closely as possible the speed of another set of LEDs (target LEDs) that were placed to the side of the mid-incisor point dot display in the z axis (ie, vertical). To determine the target amplitude of opening, participants initially opened their jaws to 20 mm of opening displacement from the postural position. This task was repeated five times (five trials) with 30 seconds of rest between each trial, and each trial was about 22 seconds in duration. The sequential illumination of the LEDs was driven by Spike2 software (micro1401, Cambridge Electronic Design) and resulted in an ideal movement speed of 2.2 mm/second. Following the

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Fig 1 Experimental procedure performed on the day of the experiment. All questionnaires, except the McGill Pain Questionnaire (MPQ), were completed first (Pre). Tasks were repeated in 3 blocks each ~10 minutes long, with 10-minute rest periods between each block (repeated-measures design). Hypertonic saline or isotonic saline was infused during blocks 2 and 3, with the sequence alternated between successive participants. Tasks = standardized open/close, standardized chewing, and free chewing; VAS = visual analog scale.
opening phase, the participant held the jaw steady at the target opening displacement for 2 to 3 seconds (holding phase) and then moved the jaw back to the postural jaw position (closing phase), again following the illuminated LED sequence.

**Free Chewing.** Participants softened a piece of sugar-free chewing gum (0.14 g) (EXTRA, The Wrigley Company) on the left side for about 30 seconds. Participants started a trial with their jaw at the postural position, and after about 2 seconds, they were asked to chew the gum naturally on the right side for 15 seconds. Two trials were performed of free chewing.

**Standardized Chewing.** Participants softened a new piece of chewing gum (as above), and after the initial approximately 2-second period with the jaw at the postural position, chewed it on the right side following the timing only of the target LEDs, which were sequentially illuminated to result in a chewing speed of 900 milliseconds/chewing cycle.48 Two 15-second trials were performed.

Prior to any of the recordings, participants performed a few trials of each task. All tasks were repeated in the same order during the three blocks (Fig 1). At the end of the recording session, the clutches were detached from the teeth, and the teeth were cleaned to remove any remnant of cyanoacrylate adhesive.

**Experimental Jaw Muscle Pain Induction.** After the completion of block 1 (see Fig 1), an intravenous (IV) catheter (JELCO, 22 G x 1 inch, Smiths Medical Australasia) was placed into the right anterior temporalis muscle to a depth of ~1 cm, ~1 cm behind the posterior border of the frontal process of the zygoma, and ~1 cm above the zygomatic arch. Another IV catheter was placed in the masseter muscle about midway between the superior and inferior palpated borders and the anterior and posterior palpated borders of the muscle. The IV catheter was inserted until bone was contacted and then retracted about 2 mm, or, if there was no bone contact, ~20 mm. The insertion site was swabbed with an alcohol wipe prior to each insertion.

A 1-mL syringe confirmed negative aspiration prior to attaching a polyethylene extension set (75-cm length, Medical Australia) to each IV catheter, and each extension set was connected to a 10-mL syringe (Becton Dickinson) in separate syringe pumps (IVAC Model P2000, Alaris Medical Systems). Experimental jaw muscle pain was induced in each muscle by the tonic infusion of 5% sterile hypertonic saline (Phebra Pty) into each muscle simultaneously so as to achieve a moderate pain intensity of 40 to 60 on a 100-mm visual analog scale (VAS) for the temporalis and on a separate masseter VAS. The VAS was a 100-mm straight line anchored at one end as “no pain at all,” and the other end with “the worst imaginable pain.” To control for volume effects, sterile isotonic saline (0.9%) was also infused either for block 2 or for block 3 (Fig 1). The sequence of hypertonic or isotonic saline infusion was alternated between participants who were blinded to the administration sequence. Hypertonic saline at ~5% is a frequently used algesic chemical that reliably evokes muscle pain with minor or no adverse effects reported over many years of use.49–52

When hypertonic saline was infused first into both jaw muscles, the infusion paradigm began with a bolus infusion of 0.2 to 0.4 mL of hypertonic saline over 20 seconds into the masseter muscle and 0.1 to 0.2 mL bolus infusion of hypertonic saline over 20 seconds into the temporalis muscle. These initial bolus injections into both the temporalis and masseter muscles ensured a rapid attainment of moderate pain intensity as scored on both pain scales from both muscles. This was followed by a continuous infusion at a rate of 3 to 6 mL/hour for the masseter infusion and 1 to 3 mL/hour for the temporalis infusion. To maintain a constant level of pain at both muscle sites, individual changes in the infusion rate were made in steps of 1 to 3 mL/hour on both pumps or by halting the infusions. These changes in infusion rates were made on the basis of the masseter VAS and the temporalis VAS scores, which were scored after the completion of each jaw task trial during both saline infusion sessions. One experimenter continually monitored these scores and adjusted the infusion rates for both muscles so as to maintain as closely as possible the VAS scores for both muscles in the range of 40–60/100 mm. After each trial, participants also outlined on lateral-profile outlines of the head and neck the extent of jaw muscle pain associated with each infusion. After the termination of each infusion, participants completed a McGill Pain Questionnaire (MPQ)53 to record verbal descriptors for the sensory, affective, evaluative, and miscellaneous dimensions of the pain experience. Two MPQs were used, one for each muscle. There were no side effects with the simultaneous infusion into the anterior temporalis and masseter muscles, and all participants scored 0 mm on both VAS scores within 5 to 7 minutes after termination of the hypertonic saline infusions.

**Data Analysis**

Pain referral sites were defined as outlines drawn on the pain maps that were distinct from the pain areas around the injection site. For each muscle, rank and weighted score calculations were made for each of the MPQ dimensions, and pain rating index (PRI) scores were summed.53 For each muscle, paired-samples t tests were used for comparisons between hypertonic saline and isotonic saline infusion blocks of the...
total volume of solutions infused, the mean VAS pain intensity scores (mean of all trials of a jaw task within each block), and the MPQ-weighted PRI scores and number of words chosen.

The outputs from both jaw tracking systems provide negative amplitude values for jaw opening in the z axis (inferior-superior axis), but only absolute values were used for display and analyses. The raw data files were converted using a customized program, and the trajectories of the mid-incisor point in the z axis were plotted as displacement time plots (GNU Plot, Manning). The standardized open/close tasks were analyzed with a customized computer program that calculated the mid-incisor point displacement from, and return to, the postural jaw position along the z axis. The onset of the opening phase in a standardized open/close jaw movement was the time when the mid-incisor point had displaced 0.5 mm along the z axis (inferior-superior axis) from the postural jaw position, and the end of the opening phase was when the mid-incisor point was within 0.5 mm of the holding phase of the open/close jaw movement task. The closing phase was taken from the end of the holding phase to within 0.5 mm of the postural position.

For analysis of the chewing cycles, all aberrant or incomplete cycles were identified visually and removed from further analysis. These included chewing cycles that were < 50% or > 150% of the mean amplitude for that individual. Then, for each chewing trial, the beginning of the opening phase was labeled at maximum closure along the z axis (inferior-superior axis) of the previous chewing cycle or when the mid-incisor point (between the incisal edges of the mandibular central incisors) had displaced downwards by 0.5 mm from the postural jaw position when analyzing the first chewing cycle. The end of the opening phase was the point of maximum opening displacement. The closing phase was the period between maximum opening (defined as the beginning of closing) to 0.5 mm from maximum closure (defined as the end of closing) of that chewing cycle. For every cycle of chewing in all trials, as well as every trial of standardized open/close movements, calculations were made of the displacement of the jaw along the z axis during each jaw opening and closing phase to provide values for amplitude of movement (mm). The amplitude was divided by the duration of each phase to provide velocity (mm/second) for each opening and closing phase. A repeated-measures analysis of variance (ANOVA) with adjusted pairwise comparisons was used to explore the effect of block (baseline, hypertonic saline infusion block, isotonic saline infusion block) on the mean amplitude and velocity values for the opening and closing phases of each task. The associations between amplitude and velocity variables during hypertonic saline infusion in relation to PCS scores and DASS-21 scores were explored with nonparametric correlation (Spearman rank correlation coefficient). Statistical significance for all tests was accepted at \( P < .05 \), and data were analyzed with SPSS for Windows (Version 21: SPSS).

Results

The total volume of hypertonic saline infused into the masseter muscle (mean: 1.10 mL [standard deviation (SD) 0.43 mL]) was significantly greater \( (P = .0006) \) than the volume infused into the anterior temporalis muscle (0.61 [0.46] mL). There was no significant difference between the total amount of hypertonic saline (0.61 [0.46] mL) and isotonic saline (0.48 [0.23] mL) infused into the right anterior temporalis \( (P = .14) \). The amount of hypertonic saline (1.10 [0.43] mL) infused into the right masseter muscle was significantly \( (P = .009) \) higher than the amount (0.86 [0.34] mL) of isotonic saline infused.

Perceived Pain Intensity

Hypertonic saline infusion into the right masseter and right anterior temporalis muscles was reported to be painful in all participants. The mean VAS scores for the hypertonic saline infusions into the right anterior temporalis (47.2 [8.1] mm) and right masseter (41.8 [7.9] mm) muscles were significantly higher \( (P < .0001) \) than the mean VAS scores during isotonic saline infusions into the same muscles (4.23 [0.3] mm and 1.5 [0.6] mm), respectively. Figure 2 shows the mean VAS scores for each of

![Fig 2 Mean visual analog scale (VAS, 0–100 mm) scores from the 15 participants after completion of each of the jaw tasks during the simultaneous infusion of hypertonic saline or isotonic saline into the right masseter and anterior temporalis muscles. Std chew = standardized chewing.](image)
the masseter and anterior temporalis muscles during each task for simultaneous hypertonic or isotonic saline infusions into the right masseter and anterior temporalis muscles. During the postural jaw position task only, the mean VAS pain intensity score across all participants during hypertonic saline infusion (or isotonic saline infusion) into the anterior temporalis muscle (hypertonic: 48.7 [14.45] mm; isotonic: 3.9 [4.7] mm) was significantly ($P < .05$) greater than during hypertonic saline infusion (or isotonic saline infusion) into the masseter muscle (hypertonic: 40.3 [12.2] mm; isotonic: 1.1 [1.9] mm); there were no significant differences for the other tasks. Eight participants were pain-free during isotonic saline infusion into the right masseter muscle, whereas five participants scored 0 for pain during the isotonic saline infusion into the anterior temporalis muscle.

**Perceived Pain Areas**
Simultaneous infusion of hypertonic saline into the right anterior temporalis and masseter muscles usually produced two discrete pain areas originating from the site of each infusion. In 7 of 15 participants, pain spread or referred to adjacent ipsilateral sites; eg, in or around the right ear (Fig 3a, P12), angle of the mandible (Fig 3a, P7), body of the mandible (Fig 3a, P7 and P15), temporomandibular joint and frontal region (Fig 3a, P7), lower lip (Fig 3a, P7 and P11), and near the top of the head or the posterior temporalis region (Fig 3a, P12 and P15). Pain maps from one representative participant (P10) demonstrate the qualitative observation that the distribution and spread of pain could vary between tasks (Fig 3b). Pain areas drawn after the postural position task were usually enclosed within the boundaries of the infused muscles and surrounded the injection sites; spread and referral tended to be noted later in the infusion (Fig 3b). During isotonic saline infusion, 10 participants drew a pain map over the anterior temporalis muscle and 6 drew a map over the masseter muscle. These maps were all small and did not cross the boundaries of the infused muscles.

**MPQ Descriptors**
For the description of pain caused by the hypertonic saline infusion into the anterior temporalis muscle, the most frequently chosen words were aching (9 participants, 60%), annoying (7, 47%), drilling (6, 40%), and piercing, hurting, and tight (5 for each, 33%). For the right masseter muscle, the most frequently chosen words were pressing and spreading (5 for each, 33%), dull (4, 27%), and hurting, sore, boring, and annoying (3 for each, 20%). During the isotonic saline infusion, the most common word descriptors for the anterior temporalis muscle pain were dull (5, 33%) and pricking (4, 27%). For the masseter, the most common descriptors were dull (4, 27%) and sore and tender (2 for each, 13%).

The mean weighted PRI scores for the sensory, affective, evaluative, and miscellaneous dimensions and the total score of the MPQ to describe hypertonic and isotonic saline pain are illustrated in Table 1. The PRI scores following hypertonic saline infusion into the anterior temporalis and masseter muscles were significantly higher ($P < .05$) than those during the isotonic saline infusion into these muscles. Each of the PRIs, except for the affective PRI, were significantly higher for the pain evoked by the hypertonic saline infusion into the anterior temporalis than for the masseter muscle ($P < .05$; Table 1). The sensory pain rating index (S-PRI) and the total PRI (PRI-Total) during isotonic infusion into the anterior temporalis muscle were significantly higher than the pain rating indices for the masseter muscle ($P < .05$; Table 1).
Jaw Movement Effects of Simultaneous Experimental Anterior Temporalis and Masseter Muscle Pain

The 15 participants were able to complete all of the required jaw movements under the different experimental conditions. Table 2 shows the standardized open/close jaw movements, free chewing and standardized chewing, and the mean (standard deviation) amplitudes and velocities for the opening and closing phases during the three blocks, as well as showing significant comparisons.

For the standardized open/close jaw movements, there were significant effects of block on the opening ($P = .027$) and closing ($P = .013$) amplitudes with significantly ($P = .042$) greater amplitude for the hypertonic block compared to baseline for the closing amplitude only. The opening ($P < .001$) but not closing velocity showed a significant effect of block with significantly lower baseline velocity than both the hypertonic ($P = .003$) and isotonic ($P = .009$) blocks (Table 2). There were no significant effects on amplitude or velocity for the comparison between isotonic saline and hypertonic saline.

During free chewing, a significant effect of block was found for both the opening ($P = .003$) and closing ($P = .035$) amplitudes and opening ($P = .007$) and closing ($P < .001$) velocities. Significant increases in the isotonic compared to the hypertonic blocks were found for the opening ($P = .045$) and closing ($P = .044$) amplitudes and for closing velocity ($P = .013$). A significant increase in velocity was found for the isotonic compared to the baseline blocks for the opening ($P = .008$) and closing ($P = .001$) velocities.

Analysis of the standardized chewing tasks found no significant effect of block on the opening or closing amplitudes; however, there was a significant effect of block for the opening ($P = .02$) and closing ($P = .015$) velocities. Comparison between the blocks found a significant ($P = .008$) increase in velocity during the isotonic compared to the baseline block. For the comparison between isotonic saline and hypertonic saline, there were no significant effects on amplitude or velocity.

Table 3 summarizes a qualitative analysis comparing absolute changes in amplitude and velocity between hypertonic saline infusion and isotonic infusion in each participant. For the purposes of this arbitrary analysis, a “+” indicates an increase in amplitude or velocity during hypertonic saline infusion in comparison with isotonic infusion, and a “−” indicates a decrease. Changes of ≤ 0.5 mm or ≤ 0.5 mm/second were arbitrarily classified as no change (“o”). Different participants could exhibit increases or decreases in amplitude and velocity within a task, and these changes were not consistent between tasks.
no significant reductions of the magnitude of kinematic parameters of standardized open/close jaw movements or standardized chewing. Individual participants could show increases, decreases, or no change in amplitude and velocity during hypertonic saline infusion in comparison with isotonic saline infusion (Table 3). These findings provide some support for the first hypothesis, that simultaneous noxious stimulation of the masseter and anterior temporalis muscles results in reductions in the amplitude and velocity of jaw movements during standardized open/close jaw movements and during free and standardized chewing. Significant correlations were noted between PCS scores and some amplitude and/or velocity values during open/close jaw movements or standardized chewing in isotonic or baseline blocks only; no significant correlations were noted during simultaneous noxious stimulation. These findings do not support the second hypothesis, that simultaneous noxious stimulation of the masseter and anterior temporalis muscles results in changes in amplitude and velocity of jaw movements that correlate with scores of mood or pain-related cognitions. The accompanying paper (Amhamed et al[23]) has provided evidence from the same participants in the present paper that, in comparison with control, the noxious stimulation results in changes in superficial jaw muscle EMG activity that can vary with the task, the muscle, the participant, and some psychologic variables. Further, and as pointed out

Correlations Between Psychologic Variables and Kinematic Variables

The score for the PCS taken before the experiment (12.7 [9.8]) was not significantly ($P = .96$) different from the PCS obtained after (12.6 [9.6]); therefore, the scores were averaged to obtain a mean PCS score for each participant. The scores for the DASS-21 indicated low levels for depression (1.4 [1.8]), anxiety (1.2 [1.3]), and stress (1.9 [1.7]). There was a significant positive correlation between PCS scores and closing amplitude ($r = 0.490$, $P = .032$) during the isotonic saline infusion for the standardized open/close task. During standardized chewing in the baseline block, there was a significant positive correlation between PCS scores and closing amplitude ($r = 0.586$, $P = .011$) and opening velocity ($r = 0.502$, $P = .028$), and a negative correlation with the opening amplitude ($r = -0.581$, $P = .012$). There were no other significant correlations.

Discussion

Simultaneous noxious stimulation of the right anterior temporalis and masseter muscles (by hypertonic saline infusion) resulted in significantly smaller opening and closing amplitudes and a significantly lower velocity of closing for free chewing in comparison with simultaneous control infusion of isotonic saline into the right anterior temporalis and masseter muscles. However, there were

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$+$ = increase in amplitude or velocity during hypertonic saline infusion in comparison to isotonic infusion; $–$ = decrease; $o$ = no change (changes of $\leq 0.5$ mm or $\leq 0.5$ mm/second were arbitrarily classified as $o$).

Table 3 Changes in Amplitude and Velocity for Each Jaw Movement Task Under Isotonic vs Hypertonic Saline Infusion for Each Participant
in the accompanying paper, some significant EMG changes were noted even when there were no significant changes in the associated jaw movement kinematic features.

Previous human studies in experimental masseter noxious stimulation alone\(^2\) and experimental temporals noxious stimulation alone\(^2\) resulting in moderate pain intensity at each jaw muscle site found no significant effects on free or standardized chewing amplitude or velocity, although simulated chewing amplitude has been shown to be significantly but only slightly reduced during experimental masseter pain.\(^\text{20}\) In an experimental animal model, bite force was significantly reduced with algesic chemical injection into the masseter in comparison with control.\(^\text{14}\) In light of these earlier findings, the significantly smaller amplitude and velocity during free chewing during moderate pain intensity at both masseter and temporal sites suggest that the spatial features of the noxious stimulus may be a factor in modulating the effects of pain on motor activity in a group of young healthy individuals. While the smaller amplitude and velocity during noxious stimulation in the present study is indeed consistent with the reductions in movement amplitude with pain as proposed by the Pain Adaptation Model, the possible requirement for spatially distributed noxious stimulation at two jaw muscle sites—which is suggested by the present findings to produce this effect—does not appear to be covered by this earlier model. The present findings in relation to the previous studies\(^\text{18,23}\) might be better interpreted in terms of more recent models of pain-motor interaction.\(^2\) One of these more recent models proposes that the multidimensional nature of pain influences the motor response to pain. Therefore, in the context of at least one of these newer theories,\(^2\) the effects of pain on the motor system may vary with the spatial distribution of the noxious stimulus as part of the sensory-discriminative aspect of the pain experience.

There is evidence for significant reductions in the excitability of the face region of the primary motor cortex with noxious stimulation of the masseter muscle in humans\(^5\) and with noxious stimulation of the tongue in anesthetized rats,\(^3\) and these observations are consistent with the inhibitory effects observed on limb primary motor cortex excitability with limb noxious stimulation.\(^5,5,6\) Simultaneous masseter and anterior temporalis noxious stimulation is likely to result in reductions in brain excitability covering a larger area within the face motor cortex than would occur with noxious stimulation of the individual muscles alone. Given the proposed role of the primary motor cortex in the fine control of jaw movements during chewing,\(^30,57\) the simultaneous noxious stimulation at two muscle sites may therefore have a more profound effect on the fine control of jaw movements during chewing, and this may contribute to the reduction in amplitude and velocity of free chewing as demonstrated in the present study. Remarkably, however, these possible more profound effects on the face motor cortex did not appear to be sufficient to have an effect on amplitude and velocity during goal-directed standardized open/close jaw movements and standardized chewing tasks. There appears to be sufficient redundancy in the motor system to allow performance of these tasks at control levels, at least at the level of assessment employed in the present study.

**Methodologic Limitations**

The sample size was small and the chances of a type I error were increased by the number of analyses performed. The findings must therefore be regarded as exploratory, with further studies being required for confirmation—or not—of the main findings. Another issue is that the sample was mostly health professional staff and postgraduate students, who will have more knowledge than the general population about the orofacial motor system. In addition, the sample was almost exclusively men, even though TMD is more common in women. Therefore, the findings cannot be readily translated to the general population, nor to chronic pain patients in whom complex neuroplastic changes and adaptations together with psychologic distress are likely to have a significant impact. Future studies can therefore be directed toward studies in patients with chronic pain.

**Pain Intensity, Pain Maps, and McGill Pain Questionnaire Findings**

The VAS pain intensity scores for the anterior temporalis muscle were significantly ($P < .05$) higher than for the masseter muscle during the postural position task only, with no significant differences for the other tasks despite significantly more total volume of solution being infused into the masseter than the anterior temporalis muscle. The temporalis MPQ pain rating indices were also mostly significantly higher than from the masseter muscle. Previous studies have reported that noxious stimulation of the anterior temporalis muscle can result in higher, lower, or no change in pain scores when compared with noxious stimulation of the masseter muscle,\(^18,23,58–60\) although in a well-controlled study,\(^5\) glutamate injections into the temporalis muscle generated significantly higher pain intensity than identical glutamate injections into the masseter in the same individuals. Another factor to consider is the possible role of conditioned pain modulation (CPM), which has been demonstrated to operate in asymptomatic individuals.\(^61,62\) It is possible that the temporalis nociceptive input is more effective...
than the masseter nociceptive input in recruiting descending inhibitory systems to inhibit the nociceptive input from the masseter muscle. The higher pain intensity and pain rating indices for the temporals than the masseter muscle may also reflect differences in anatomy between the sites, for example, a lower tissue distensibility at the anterior temporalis injection site in comparison to the masseter injection site. More nociceptors may be activated because of a possibly greater intramuscular pressure at the anterior temporalis injection site than at the masseter muscle injection site.

The pain maps represented a simple combination of maps that might be expected with single muscle injections, or in some participants the maps that did not necessarily include the injection sites (eg, Fig 3). As has also been noted during single injections, spread and referral of pain could occur, The PRI-S from the MPQ for each muscle was significantly higher than the other indices, and this is similar to findings from previous studies that have injected algogenic chemicals into the jaw muscles or have applied noxious stimuli to the orofacial area, as well as in TMD patients. Pain evoked from injections into the anterior temporalis muscle was most commonly described by participants as aching, annoying, and drilling, whereas pain evoked from the masseter muscle was commonly described as pressing and spreading. Some of these words have also been previously reported to be chosen in both experimental and clinical pain.

Motor Consequences of Experimentally Induced Jaw Muscle Pain

The baseline opening jaw movement amplitudes at the mandibular mid-incisor point during free and standardized chewing (15.1 [2.5] mm; 15.3 [3.8] mm) are comparable to previous pain-free chewing of gum (14.9 [4.0] mm; 15.3 [3.5] mm) and to chewing gum or peanuts (13.7 [2.4] mm), although higher than other previous studies (10.9 [2.5] mm and 12.1 [3.9] mm). The mean opening velocity in pain-free trials of free and standardized chewing (35.3 [9.6] mm/second; 37.1 [9.1] mm/second) are comparable to (37.7 [10.6] mm/second; 37.2 [9.1] mm/second) or higher than other previous studies (25.7 [9.2] mm/secondand31.4 [11.5] mm/second; 26.7 [8.8] mm/second and 24.9 [9.4] mm/second).

The following refers principally to the comparison between the block during which there was simultaneous noxious stimulation of the masseter and anterior temporalis muscles with hypertonic saline and the block during which isotonic saline was infused. The isotonic saline controls for possible effects related to volumetric changes in the infused muscles. During the simultaneous noxious stimulation, significant decreases were noted for opening and closing amplitude and for closing velocity in free chewing in comparison with isotonic infusion; there were no significant changes noted for the standardized chewing or open/close jaw movement tasks.

Amplitude and velocity of free chewing were not significantly different between the baseline trials and noxious stimulation trials. However, the baseline trials were performed first and allowed participants to become accustomed to the tasks. There is also recent evidence that repetitions of free chewing movements in a paradigm similar to that carried out in the present study are associated with progressive increases in velocity and amplitude of jaw movement and are attributed to practice effects and motivational factors. The alternation between successive participants of the sequence isotonic infusion first or hypertonic infusion first helped to distinguish an effect related to the hypertonic saline as distinct from an effect related to other effects, such as practice or motivational effects, or to volumetric changes associated with the infusion. Thus, if only practice or motivational effects were present (and no pain effects), then there should be no significant differences in the kinematic parameters between the noxious stimulation and the control isotonic saline infusion.

The findings (Table 2) demonstrate that simultaneous noxious stimulation of both the anterior temporalis and the masseter muscles results in a reduction in kinematic parameters during free chewing. These findings are consistent with the predictions of the Pain Adaptation Model, which proposes that pain results in a decrease in jaw movement kinematic parameters. They are also consistent with the findings of previous human and animal studies in the trigeminal and spinal systems and which have reported that experimental or clinical pain results in reductions in the amplitude and/or velocity of movements or reductions in maximum voluntary contraction force or bite force, For example, in both human and animal experimental pain models, bite force was significantly reduced with algogenic chemical injections into the masseter in comparison with control. The findings are however not consistent with the findings from the only other study in the trigeminal motor system of simultaneous algogenic chemical injections into two jaw muscles, namely bilateral masseter muscle injections. In this study, while effects on agonist and antagonist muscle activity were noted, there was no significant effect on maximum amplitude of chewing or the duration of the opening or closing phases of chewing.

The absence of significant reductions in velocity and amplitude during the standardized open/close task and standardized chewing is not consistent with the Pain Adaptation Model and is remarkable given the moderate pain induced in two jaw muscles.
A previous study did report a reduction in the amplitude of open/close jaw movements during experimental masseter noxious stimulation18; however, other studies reported that experimental jaw muscle or intraoral mucosal pain did not result in significant reductions in kinematic parameters of some jaw movements18,23,76,79 or in clinical TMD pain.40,80

The presence of mild reductions or no effects on kinematic parameters with simultaneous masseter and temporalis muscle noxious stimulation deserves comment. First, the participants had low distress scores and were highly motivated health care professional staff and students who were expecting a brief period of experimental pain. Effects may be different in individuals with higher distress scores experiencing unexpected acute muscle pain. Second, qualitatively (Table 3) and consistent with the observations from previous studies,18,23 some participants exhibited increases in kinematic parameters, while others experienced decreases or no change during noxious stimulation. Grouping of the data sets in the statistical analysis may therefore obscure these individual effects. Third, the pain intensity in the present study (temporalis: 47.2 [8.1] mm; masseter: 41.8 [7.9] mm) may have been too low, as significant reductions in kinematic parameters have been noted in chewing64 with the high VAS pain intensity scores used in this earlier study (70 [10] mm). Nonetheless, the pain intensity scores employed in the present study were higher than the pain ratings reported by TMD patients (37 [22] mm),66 but were lower than the pain intensity scores (64 [21]) in a normative dataset from a sample of patients with chronic noncancer pain presenting to a tertiary referral multidisciplinary pain management center (1994 to 2004).37 The pain intensity levels employed in the present study could certainly be classified as sufficient in terms of pain intensity levels that can be experienced by TMD patients.

**Association Between Psychologic Variables and Jaw Motor Behavior During Experimental Pain**

There is an emerging body of evidence that identifies relationships between the emotional and motor systems.42,43,76 In humans, the presence of lower pain thresholds in individuals with high anxiety and depression (affected with major depression)44,45,46 and with chronic pain47 compared to healthy controls has been noted. In clinical TMD pain, the presence of only a few significant correlations between the kinematic measures and the psychologic variables likely reflects these low scores from a group of psychologically well-functioning participants.

**Conclusions**

The present findings show that, in comparison with control, simultaneous noxious stimulation of both the anterior temporalis and masseter muscles results in no significant changes in jaw movement during standardized open/close jaw movements and standardized chewing, and only mild reductions in amplitude or velocity of jaw movement during free chewing. The findings for free chewing support the Pain Adaptation Model, but the findings for the standardized jaw tasks do not. There were no significant associations between the DASS or PCS scores and amplitude or velocity during hypertonic saline infusion, and this lack of significant association may reflect the low scores in this small study sample.

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**References**

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