The Effect of Topical Anaesthesia on Pressure Pain Thresholds

in Symptom-free Subjects

Umeå University

Anna Axelsson, Elsa Carlson

Tutors Anders Wänman and Birgitta Häggman Henrikson

ABSTRACT

Palpation, a standard procedure in the diagnostic process of muscular pain conditions – *myalgia,* aims to provoke a muscular pain response. The origin of pain elicited by palpation is not fully understood. There is also a possibility that the pressure elicit response from mechanoreceptors in skin, sub mucosal tissue or periosteum, which may affect the validity of the method.

The aim of this study was to evaluate the effect of topical anesthesia on pressure pain thresholds at masseter, temporal and thumb muscle sites in healthy individuals.

Twenty symptom-free subjects (ten males/ ten females) mean age 24.6 years (SD 2.6) participated in this double blind randomized experimental trial. An algometer (Somedic AB) was used to measure Pressure Pain Thresholds (PPT), on the masseter, temporal and thumb before and after topical application of EMLA or a placebo cream.

There was no statistically significant change in PPTs at any of the tested sites between the baseline and after application of EMLA and placebo cream. Furthermore, there was no significant difference in PPTs between sexes. There was no significant difference in PPTs between the different muscle sites, apart from a higher PPT for the thumb site compared to the masseter muscle.

In conclusion, PPT was not affected by topical anaesthesia in symptom-free subjects. This indicates that palpation pain is not dermal. Furthermore did PPT not differ between the masseter and temporal muscles. The study indicates that the same palpation pressure may be used for extra-oral palpation of the temporal and masseter muscle for both sexes.

INTRODUCTION

In the 1950's Travell and Simons (Travell and Simons, 1983) advocated that muscle pain was the origin of many diffuse pain conditions in the body. The hypothesis was adopted by Schwartz (Schwartz, 1955) and Laskin (Laskin, 1969). The previous theory suggested by Costen (Costen, 1934) which related unstable occlusion to temporomandibular joint (TMJ) disorders was rejected (Molin, 1999). Instead it was suggested that muscle pain and the development of TMJ-disorders was related to psychological factors such as anxiety and stress. These hypotheses lead to a paradigm shift in the field of TMJ disorders.

Studies of muscle activity with the aid of electromyography (EMG) indicated that pain within the masticatory muscles showed similarities with pain during spasm conditions (Molin, 1999). The results provided an explanation of how, but not why pain developed. TMJ-disorders also displayed similarities to work-related musculoskeletal disorders elsewhere in the body. Thus, overstrain of muscles may develop after sustained strain of specific muscle-fibres while other parts of muscles may have a lighter workload (Molin, 1999). Today both psychological factors and pain physiology are viewed to be important for the aetiology of temporomandibular disorders (TMD) (Laskin, 2007).

The prevalence of TMD pain is higher among women than among men (Marklund and Wanman, 2008) and studies indicate that women have a lower pressure pain threshold at jaw muscle sites than men (Fredriksson et al., 2000). It has accordingly been discussed whether pain conditions are associated with gender (Fillingim, 2000), differences in central processing of nociception or other variations in biological, physiological, psychological, social and hormonal factors (Racine et al., 2012a). The results are yet not conclusive due to various methodological reasons (Racine et al., 2012b). Today, the same criteria and muscle palpation pressure are used for diagnosing men and women. If there is a difference between the PPTs between asymptomatic men and women the palpation pressure should perhaps be adjusted in coherence with sex-differences.

The sensory receptors that signal pain and discomfort to the central nervous system (CNS), via the afferent neurons, are mechanoreceptors termed nociceptors. They are

located in the skin, mucosa, cornea, muscles, joints, as well as in the periosteum and bone. There are several types of mechanoreceptors, which respond to different stimuli. Some of these are wide ranged, responding to both tactile and noxious stimuli, whereas others are specialized in noxious mechanical and thermal stimuli. These specialized nociceptors also transmit signals of potential tissue damage to the CNS. Thus the nociception differs from the term *Pain*, which concerns a cognitive process in the brain. According to the International Association for Study of Pain (IASP) the definition of pain is "an unpleasant sensory and emotional experience associated with actual or potential tissue damage, or described in terms of such damage "(Merskey and Bogduk, 1994).

Pain can be experienced regardless of nociceptive signalling, even if it normally is the response to a nociceptive input. The signal is modulated by the CNS before reaching its cortical destination (Okeson, 2008) and can be both magnified and reduced. The CNS thus has a significant influence how the signal is perceived. An individuals' reaction to pain can be termed suffering (Okeson, 2008). This is an emotional experience, affected by an individual's expectations, previous knowledge and consequences, involving physiological, psychological and social factors. Hence, suffering, nociception and pain intensity do not necessarily coincide.

Muscular pain is clinically diagnosed by the use of palpation. Palpation is defined as "*the application of the fingers with light pressure to the surface of the body for the purpose of determining the consistency of the parts beneath in physical diagnosis*" (Anderson et al., 2012). It is a cost-effective and available technique used worldwide even though its validity may be questioned, since the origin of the elicited pain has not been determined.

Temporomandibular disorders refer to conditions in the jaws including pain, TMJ sounds and limited jaw movement capacity. The criteria used for diagnosing specific TMD conditions were recently revised by the International DC/TMD Consortium Network (Schiffman, 2014). In the Axis 1 classifications system both symptoms and specific signs are assessed. The diagnosis of myalgia is based on a history of pain

located in the masseter or temporal muscles, within the past month together with clinical signs of familiar pain during extra-oral palpation over the temporal - and/or the masseter muscle with a palpation pressure of 1 kg. During examination in accordance with the criteria set up by the International DC/TMD Consortium Network the masseter and temporalis muscles are both extra-orally palpated with the same pressure (1 kg) and equally important for giving the diagnosis. A significant difference in PPTs between these two muscles would indicate that the palpation pressure over these areas ought to be adjusted.

The examination procedure should have a high specificity (i.e. the probability of finding patients that are truly free from a TMD), with these included diagnostic criteria, together with a high sensitivity (i.e. identifying patients correctly diagnosed with a TMD diagnosis) (Schiffman, 2014). For both the clinician and for the individual a high specificity and sensitivity is beneficial since it helps to arrive at a correct diagnosis. It is also of importance for the society that unnecessary costs due to misdiagnosis and ineffective treatments are avoided. If a pain response can be elicited from superficial dermal nociceptors and not from nociceptors more deeply in the muscle tissue the specificity may be affected. The pressure pain threshold (PPT) is defined as the minimum force required to elicit a sensation of pain. This threshold can be measured with the aid of an algometer which is a device that quantifies pressure.

The overall aim of this study was to examine if PPT over masseter and temporal muscles is affected by application of topical anaesthesia in men and women. Our first hypothesis was that there would be no significant change in the PPT following application of topical anaesthesia in healthy subjects. The second hypothesis was that there would be a difference in PPTs between men and women. The third hypothesis was that there would be no difference in PPTs between the different muscle sites.

MATERIAL & METHOD

Subjects

Ten men (mean age 24.7 yrs., SD: 2.8) and ten women (mean age 24.5 yrs., SD: 2.5), participated in the study. The inclusion criteria used were asymptomatic individuals with no history of temporomandibular disorders. Exclusion criteria were pregnancy, elite athletes and current use of analgesics. Recruitment was made by advertisement. All subjects were students at the Medical faculty, Umeå University. The screening examination to exclude subjects with temporomandibular disorder was carried out by a dentist specialized in TMD according to the RDC/TMD criteria (Dworkin and LeResche, 1992). The information given to the participants two days before the study included prohibition of alcohol consumption and exercise 24 hours prior to the examination.

In total 21 women and 18 men responded to the advertisement, of these 12 men and 14 women were screened. The subjects were included consecutively according to the inclusion criteria until we had a total of 20 healthy participants (10 women and 10 men).

Definition of test sites

The test sites were the anterior part of the temporal muscle bilaterally, the central part of the masseter muscle bilaterally and the abductor pollicis transversa muscle of the test person's dominant hand. The test sites were determined by palpation and marked with a pen (Fig. 1). The subject was seated in an upright position in a seat with backrest and instructed to keep their mouth closed and jaws in a relaxed position.

Algometric recording

The order of examined sites was randomized using SPSS v 20 (random number function). An electronic algometer (Somedic AB) with a probe area of one square centimeter was used according to the manual for the PPT measurements with the pressure amplitude increased by 20 kPa/s. The examiner was calibrated beforehand to perform a constantly increasing pressure with the aid of a graph on the computer screen indicating the increase of pressure amplitude. The computer connected to the algometer registered the exact values for the PPTs.

The algometer included a stop-button, which the subject pressed when the PPT was reached. Before the recordings a test trial was carried out on m. abductor pollicis transversa on the non-dominant hand.

Every test site was measured three times consecutively, hence a total of 15 registrations were carried out for the baseline recording. Thereafter a topical anesthetic cream (EMLA 25mg/gram, Astrazeneca) and placebo cream (LIV, skin lotion Lahega Kemi AB) was applied to the test sites in randomized order. The creams were applied with the aid of a cut-out template with a two-centimeter diameter centered on the pen marking. The creams were applied in a thick layer covering the skin in the decided area and left on the skin for five minutes, and then removed. The same procedure for the second set of measurements, as in the baseline recording, was followed.

The study had a double-blind design with both subjects and the examiner performing the measurements kept unaware of which cream that was used on respective site. To eliminate differences in the execution of the examinations a test protocol was made prior to the experimental part.

Literature review

A literature search was done on PubMed using a free text search with the terms (without citation-marks); "pressure pain threshold algometer", which resulted in 267 studies. Including an additional term "temporalis" the search "pain pressure threshold algometer temporalis", narrowed the search down to 36 studies of which two references were deemed relevant and included. The search was repeated with the term "masseter" instead of "temporalis" which resulted in 33 studies of which three were included. Further terms used; "EMLA cream pressure pain threshold" resulted in five studies of which one reference was included. All together this resulted in a total of 75 articles, which were screened by titles and abstracts. We included studies using methods with pressure pain, masticatory muscles and non-symptomatic subjects. Of these, we excluded articles not written in English and not available in full text. The relevant articles were hand searched for further references. In Pubmed, we used the "related articles" function to expand our search. Additional articles were recommended by our

tutors. Inclusion criteria for textbooks on physiology and anatomy were physiology of pain perception, anatomy of the head and availability.

Statistical methods

During the experiment the data was recorded in Somedic software and then transferred to Excel and to SPSS v. 20 for the statistical analysis. An analysis with Q-Q plot indicated that the data was normally distributed. To individually assess the significance of the differences between the mean pressure pain thresholds, before and after application of EMLA and placebo cream, paired samples t-test was used. Comparisons of PPT values between men and women as well as between sites were done with independent sample t-test. The null-hypothesis was rejected if the P-value was < 0.05.

Ethical reflection

Prior to the experiment, the subjects received oral and written information about the test procedure and the possibility to discontinue the experiment at any time. Detailed information about all aspects of the test procedure was not presented to the participants due to the double-blind design of the study. The information stated that the participation was voluntarily and all subjects signed a written informed consent. All data gathered in the study was kept confidential and not spread to unauthorized personnel. The individual results and personal data will be kept in a safe deposit for ten years in the department of Odontology, Umeå University. The subjects volunteering for the study received a compensation for their participation (200Sek).

The pressure caused by the algometer can cause transient pain and flush of the skin for the subjects. The pain is at a low-level since we measured the perception and not the tolerance threshold; it is transient and controlled by the subject with a switch button to signal when the sensation of pain was evoked. The subjects were carefully instructed to signal the perception of pain immediately to avoid unnecessary pain and discomfort. No side-effects were expected for the subjects due to participating. The algometer was used in accordance to the manual and for safety not connected to the main power supply during the experiments. The subjects were instructed to abstain from alcohol, painkillers

and heavy physical activities 24 hours prior to the experiment. The benefit of the study for further research on diagnostic methods related to temporomandibular disorders and its use in clinical practise was considered greater than the inconvenience caused to the participants during the experiments. A local ethical committee approved the study and the research protocol.

RESULTS

The mean values and standard deviation (SD) of PPT before and after appliance of EMLA and placebo creams are presented in Table 1. There was no statistically significant difference in PPT between baseline and after the application of EMLA or placebo cream at the tested sites. The PPTs for the different sites are presented in box plots before and after application of the EMLA and placebo creams (Fig. 2). No statistically significant difference in PPTs was found between men and women (Fig. 3). At baseline there was no significant difference in PPT between the masseter and the temporal muscle sites, nor between temporal and the thumb muscle sites. A significant difference between the masseter muscle and the thumb was observed with a higher mean PPT for the thumb muscle compared to the masseter muscle (P= 0.01).

DISCUSSION

The main finding of our study was that there was no statistically significant difference in PPTs after applying topical anesthesia to masseter and temporal muscles sites in symptom free individuals. The PPT values at temporal and masseter muscle sites did not differ significantly between men and women or between these two muscles.

The mean PPT values in the present study were higher for the females and lower for the males for both masseter and temporal muscles sites compared to one previous study (Fredriksson et al., 2000) and lower for females and higher for the males for the masseter muscle compared to another study (Komiyama and De Laat, 2005). These differences may be related to the different inclusion criteria and ages of subjects in these studies. The mean age of the participants in our study was similar to the subjects included in the latter study (Komiyama and De Laat, 2005) while the mean age in the first study (Fredriksson et al., 2000) was somewhat higher. Age may affect pain

perception, at least there seems to be a difference between younger and older subjects (Gibson and Farrell, 2004). How much age accounts for the difference between the studies is difficult to evaluate.

The finding that PPT did not differ significantly between men and women is in accordance to Isselee et al (Isselee et al., 1997) but contradictory to other studies (Fillingim, 2000; Fredriksson et al., 2000). The inclusion criteria in the other studies were asymptomatic subjects for pain in the head and neck region but pain to palpation was not assessed. In our study the participants were screened for both absence of subjective symptoms and clinical signs of dysfunction. The study population thus represented young individuals without symptoms and without signs of tenderness to palpation. This difference in inclusion criteria might be a possible explanation of the lack of a significant difference between men and women. The PPT values in our study were within the range of the other studies, indicating that our results should be regarded as reliable for the included sites and symptom free individuals.

When comparing the examined sites the PPT was highest for the thumb, and lowest for the masseter muscle which is in accordance with a previous study (Isselee et al., 1997). One explanation for this difference between the sites can be anatomical differences. The masseter and temporal muscles both have bone with periosteum underneath, contrary to the pollicis transversa muscle on the thumb. Thus, the higher PPT values of the thumb compared to masseter and temporalis muscles, might be due to a response from nociceptors in the periosteum (Zhao and Levy, 2014). The temporal muscle has a thick fascia covering the muscle tissue (Sobotta, 2011). The fascia is perhaps to some extent protecting the muscle against mechanical pressure. In summary our baseline PPT values were similar to those of the studies used for comparison (Fredriksson et al., 2000; Isselee et al., 1997; Komiyama and De Laat, 2005)

The probe of the algometer used was to resemble the size of a fingertip, which is a frequently used size in other studies (Takahashi et al., 2005). During the experiment we experienced that the tip of the algometer had a tendency to slip of the muscle; hence it was difficult to keep in the correct position, especially after the cream had been applied.

There were also difficulties in gradually increasing the pressure of the algometer at the predetermined rate even though the examiner had practiced beforehand. We also had some problems with the software from Somedic, which meant that some of the registrations had to be repeated, which may have affected the results. The subject pressed a button when the PPT was reached which stopped the recording, and thereby limited the time lag between the time when PPT was reached and the moment when the pressure was stopped. The increase of pressure used in this study was 20 kPa/sec. Previous studied has used a value from 30 to 50 kPa/sec. We have not found any studies giving arguments for best practice of increasing pressure in PPT measurements.

In this study we used topical application of EMLA ®, a combination of prilocain and lidocain, which is an effective dermal anesthetic (Barcohana et al., 2003). The application time, five minutes, for the EMLA-cream was chosen in order to only have a superficial anesthetic effect. In the clinical use the recommendation of application before needling is 60 minutes or longer (Kosek et al., 1999). Since we wanted a superficial anesthetic effect on dermal skin, five minutes duration was considered enough. The effect of the EMLA-cream is dependent on the skins thickness and condition, which might have affected our results. (Läkemedelindustiföreningens Service AB, 2014). Some subjects claimed to have felt an anesthetic effect after the application of cream, even for some of the placebo sites. From this, we conclude that it is difficult to determine if we obtained the expected anesthetic effect of the EMLA-cream. Further research may be needed to determine the anaesthetic effect of EMLA on the dermal nociceptors and the time and dosage needed to prevent spread the aesthetic effect to deeper tissues.

The subjects and the examiner were blinded to on which sites the EMLA-cream was used. Therefore the psychological effect of previous bias was limited. The subjects deliberately decided to press the button, when they consciously were able to percept pain. This may have affected the results as the subjects subjective perception of pain may not be the same as the "true" pressure pain threshold.

It has been discussed that the sex of the investigator might affect the subjects' response to pain. An investigator of the opposite sex might lead to higher PPT due to the subjects being biased by their genus (Gijsbers and Nicholson, 2005). Other studies has concluded that the sex of the investigator is of no clinical significance for the PPTs and therefore an irrelevant factor (Racine et al., 2012a). In this study of difference between baseline and after application of a cream this may not be relevant for the result, as the subjects were their own control, but using a female investigator for all subjects may have affected the overall levels of the PPTs. The overall difference in PPT levels between men and women was not substantial, and followed the same pattern and approximate levels.

In asymptomatic individuals an inter-individual variability of the PPTs has been found, even though the PPTs have been seen to be relatively constant intra-individually (Kosek et al., 1999). However, other studies have concluded that for the first measurement the values are significantly higher than in the following and should thus be dismissed as misdirecting (Fredriksson et al., 2000). For this study we used the mean value of three measurements to increase validity.

An overstrained muscle can develop peripheral hypersensitisation of the nociceptors, (Miles et al., 2004). Therefore, the conclusions reached by the present study may not be transferable to people with myalgia. There are different views on the rational of using healthy subjects to compare pressure pain perception between genders, as well as the clinical significance of these studies (Racine et al., 2012a). In the diagnostic process it is essential that the normal pattern is known and palpation tenderness is frequently observed in the general population (Wanman, 1995).

We transferred our mean PPTs from kilopascal to kilogram per square centimetre resulting in the following mean values and 2 SD values: temporal muscle 1.9 kg/cm^2 [2 SD 1,3], masseter muscle 1.6 kg/cm^2 [2 SD 1,1]. An interpretation of 2 SD is that the lowest pressure to elicit a pain response in an otherwise symptom free subject of the temporal and masseter muscle would be approximately > 0,5 kg/cm^2. This might indicate that recommended 1 kg pressure can be slightly too high to reach an optimal specificity. To

judge best practice also the methods sensitivity must be calculated and that was not possible in this study with reference to only symptom free subjects included. The result of our study can thus not resolve the question of best practice in palpation pressure technique but can be used as a contribution to the debate. In the clinical perspective, the non-significant difference of the PPTs between the sites indicates that the same pressure can be used for the masseter and the temporal muscles. The pressure of one kilogram used when diagnosing TMD pain may be specific enough to exclude most of asymptomatic subjects.

Further research is required to determine the anaesthetic effect of EMLA on the dermal nociceptors and the time and dosage needed to prevent spread to deeper tissues. Additional research should be done on sex differences in PPTs in asymptomatic subjects examined by palpation to exclude TMD.

As we saw a significant difference in PPTs between the thumb and the masseter muscle a study on eventual difference between trigeminally and spinally innervated areas may be fruitful. The influence of nociception from the periosteum at extraoral palpation should also be analyzed.

In conclusion, the non-significant difference in PPTs indicate that palpation pain, in the temporomandibular area in asymptomatic persons after applying dermal anesthetics for five minutes, is not dermal. The same palpation pressure can be used on men and women. The same palpation pressure can be used on both the masseter and temporal muscle sites.

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REFERENCES

Anderson DM, Novak PD, Jefferson K, Elliott MA (2012). Dorland´s Illustrated Medical Dictionary. 32nd ed. Philadelphia: Elsevier Saunders.

Barcohana N, Duperon DF, Yashar M (2003). The relationship of application time to EMLA efficacy. *J Dent Child (Chic)* 70(1)*:*51-54.

Costen JB (1934). Syndrome of ear and sinus symptoms dependent upon disturbed function of the temporomandibular joint. *Ann Oral Rhinol Laryngol* 43(1)*:*1-15.

Dworkin SF, LeResche L (1992). Research diagnostic criteria for temporomandibular disorders: review, criteria, examinations and specifications, critique. *J Craniomandib Disord* 6(4)*:*301-355.

Fillingim RB (2000). Sex, gender, and pain: women and men really are different. *Curr Rev Pain* 4(1)*:*24-30.

Fredriksson L, Alstergren P, Kopp S (2000). Absolute and relative facial pressure-pain thresholds in healthy individuals. *J Orofac Pain* 14(2)*:*98-104.

Gibson SJ, Farrell M (2004). A review of age differences in the neurophysiology of nociception and the perceptual experience of pain. *Clin J Pain* 20(4)*:*227-239.

Gijsbers K, Nicholson F (2005). Experimental pain thresholds influenced by sex of experimenter. *Percept Mot Skills* 101(3)*:*803-807.

Isselee H, De Laat A, Lesaffre E, Lysens R (1997). Short-term reproducibility of pressure pain thresholds in masseter and temporalis muscles of symptom-free subjects. *Eur J Oral Sci* 105(6)*:*583-587.

Komiyama O, De Laat A (2005). Tactile and pain thresholds in the intra- and extra-oral regions of symptom-free subjects. *Pain* 115(3)*:*308-315.

Kosek E, Ekholm J, Hansson P (1999). Pressure pain thresholds in different tissues in one body region. The influence of skin sensitivity in pressure algometry. *Scand J Rehabil Med* 31(2):89-93.

Laskin DM (1969). Etiology of the pain-dysfunction syndrome. *J Am Dent Assoc* 79(1):147-153.

Laskin DM (2007). Temporomandibular disorders: the past, present, and future. *Odontology* 95(1):10-15.

Läkemedelindustiföreningens Service AB LFv (2014). [Online][Cited 2014 Oct 27] EMLA AstraZeneca Kräm 25mg/g+25mg/g. Available from: http://www.fass.se/LIF/product?userType=0&nplId=19841101000029.

Marklund S, Wanman A (2008). Incidence and prevalence of myofascial pain in the jaw-face region. A one-year prospective study on dental students. In: Acta Odontol Scand. Norway, pp. 113-121.

Merskey H, Bogduk N (1994). Part III: Pain Terms, A Current List with Definitions and Notes on Usage. 2nd ed. Seattle: IASP Press, IASP Task Force on Taxonomy.

Miles TS, Nauntofte B, Svensson P (2004). Clinical oral physiology Copenhagen. Denmark: Quintessence Publishing Co. Ltd.

Molin C (1999). From Bite to Mind: TMD - A Personal and Literature Review. *International Journal of Prosthodontics* 12(279-188.

Okeson JP (2008). Management of Temporomandibular Disorders and Occlusion. 6th ed. St. Louis, Missouri: Elsevier Mosby.

Racine M, Tousignant-Laflamme Y, Kloda LA, Dion D, Dupuis G, Choiniere M (2012a). A systematic literature review of 10 years of research on sex/gender and

experimental pain perception - part 1: are there really differences between women and men? *Pain* 153(3)*:*602-618.

Racine M, Tousignant-Laflamme Y, Klouda LA, Dion D (2012b). A systematic literature Review of ten years of research on sex/gender and pain perception - part two: do biopsychosocial factors alter pain sensitivity differently in women and men? Journal of Pain.

Schiffman E (2014). Diagnostic Criteria for Temporomandibular Disorders (DC/TMD) for Clinical and Research Applications: recommendations of the International RDC/TMD Consortium Network* and Orofacial Pain Special Interest Group. *Journal of Oral & Facial Pain and Headache* 28:1(6-27).

Schwartz LL (1955). Pain associated with the temporomandibular joint. *J Am Dent Assoc* 51(4)*:*394-397.

Sobotta J (2011). Sobotta Atlas of Human Anatomy, Head, Neck and Neuronatomy. 15th ed. Munich, Germany: Elsevier GmbH. p 42.

Takahashi K, Taguchi T, Itoh K, Okada K, Kawakita K, Mizumura K (2005). Influence of surface anesthesia on the pressure pain threshold measured with different-sized probes. In: Somatosens Mot Res. England, pp. 299-305.

Travell J, Simons D (1983). *Myofascial Pain and Dysfunction: TheTrigger Point Manual*. Baltimore: MD: Williams & Wilkins.

Wanman A (1995). The relationship between muscle tenderness and craniomandibular disorders: a study of 35-year-olds from the general population. *J Orofac Pain* 9(3)*:*235-243.

Zhao J, Levy D (2014). The sensory innervation of the calvarial periosteum is nociceptive and contributes to headache-like behavior. *Pain*.

Table 1. Mean pressure-pain thresholds (PPT) and standard deviations (SD) and significance levels (P) before and after the application of placebo cream or a topical anaesthetic (EMLA). The test sites were the anterior temporal muscle (temporalis), the belly of the masseter muscle (masseter) and the pollicis transversa (thumb)

Test site	Placebo/EMLA	Nr of subjects	PPT before (SD)	PPT after (SD)	P
Temporalis	Placebo	22	181 (56.8)	197 (75.3)	0.076
Temporalis	EMLA	18	186 (70.6)	210 (83.4)	0.058
Masseter	Placebo	18	163 (53.6)	153 (58.2)	0.149
Masseter	EMLA	22	156 (57.0)	154 (51.4)	0.752
Thumb	Placebo	11	283 (75.1)	268 (106)	0.546
Thumb	EMLA	9	233 (68.7)	230 (65.6)	0.747
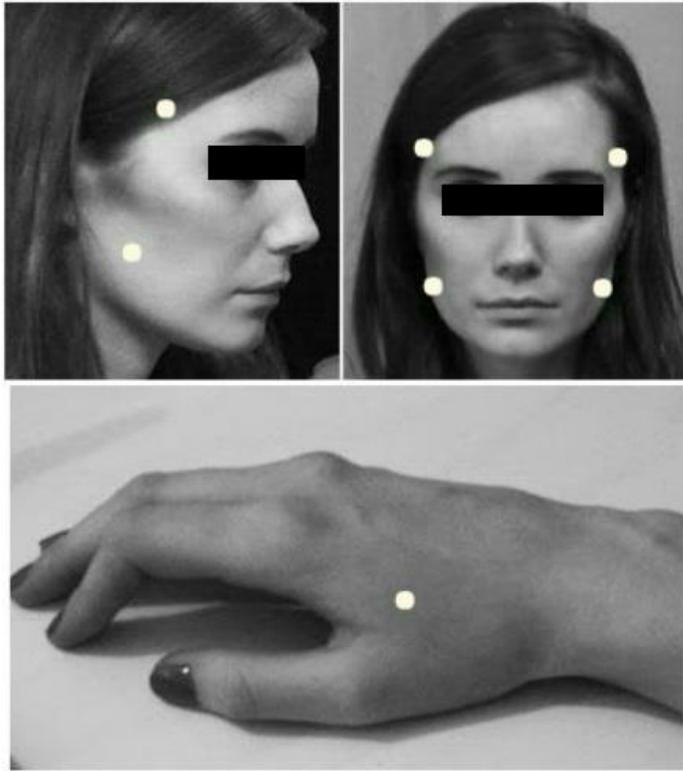
Figure 1. The white dots indicate the tested sites; anterior part of temporalis muscle, central part of masseter muscle and the abductor pollicis transversa muscle on the thumb.
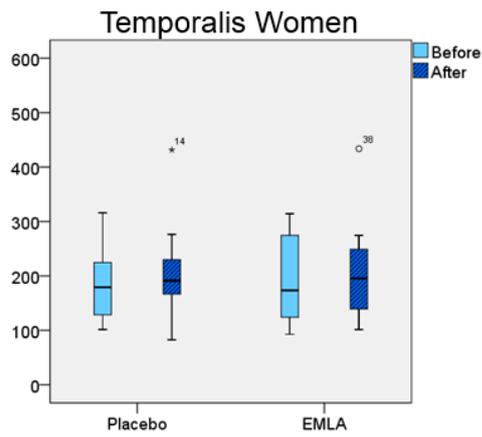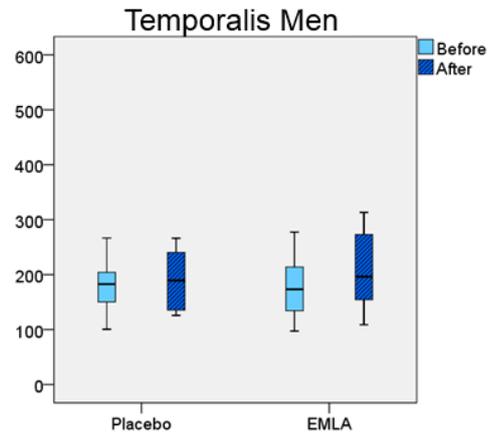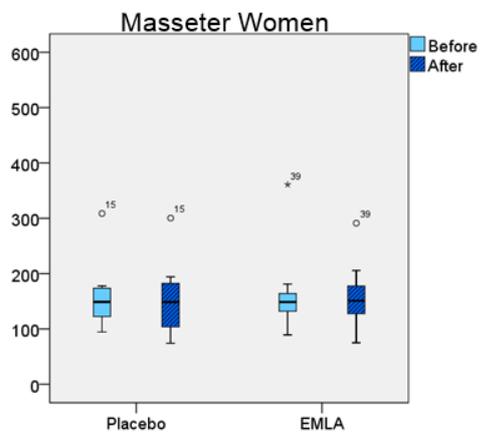
M. Temporalis

M. Masseter

A

B

Thumb

C
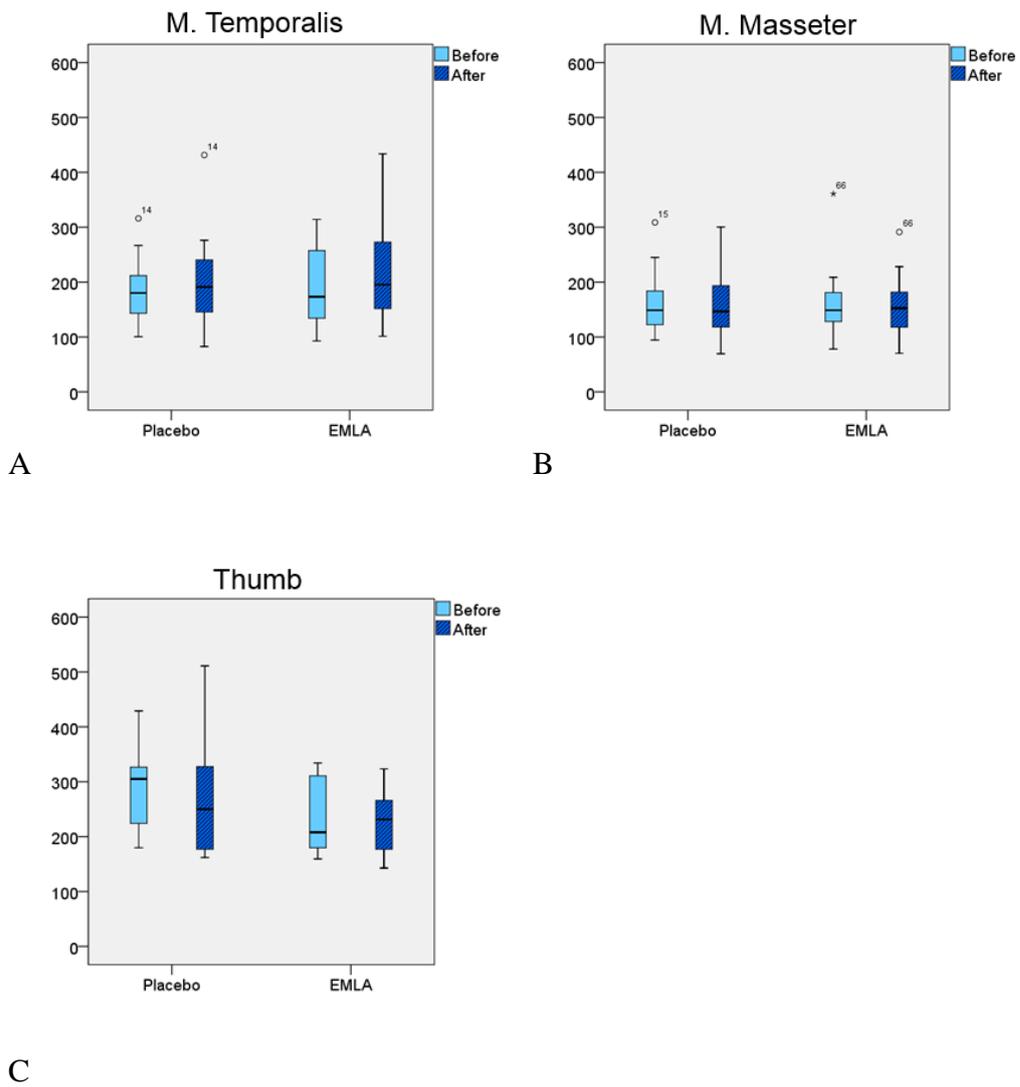
Figure 2. Mean pressure-pain thresholds before (light blue box) and after (dark blue box) the appliance of placebo-cream and EMLA-cream on the temporalis muscle (A) , masseter muscle (B) and thumb (C) for all subjects (n=20).
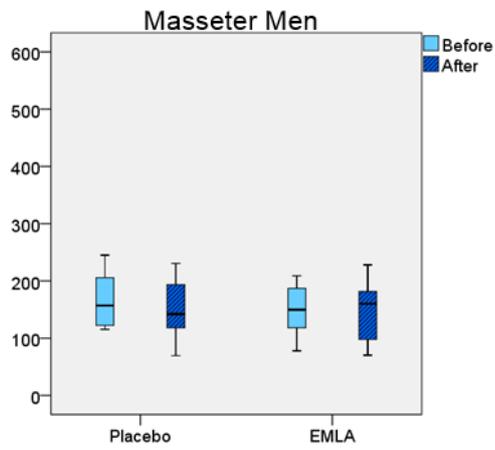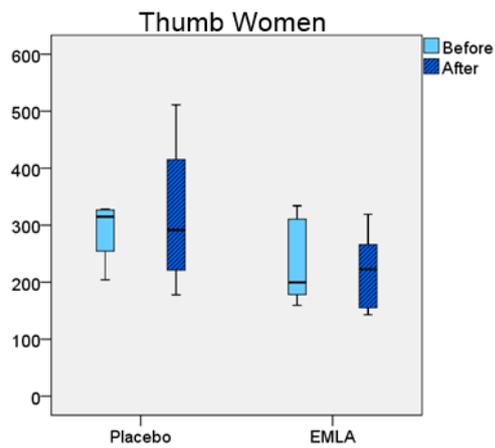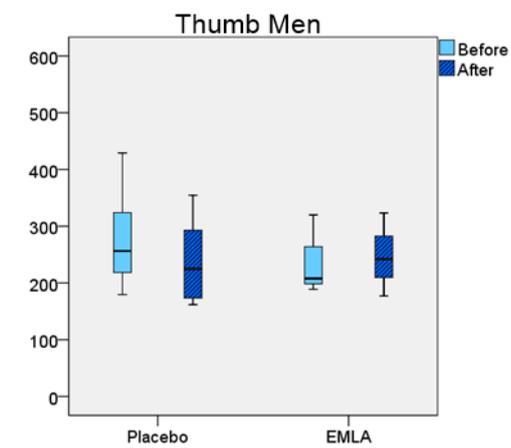
Figure 3 *Differences between men (n=10) and women (n=10) in mean pressure-pain thresholds before (light blue box) and after (dark blue box) appliance of placebo-cream and EMLA-cream on the temporalis (A-B), masseter muscle (C-D) and thumb (E-F).*