Referred Pain and Sensations Evoked by Standardized Palpation of the Masseter Muscle in Healthy Participants

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Aims: To determine if standardized palpation of the masseter muscle can evoke referred pain and/or sensations in healthy individuals and to compare the mechanical sensitivities in response to three different levels of palpation force. **Methods:** A total of 32 pain-free individuals participated. The right masseter muscle was divided into 15 test sites. Mechanical sensitivity of the masseter was assessed with three mechanical stimuli (0.5 kg, 1.0 kg, or 2.0 kg) applied by palpometers to the 15 test sites for 5 seconds each site. Participants scored the perceived intensity of pain and unpleasantness of each of the three mechanical stimuli on 0–100 numeric rating scales (NRS). After each stimulus, the duration of aftersensation was measured, and the participants were also asked to indicate areas within the orofacial region with referred pain/sensations. Data were tested using analysis of variance, Tukey post hoc, and McNemar's tests with a 5% level of significance. **Results:** Referred pain/sensations were most commonly evoked with the 2.0-kg stimulus (34.4% of participants; $P < .05$) compared to the 1.0-kg (12.5%) and 0.5-kg stimuli (3.1%). There were significant effects of stimulus intensity on NRS scores for pain and unpleasantness, as well as for aftersensation ($P < .05$). There were significant effects on NRS scores for pain and unpleasantness for the 1.0- and 2.0-kg stimuli ($P < .05$) and on aftersensation for the 2.0-kg stimulus ($P < .05$). **Conclusion:** These results indicate that referred pain/sensations in the orofacial region are frequent phenomena among healthy individuals during standardized palpation of the masseter muscle. *J Oral Facial Pain Headache 2018;32:159–166. doi: 10.11607/ofph.2019*

Keywords: *aftersensation, masseter muscle, mechanical sensitivity, palpation, referred pain*

Temporomandibular disorders (TMD) involving myofascial pain in the jaw muscles are prevalent in the general population, ranging from 21.5% to 51.8%,[1–4] and are twice as common in women as in men.[5,6] Common symptoms of myofascial pain conditions in the jaws are fatigue, soreness, pain in the jaw muscles, and restricted jaw function.[7,8] Muscle pain is typically described as diffuse and difficult to localize and is often referred to regions remote from the muscle regions.[9] Pain localized to the source of pain is termed local pain, whereas pain felt in a region or structure away from the source of pain is termed referred pain.[10] If the source of the pain is not identified, the clinician may make an incorrect diagnosis and provide inappropriate treatment. Although several theories of referred pain have been proposed to explain this phenomenon,[11–13] the precise neural pathways of referred pain from the masticatory muscles are unclear. Despite this, the extensive convergence of afferent inputs from various tissues onto nociceptive neurons in the trigeminal brainstem sensory nuclear complex is believed to be crucially involved.[14,15]

The Diagnostic Criteria for Temporomandibular Disorders (DC/TMD) provide a comprehensive assessment of the most common TMD based on the biopsychosocial model of chronic pain.[16] According to the DC/TMD procedure, the examiner palpates the masseter muscle by increasing the stimulus intensity to 1.0 kg and holding the pressure for a specified time. During palpation, a duration of either 2 seconds (for diagnosis of myalgia) or 5 seconds (for diagnosis of referred pain) is recommended. However, clear evidence is lacking regarding the optimal stimulus intensity and duration of palpation for examining referred pain in the orofacial area. To clarify the relationship between mechanical sensitivity and referred pain in the orofacial area, the identification of duration- or intensity-dependent relationships with local pain in the masticatory muscles is essential.

While manual palpation is the most widely used clinical method to diagnose TMD and other musculoskeletal pain conditions, including fibromyalgia,[16,17] many factors can influence the outcomes of manual palpation (eg, patient bias, examiner experience, instructions, training, and psychological state).[18] These factors need to be considered, and if the physical stimulus (ie, pressure) is not well controlled, the results will not be reliable. The reliability of manual palpation applied to the masticatory muscles is normally considered adequate but not optimal.[19–21] In efforts to improve the reliability of manual palpation, several devices have been proposed for assessing mechanical sensitivity.[22–25]

It has been proposed that the distribution of pain should be considered to enrich the characterization of different diagnoses of muscle pain.[16,26–28] Pain perception in the masseter muscle would also be expected to vary depending on the part of the masseter muscle that is affected. Systematic assessment of the spatial distribution of mechanical sensitivity in the masseter muscle may contribute new and crucial information to the characteristics of orofacial muscle pain. Since entropy measures complexity and the degree of diversity of information,[29–31] it could be used to assess localized muscle mechanical sensitivity in response to standardized palpation with a palpometer and may be useful for establishing optimal stimulus intensity of muscle palpation to cause referred pain for diagnosing myofascial pain in the masseter muscle. Also, it may be helpful for better comprehension of the mechanical sensitivity and referred pain mechanisms in the masseter muscle. Therefore, the aims of this study were to determine if standardized palpation of the masseter muscle can evoke referred pain and/or sensations in healthy individuals and to compare the mechanical sensitivities in response to three different levels of palpation force.

Materials and Methods

Participants

A total of 32 healthy volunteers (16 men, mean ± standard deviation [SD] age 32.4 ± 12.7 years; 16 women, mean age 25.4 ± 6.1 years) were recruited from the Section of Orofacial Pain and Jaw Function, Department of Dentistry and Oral Health, Health, Aarhus University. Inclusion criteria were: no ongoing pain in the face or any other reported chronic pain in the last 6 months; no medical history of systemic disease; no current pregnancy (participant-based report); no medications (eg, nonsteroidal anti-inflammatory drugs, muscle relaxants, anxiolytics, or hypnotics); and no orofacial pain or temporomandibular pain symptoms assessed with the DC/TMD.[16] The study protocol followed the guidelines of the World Medical Association Declaration of Helsinki II. All participants signed an informed consent document agreeing to participate in the study after being provided written and oral information about the experiment. This protocol was approved by the Central Denmark Region Research Ethics Committee (1-10-72-286-14).

Study Design

The study was performed as a single-blinded, randomized study. The anterior-posterior and superior-inferior borders of the right masseter muscle were identified by palpation during repetitive clenching, and the area was divided into 15 test sites (5 vertical and 3 horizontal) for the right masseter muscle (Fig 1a). Mechanical sensitivity was assessed by using a palpometer (Palpeter; Sunstar Suisse SA) applied to each of the 15 test sites with three different stimulus intensities (0.5 kg, 1.0 kg, 2.0 kg).[23,25] The duration of a single stimulus at each test site was 5 seconds, in accordance with the DC/TMD.[16] The order of stimulus intensity and site tested was randomized using a randomization program available on the internet (www.randomization.com).

Each stimulus was repeated three times per site for all participants. After each stimulus, participants were asked to score perceived intensity of pain and unpleasantness on numeric rating scales (NRS) as an indicator of mechanical sensitivity. Participants were carefully instructed in the use of the NRS for pain intensity, with 0 denoting no sensation at all, 50 denoting a just barely painful sensation, and 100 denoting the most painful sensation imaginable (Fig 1b).[32] Mean pain NRS scores were assessed for each of the 15 test sites as an overall assessment of mechanical sensitivity. On a different 0-to-100 NRS, the participants scored the intensity of unpleasantness, with 0 denoting no unpleasantness at all and 100 the most unpleasant sensation imaginable (Fig 1c). In addition, to measure aftersensations, participants were asked to lift a hand when they did not feel any sensation in their masseter muscle after each removal of the stimulus, and the examiner counted the time it took until they lifted their hand. A stopwatch was used to record aftersensations as the duration (in seconds) of the sensation perceived after removal of the stimulus.[33]

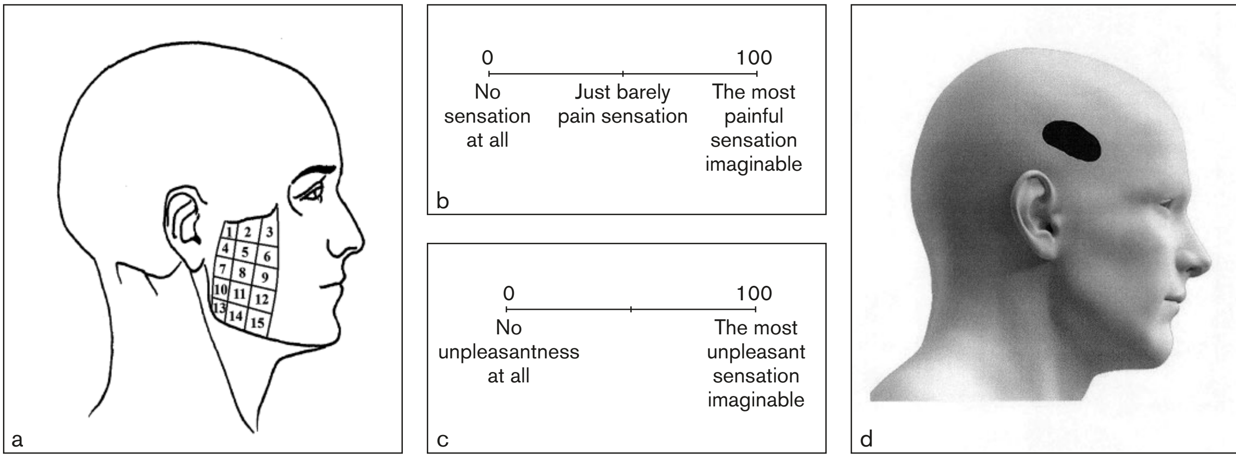

Fig 1 **(a)** The anterior-posterior and inferior-superior borders of the masseter muscle were identified, and the areas were divided into 15 test sites (5 vertical and 3 horizontal). **(b, c)** Perceived intensity of pain and unpleasantness was scored on 0-to-100 numeric rating scales. **(d)** The participants were asked to indicate the area of referred pain/sensation on a digital anatomical drawing.

Pain/sensations were considered as referred pain/sensations if the participant reported pain or any other sensation beyond the boundary of the masseter muscle being palpated (ie, perceived in another structure). Pain/sensations were not considered referred if the participant reported pain or sensation extending beyond the area of provocation while remaining within the boundary of the masseter muscle. If the participants reported referred pain/sensations, they were asked to indicate the area on a digital anatomical drawing (Navigate Pain; Aglance Solutions) after each stimulus (Fig 1d).

In the context of the diversity of mechanical sensitivity scores for the right masseter muscle, entropy indicates the degree of diversity of the NRS sensitivity scores, with higher entropy values corresponding to more diverse intensity registers of NRS scores over the grid. Entropy was calculated for NRS scores for both pain and unpleasantness of the 15 test sites for each intensity within the right masseter muscle following a previously described method.[31]

Additional Experiment: Force Values

To compare the test-retest variability of palpation in this study, the same examiner carried out an additional experiment to measure actual force values in 10 consecutive applications of forces at the times 0 seconds and 5 seconds with each stimulus intensity (0.5 kg, 1.0 kg, and 2.0 kg) and without any visual feedback.[25] Time 0 seconds was defined as the time at which the examiner felt stability of pressure for each target stimulus intensity. Actual force values were measured by a force transducer (EJ-3000; A&D). Coefficients of variation (CVs) were calculated from actual force values at 0 seconds and 5 seconds with each stimulus intensity (0.5 kg, 1.0 kg, and 2.0 kg). CV was defined as the SD divided by the mean of 10 repeated force measurements and multiplied by 100 (to report the answer as a percentage).

Statistical Analyses

Analysis of variance (ANOVA) was used to test differences in mean pain and unpleasantness NRS scores and in the duration of aftersensations for the three mechanical stimulus intensities with the following factors: gender (2 levels), stimulus intensity (3 levels), and test site (15 levels). Before ANOVA, assumption of normality was tested using the Shapiro-Wilk test, and homogeneity of variance was tested using Levene's test. Tukey post hoc test was used with correction for multiple comparisons. Entropy scores for palpation were analyzed with one-way ANOVA with stimulus intensity as the factor (3 levels). Furthermore, McNemar's test was used to test for differences in the number of participants who reported referred pain/sensations for each of the three mechanical stimulus intensities and for each of the 15 test sites. In the additional experiment, CVs of actual force with each stimulus intensity were analyzed with one-way ANOVA for duration (0 seconds and 5 seconds). For all tests, the significance level was set at $P < .05$. All data are presented as mean values and SDs.

Results

NRS Scores

Mean NRS scores for pain were 14.6 ± 8.8 for the 0.5-kg stimulus and 30.4 ± 14.9 for the 1.0-kg stimulus (nonpainful range), whereas the score for the 2.0-kg stimulus was 55.4 ± 16.5 for the 2.0-kg (painful range). The lowest NRS scores for pain for each

Table 1 Statistical Relationship of Factors for NRS Scores and Duration of Aftersensations

	Gender	Stimulus	Test sites	Gender × stimulus	Gender × test site	Stimulus × test site	Gender × stimulus × test site
Nonpain/pain NRS	.917	< .001	< .001	.945	.367	< .001	.872
Unpleasantness NRS	.498	< .001	< .001	.625	.821	< .001	.122
Duration of aftersensation (s)	.546	< .001	< .001	.165	.682	.002	.596

The *P* values are shown from ANOVAs testing differences in means of pain and unpleasantness NRS scores and duration of aftersensations for three mechanical stimulus intensities with the following factors: gender (2 levels), stimulus intensity (3 levels), and test site (15 levels).

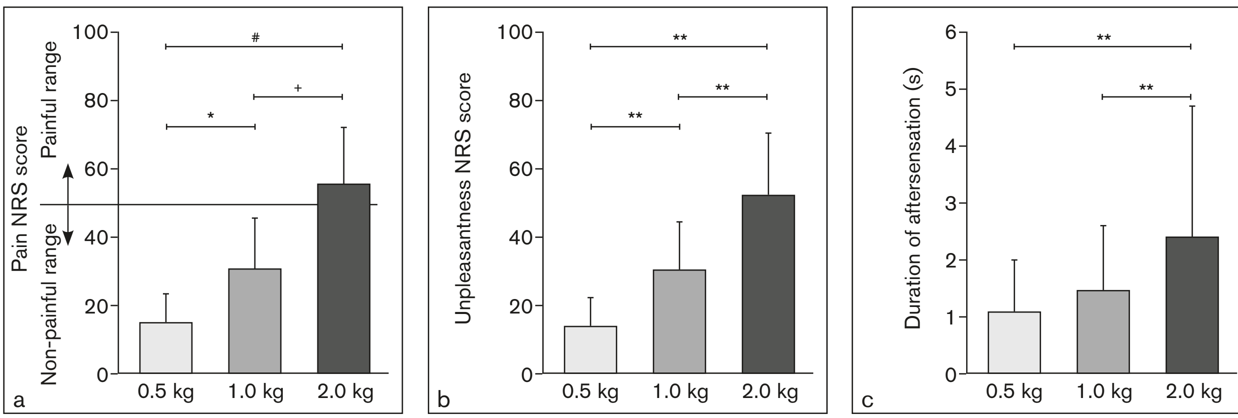

Fig 2 Comparison of **(a)** pain NRS scores, **(b)** unpleasantness NRS scores, and **(c)** duration of aftersensation between stimulus intensities. Pain and unpleasantness NRS scores with 2.0 kg were significantly higher than with 0.5 kg and 1.0 kg and were significantly higher with 1.0 kg than with 0.5 kg (***P* < .001 for both, Tukey post hoc test). The duration of aftersensation with 2.0 kg was significantly longer than with 0.5 kg and 1.0 kg (***P* < .001, Tukey post hoc test).

Table 2 Areas of Referred Pain/Sensations With Each Stimulus Intensity

Stimulus	Referred area	Participants with referred pain, n/total (%)
0.5 kg	Temporal	1/32 (3.1)
1.0 kg	Mandibular teeth	2/32 (6.3)
	Temporal	1/32 (3.1)
	Maxillary teeth	1/32 (3.1)
	Orbital	1/32 (3.1)
2.0 kg	Temporal	7/32 (21.9)
	Orbital	2/32 (6.3)
	Frontal	1/32 (3.1)
	Mandible	1/32 (3.1)
	Lip	1/32 (3.1)
	Maxillary teeth	1/32 (3.1)
	Mandibular teeth	1/32 (3.1)

stimulus intensity were 13.1 ± 7.6 at site 4 with the 0.5-kg stimulus, 26.0 ± 13.1 at site 1 with the 1.0-kg stimlus, and 50.0 ± 15.4 at site 15 with the 2.0-kg stimulus. The highest pain NRS scores for each stimulus intensity were 17.1 ± 12.0 at site 6 with the 0.5-kg stimulus, 34.9 ± 15.3 at site 9 with the 1.0-kg stimulus, and 61.7 ± 15.9 at site 9 with the 2.0-kg stimulus.

Mean NRS scores for unpleasantness were 13.7 ± 8.5 for the 0.5-kg stimulus, 29.8 ± 14.9 for the 1.0-kg stimulus, and 52.1 ± 18.4 for the 2.0-kg stimulus. No significant differences were seen in overall pain and unpleasantness NRS scores for palpation between genders (*P* > .05; Table 1), but significant differences were seen in overall pain and unpleasantness NRS scores between stimulus intensities and test sites (*P* < .001) (Table 1). Pain and unpleasantness NRS scores obtained with the 2.0-kg stimulus intensity were significantly higher than those with the 0.5- and 1.0-kg stimulus intensities. Furthermore, pain and unpleasantness NRS scores with the 1.0-kg stimulus were significantly higher than those with the 0.5-kg stimulus (Figs 2a and 2b).

Referred Pain/Sensations

Referred pain/sensations were evoked in 3.1% of healthy participants (n = 1/32) with the 0.5-kg stimulus intensity, in 12.5% with the 1.0-kg stimulus intensity, and in 34.4% with the 2.0-kg stimulus intensity. The only area of referred pain/sensations elicited by the 0.5-kg stimulus intensity was in the temporal region (3.1%). The areas of referred pain/sensations elicited by the 1.0-kg stimulus intensity were the mandibular teeth (6.3%), maxillary teeth (3.1%), temporal region (3.1%), and orbital region (3.1%). The areas of referred pain/sensations elicited by the 2.0-kg stimulus intensity were the temporal region (21.9%), orbital region (6.3%), frontal region (3.1%), lip region (3.1%), maxillary teeth (3.1%), mandibular teeth (3.1%), and mandibular region (3.1%) (Table 2).

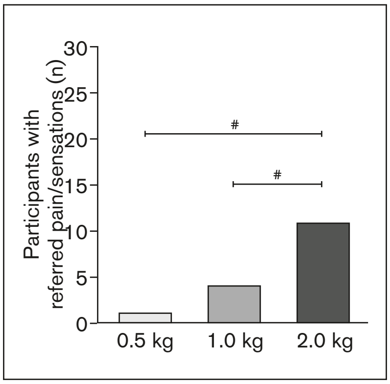

Fig 3 Comparison of number of participants with referred pain/sensations between stimulus intensities. The number of participants with referred pain/sensations with 2.0 kg was significantly higher than with 0.5 kg and 1.0 kg (*P < .05, McNemar's test).

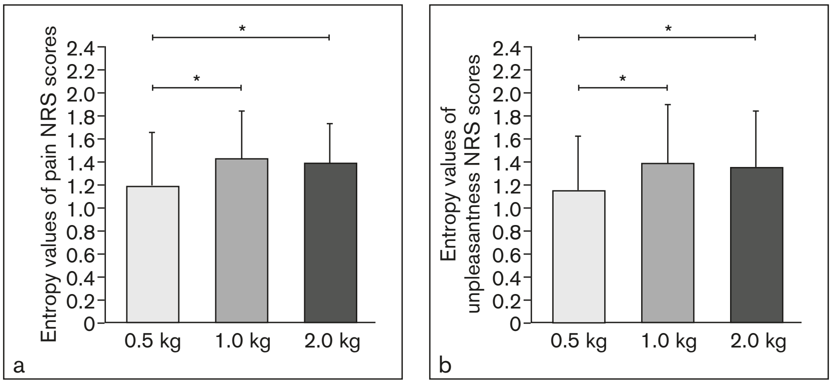

Fig 4 Comparison of entropy values of **(a)** pain and **(b)** unpleasantness NRS scores between stimulus intensities. Entropy values of both scores were significantly higher with 2.0 kg than with 0.5 kg and 1.0 kg (*P < .05, Tukey post hoc test).

McNemar's test assessing the number of participants with referred pain/sensations evoked by each test site showed no significant differences between genders or test sites. However, the number of participants with referred pain/sensations elicited by the 2.0-kg stimuli was significantly higher than those with referred pain/sensations elicited by the 0.5- and 1.0-kg stimuli (P < .05; Fig 3).

Duration of Aftersensation

No significant differences were seen in duration of aftersensation for palpation of the 15 test sites between genders (P > .05); however, significant differences were seen in the duration of aftersensation between stimulus intensities and test sites (P < .001). Duration of aftersensation also showed a significant interaction for intensity and test site (P < .001; Table 1). The duration of aftersensation with the 2.0-kg stimulus was significantly longer than with the 0.5-kg or 1.0-kg stimuli (P < .001; Fig 2c).

Entropy Analysis of Mechanical Sensitivity

ANOVA analyses of entropy values for pain and unpleasantness NRS scores showed overall statistically significant differences between stimuli (P < .01). Post hoc tests showed that entropy values of pain and unpleasantness NRS scores elicited by 1.0- and 2.0-kg stimuli were significantly higher than those elicited by the 0.5-kg stimulus (P < .05) (Fig 4). However, no significant differences in entropy values of pain or unpleasantness NRS scores were seen between the 1.0- and 2.0-kg stimuli (P > .05).

Additional Experiment: Force Values

Mean actual force at 0 seconds was 0.52 ± 0.02 kg with the 0.5-kg stimulus, 1.02 ± 0.05 kg with the 1.0-kg stimulus, and 2.17 ± 0.04 kg with the 2.0-kg stimulus. Mean force at 5 seconds was 0.51 ± 0.02 kg with the 0.5-kg stimulus, 1.01 ± 0.04 kg with the 1.0-kg stimulus, and 2.00 ± 0.05 kg with the 2.0-kg stimulus. No significant difference in actual force was seen between time 0 seconds and 5 seconds (P > .05). CVs at 0 seconds were 3.1%, 4.9%, and 3.5% with 0.5-, 1.0-, and 2.0-kg stimuli, respectively. CVs at 5 seconds were 4.4%, 4.0%, and 2.7% with 0.5-, 1.0-, and 2.0-kg stimuli, respectively. No significant differences in CVs were seen between 0 seconds and 5 seconds with either stimulus intensity (P > .05).

Discussion

The main findings in this study were that: *(1)* referred pain/sensations occurred with 0.5-, 1.0-, and 2.0-kg stimulus intensities in healthy participants; *(2)* a positive relationship existed between the number of participants reporting referred pain/sensations and stimulus intensity; *(3)* although mechanical sensitivity was dependent on test site, referred pain/sensations were not; 4) a positive relationship existed between entropy values and stimulus intensity; and *(5)* applying a 2.0-kg stimulus to the masseter muscle was likely to evoke pain in healthy participants.

The mechanism for referred pain is believed to represent a combination of central sensitization, convergence of sensory nerve fibers from multiple sites, changes in second-order neuron connectivity, and descending facilitation within the central nervous system.[11–14,34] Some studies have compared patients to healthy individuals for referred pain provoked by palpation in other regions of the body (eg, lower part of the body or low back). Torstensson et al[35] compared referred pain provoked by palpation for 13 intra-pelvic landmarks between participants with and without chronic pelvic pain (CPP); 9% of participants without CPP reported referred pain provoked by palpation. Chang-Yu et al[36] compared referred pain provoked by palpation of low back muscles between participants with and without low back pain; 1% of participants without low back pain experienced referred pain provoked by palpation. The present results also showed referred pain/sensations in the orofacial region among healthy participants upon standardized palpation of the masseter muscle. The results suggest that even participants who do not have pain or symptoms may be subject to the mechanisms of referred pain in the masseter muscles. However, the present study used only 5 seconds as the duration of the mechanical stimulus. Further studies are needed to clarify the influence that the duration of a mechanical stimulus applied to the masseter muscle may have in eliciting referred pain in the orofacial region. In addition, since the present study only applied palpation to the masseter muscle, further studies are needed to investigate the mechanical sensitivity and referred pain/sensations when mechanical stimuli are applied to other masticatory muscles (eg, temporalis muscle). Furthermore, referred pain and/or sensations were studied as one entity in the present study. Further studies are needed to clarify the proportion of healthy participants with referred pain and/or sensations.

Some studies have reported a positive correlation between pain intensity and frequency of referred pain.[37,38] The present results also showed a positive correlation between stimulus intensity and the number of participants with referred pain/sensations and also suggest that referred pain from the masseter muscle is an intensity-dependent process originating from a local stimulus. However, although the intensities of mechanical sensitivity applied were 0.5, 1.0, and 2.0 kg, the duration of mechanical sensitivity was fixed at 5 seconds. Interestingly, referred pain/sensations were not dependent on test site, whereas NRS scores of perceived pain and unpleasantness were. Further studies are needed to investigate differences in mechanical sensitivity to palpation between test sites in the masseter muscle.

Castrillon et al[31] reported that a 2.0-kg stimulus intensity applied to the masseter muscle would be sufficient to elicit a mechanical pressure pain sensation. The present results showed that mean pain NRS scores were in the nonpainful range for 0.5- and 1.0-kg stimuli and in the painful range for the 2.0-kg stimulus, supporting previous findings. These results also suggest that the 2.0-kg stimulus intensity is not suitable for clinical palpation of masseter muscles. In addition, Rainville et al[39] demonstrated that both pain intensity and unpleasantness are tightly linked to stimulus intensity across different stimulus types in cutaneous pain. However, information is currently lacking on comparisons between pain and unpleasantness NRS scores for the masseter muscle (deep pain). The present results suggest that when palpating the masseter muscle, intesnity of pain and unpleasantness are also tightly linked to stimulus intensity.

Past studies have shown that reductions in pressure pain threshold (PPT) and prolonged aftersensations are linked to chronic pain, including fibromyalgia syndrome (FMS).[40–42] Repetitive thermal stimuli applied to FMS participants produced not only increased wind-up compared to healthy participants,[40,43,44] but also prolonged aftersensations, both of which reflect central sensitization.[45,46] The present results showed significant differences in the duration of aftersensation between 0.5- and 1.0-kg stimulus intensities, between 0.5- and 2.0-kg stimulus intensities, and between 1.0- and 2.0-kg stimulus intensities. These results suggest that the magnitude of palpation intensity correlates with NRS scores and the duration of aftersensations in healthy participants. Although the present study did not reach any conclusions regarding the relationship between duration of aftersensation and central sensitization, the results suggest that the duration of aftersensation may represent important information about aftersensations in healthy individuals. Further studies are needed to directly compare duration of aftersensation between healthy participants and patients with myofascial pain.

Entropy[29] can be used to assess the diversity of mechanical sensitivity scores within the spatial distribution of the masseter muscle. Schiffman et al[16] recently indicated that the distribution of pain should be considered to better characterize the different diagnoses of myalgia. Nevertheless, little information is available about the distribution of NRS scores/mechanical sensitivity within the masseter muscle.[31] Low entropy values suggest that the mechanical sensitivity scores of the masseter muscle are quite uniform. This means that scores in the 15 test sites of the masseter muscle do not differ markedly from each other. However, it is unclear whether a similar feature characterizes myofascial pain patients. Further studies are needed to evaluate the entropy of NRS scores in patients and in healthy participants.

The present results showed that entropy values obtained with the 0.5-kg stimulus intensity were significantly lower than those with the 1.0- and 2.0-kg stimulus intensities. However, no significant differences in entropy values were seen between 1.0- and 2.0-kg stimulus intensities. This suggests that increasing intensity is associated with high entropy values, even though no significant differences were evident between 1.0- and 2.0-kg stimulus intensities.

Further studies are needed to investigate the relationship between pain intensity and entropy values to clarify the variability of mechanical sensitivity in masticatory muscles.

Kothari et al[25] showed that CVs for 2 seconds of palpation were significantly higher than for 10 seconds and suggested that control over palpation is difficult to achieve in a period as short as 2 seconds. As a result, 0 seconds was defined as the time at which the examiner felt stability of pressure for each target stimulus in the present additional experiment. The results showed no significant difference in CVs between times 0 seconds and 5 seconds. The results also suggest that palpation of the masseter muscle with a palpometer can improve standardization of the actual force value for the masseter muscle compared to palpation without a palpometer, in accordance with previous studies.[23,25]

Conclusions

The present systematic study has shown that referred pain/sensations in the orofacial region is a frequent phenomenon among healthy individuals during standardized palpation of the masseter muscle. Interestingly, referred sensations were not dependent on test site, whereas NRS scores of perceived pain and unpleasantness were. This observation could indicate differences in the mechanisms underlying mechanical pain sensitivity and referred pain/sensations from the masseter muscle, which could have implications for muscle pain diagnosis according to the DC/TMD.

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