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Effect of different dental articulating papers on SEMG activity during maximum clenching

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Title:

Effect of different dental articulating papers on SEMG activity during maximum clenching.

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1. INTRODUCTION

The comprehension of the nature and patterns of occlusal contacts is of primary relevance for all dental disciplines, both for preventive education and rehabilitation treatments. Occlusal marking media are important tools used to locate interferences and refine occlusal contacts in restorative and prosthodontics procedures (Sharma et al. , 2013). The importance of occlusal control and optimization has been recently emphasized, for example in the prosthodontics discipline, for the preservation from fractures of new all-ceramic materials (Dittmer et al. , 2011), or for increasing the success of implant-based restorations (Gore and Evlioglu, 2014). From a functional perspective, historical studies have pointed out that an effective stabilization of the mandible might be achieved in presence of an harmonious occlusal pattern, defined by multiple, time simultaneous, evenly distributed tooth contacts (Myers and Anderson, 1971; Parker, 1993). Articulating papers represent a qualitative indicator used to reveal location and numbers of tooth contacts (Sharma et al. , 2013); they are the most commonly adopted media in clinical practice to perform occlusal analysis due to their low cost and simplicity of application. When ink-based or dye-containing articulating papers are chosen, biting on them is required in order to visually disclose tooth contacts; this task involves the activation of the masticatory system and its related musculature. Commercially available paper indicators differ in material composition, thickness, mechanical ductility and tensile strength; previous studies have investigated the physical features of these media (Halperin et al. , 1982), or the influence of specific properties on the appearance of occlusal markings (Saad et al. , 2008).

It has been suggested that the presence of an interocclusal material (an articulating paper should be considered essentially like that) of excessive thickness could induce a proprioceptive response that interferes with the patient's nominal path of closure and therefore distorts the indicated contact pattern (Halperin et al. , 1982). Occlusal experimental interferences (obtained using different materials/thicknesses, placed on specific locations of the arch) can alter the function of jaw muscles and temporomandibular joints (Learreta et al. , 2007; Hugger et al. , 2013).

Surface electromyography (SEMG) is an important tool for analysis of muscular physio-pathological changes during orofacial static and dynamic activities (Hugger et al. , 2008); changes in SEMG characteristics may indicate a modification in the occlusion parameters (Ferrario et al. , 2002; Trovato et al. , 2009). Several historical and more recent studies, where occlusal parameters were analyzed, suggested that the activity of the masticatory muscles at maximum efforts depends on occlusal factors such as the numbers of posterior

occlusal contacts (Helkimo et al. , 1977; Bakke, 1993; Kerstein, 2004). A higher number of posterior contacts gives a stable intercusp support that allows elevator muscles to achieve higher levels of muscular activity during clenching and chewing (Bakke, 1993; Kerstein, 2004). Moreover, alteration of the activity of temporalis and masseter were observed during clenching and chewing in presence of experimental occlusal interferences or prematurities. For example, Christensen and Rassouli have reported motor facilitation on the masseter muscle on the side of a rigid unilateral interference and motor inhibition on the opposite side during forceful clenching (Christensen and Rassouli, 1995). According to Baba, an increase in the activity of anterior temporalis ipsilateral muscle will also occur, when an interference is placed (Baba et al. , 2000). Using EMG, Belser and Hannam (Belser and Hannam, 1985) reported that lateral clenching efforts on the natural canine caused unilateral activity in the anterior and posterior temporal muscles with clear dominance on the working-side; the introduction of an experimental balancing interference changed these unilateral activity patterns to bilateral. The effects of occlusal indicators, frequently used for fine-tuning of prosthetic restorations (and in some instances for clinical discovering or regulation of interferences itself) on EMG activity of masticatory muscles are much less investigated. Occlusal indicators might also be considered as a form of interferences themselves. One study analyzed whether occlusal marking media affect the neuromuscular function: Forrester et al. used a SEMG to record muscle activity of the temporalis anterior (TA) and superficial masseter (MS) during biting and clenching. According to their findings, an elicited muscle activity similar to that of natural occlusion was revealed exclusively for some types of tested occlusal indicators (i.e: silk strip) (Forrester et al. , 2011). While Forrester et al. provided information on maximum SEMG activity for TA and MS, the coordination of muscles between right and left sides was not assessed (Forrester et al. , 2011).

The aim of the present study was to evaluate the influence of articulating paper on the SEMG activity of TA and MS muscles in healthy subjects, during maximum voluntary dental clenches (MVC) by computing indexes of muscular coordination and symmetry. Among the evaluation of three different standardized SEMG indices, a specific parameter is used to collect information on muscular coordination (comparison of muscular activities between right and left sides). The null-hypothesis tested is that the presence of a paper indicator between the lower and the upper dental arch affects SEMG activity compared with the activity recording during occlusion on natural dentition.

2. MATERIALS AND METHODS

2.1 Subjects

Thirty healthy young adult subjects (21 women - mean age in years: 30.8 ± 9.6 ; and 9 men - mean age in years: 28 ± 4.9) were recruited among institution staff. Inclusion criteria were: complete natural dentition except for occasionally missing third molars; normal physiological occlusion as defined by Mohl (1988) and Angle Class I molar and canine relation; no pathological periodontal conditions; good compliance with oral hygiene; no previous orthodontic, prosthodontics or direct restorative treatments; no signs or symptoms of temporo-mandibular disorders (TMD) according to the Research Diagnostic Criteria for TMD (Dworkin and LeResche, 1992); no current use of analgesic, anti-inflammatory and psychiatric drugs, no previous orofacial injuries or surgical procedures. The Institutional Ethical Committee approved the trial protocol and all participants gave written informed consent.

2.2 Occlusal contact areas evaluation

The occlusal status of recruited subjects was further investigated for quantification of contact areas according to the study and methods of Owens et al. (Owens et al. , 2002). Briefly, silicone-based occlusal registrations were obtained with the subjects in maximum intercuspation (static occlusion). Bite registrations were digitally imported and analyzed by a computer software to perform image processing and scientific measurements (ImageJ, NIH, US). Contact areas (mm^2) of the posterior occlusion with a thickness between 0 and $350\mu\text{m}$ were digitally measured after the following calibration steps: 1) imported images of bite registrations were converted to 8-bit channel; 2) wedges of impression material of known thickness (measurements accomplished with a Digimatic Micrometer, Mitutuyo Corp, Tokyo, Japan – accuracy of $0.001\mu\text{m}$) were used to establish the relationship between each of the 256 gray scales and the thickness of the occlusal registrations.

2.3 Data collecting

Data collection was conducted in a dental office by a single experienced examiner; the subject was seated on a comfortable chair with no headrest, with the feet resting on the floor and arms resting on the lap. The head was positioned upright and the Frankfort posture/plane was used as a positioning parameter (De Felicio et al. , 2009). The subjects were allowed to familiarize with the experimental apparatus and procedures before actual data collection.

2.4 Instrumentation and SEMG recordings and measurements

The MS and TA muscles of both sides were examined. Subject's skin was cleaned with alcohol to eliminate fat and pollution residues. Circular pre-gelled disposable electrodes (18 mm in diameter; Ag-AgCl sensor; Covidien™ Electrodes) were applied to the muscle belly, determined through palpation, parallel to the fiber direction with an inter-electrode distance of 18 mm.

For each subject, the electrodes were positioned at the beginning of the experimental session, and all trials were performed without any modification of the electrodes and/or their position.

Electromyographic activity was recorded using a wireless computerized instrument (BTS TMJOINT; BTS SpA, Italy). Two paper occlusal indicators were tested in this study: BK01 (Thickness 200 μ m; Bausch Inc., Nashua, USA – Batch n. 83048) and BK10 (Thickness 40 μ m; Bausch Inc., Nashua, USA – Batch n. 83009).

The analogue EMG signals were amplified (gain 150, bandwidth 0-10 KHz, peak-to-peak input range from 0 to 2000 μ V) using a differential amplifier with a high common mode rejection ratio (CMMR > 110 dB in the range 50-60 Hz, input impedance 10 GOhm), digitized (12b resolution; acquisition frequency up to 1KHz with +/- 2% accuracy), and digitally filtered (high-pass filter set at 10Hz; low-pass filter set at 500 Hz). The signals were averaged over 25 ms, with muscle activity assessed as the root mean square (r.m.s) of the amplitude (μ V).

For all the trials the subject was invited to clench as hard as possible and to maintain the same level of contraction for 5 s. To standardize the SEMG potentials of the four analyzed muscles with tooth contact, two 10-mm thick cotton rolls were positioned on the mandibular first and second molars of each subject, and a 5 seconds MVC was recorded.

Each test consisted of six trials: (a) MVC with cotton rolls as described above; (b) MVC on natural dentition (c) MVC onto the 40 μ m paper indicator positioned on right side of the dental arch (d) MVC onto the 40 μ m paper indicator positioned on left side of the dental arch; (e) MVC onto the 200 μ m paper indicator positioned on right side of the dental arch; (f) MVC onto the 200 μ m paper indicator positioned on left side of the dental arch.

The order of trials performed with occlusal paper indicator from (c) to (f) was randomized; no information on the nature of each indicator were given to the subject. For each trial, the operator positioned a new piece of 40- or 200- μ m paper; strips were applied to adequately cover the length and width of the teeth considered for each side (the paper extended from the most distal molar of the lower arch to canine). The operator also maintained the same working position with respect to the chair of the patient to simulate the usual working practice. To avoid any fatigue's effect of the patients' musculature, a rest period of at least 3 min was allowed

between standardization recording and tests as well as between each test. During clenching (regardless the type of trial) a relative prevalence of the activity of a particular side of TA (or MS) muscle with respect to the other side is found; for example, in case of a relative prevalence of the activity of right TA with respect to the activity of left TA, a “RIGHT” label was assigned for that trial. Vice-versa (in case of a relative prevalence of the activity of left TA with respect to the activity of right TA), a “LEFT” label was assigned for that trial. Those labels were used for subsequent elaboration of contingency tables (see next, results section).

After test recordings, a series of SEMG indices were computed as detailed by Ferrario et al. (Ferrario et al. , 2000) and Tartaglia et al. (Tartaglia et al. , 2008): POC temporal (POC_TA), POC masseter (POC_MS), anterior-posterior coefficient (APC) and IMPACT (IMP) as detailed below.

For each subject, the SEMG potentials recorded during the MVC tests were expressed as per cent of the mean potential recorded during the MVC on the cotton rolls (unit $\mu\text{V}/\mu\text{V} \times 100$). All following calculations were made with the standardized potentials.

To assess muscle symmetry, within each subject, the EMG waves of left and right masseter and temporalis anterior muscles were compared by computing a percentage overlapping coefficient (POC, unit: %). POC ranges between 0% (no symmetry) and 100% (perfect symmetry).

The APC index is a measure of the total masseter activity compared to total temporalis anterior activity (100% indicates that masticatory muscles are exactly balanced throughout the occlusion).

The IMP index, i.e. mean (masseter and temporalis) total standardized muscle activities (unit: $\mu\text{V}/\mu\text{V} \cdot \text{s}$) was computed as the integrated area of the SEMG potential over time.

To ensure reproducibility of the EMG indices the test-retest recordings were made from 50% of the sample assessed during the same session, with the subjects repeating the entire measurement protocol. The first and second sets of measurements were compared statistically.

2.5 Statistical analysis

For each subject, the differences between recordings of SEMG indices on natural MVC (POC, APC and IMP values at MVC) and values obtained with interposition of occlusal indicators were calculated (i.e: $\Delta\text{POC} / \Delta\text{APC} / \Delta\text{IMP}$ when introducing 40 or 200 μm papers). Medians and percentiles (first and third quartile) of the obtained differences were computed and used for subsequent statistical tests. Due to slight asymmetries

(detected using Kolmogorov-Smirnov normality tests) in distributions of the analyzed variables and of the obtained differences, non-parametric tests were chosen.

Descriptive statistics were computed for the subject data and analyzed variables. For description of static occlusion, the total posterior occlusal contact areas for left and right sides were calculated; for each subject, the side with highest occlusal surface was defined as dominant side (DS); this information was subsequently used in the statistics to discover potential relationships between DS and SEMG indices in MVC or during interposition of articulating papers. The EMG indices reproducibility was calculated by using Wilcoxon signed-rank tests; the Technical Error of Measurement (TEM, random error) was also computed as $[(D^2)/2 \times N]^{0.5}$, where D is the difference between the two repeated measurements and N is the number of subjects. Indicator effects on SEMG parameters, comparison between paper locations (right vs left side) and thicknesses, potential relationships with socio-demographic parameters (i.e: gender) were assessed using Mann-Whitney and Kruskal-Wallis analyses. All statistical tests were made using the JMP 8.0.2 (SAS Institute Inc., NC, USA); significance was set at 5% ($P = 0.05$).

3. RESULTS

Table 1 reports the descriptive statistics of the analyzed variables. Excellent repeatability of SEMG recordings was found; median differences between test-retest values were of 0.34, 1.59, -0.17 and 1.00 for POC_TA, POC_MS, APC, and IMP, respectively. There were no statistically significant differences between SEMG indices obtained in test and retest values ($P>0.05$, ranging from 0.32 to 0.93). Regarding the occlusal status, in 14 subjects the side with highest occlusal contact surface was left; in the remaining 16 cases it was on the right. A comparable distribution of measured occlusal surfaces between right and left sides was detected (mean % of posterior right contact surfaces out of the total posterior contact area: 49.56; mean % of posterior left contact surfaces out of the total posterior contact area: 50.44). No statistical association between occlusal side dominance and POC, APC and IMP values was found for all testing conditions ($P>0.05$).

During MVC the median values for all indices of standardized muscular symmetry (POC_TA; POC_MS) and median APCs (relative activities of masseter and temporalis muscles) were $> 80\%$. On natural dentition all recruited subjects showed POC_TA and POC_MS values within the physiological range (median POC_TA: 86.76; median POC_MS: 84.78).

The use of an occlusal indicator (both for 40 μm and 200 μm) statistically changed POC_TA and POC_MS median values ($p < 0.05$) with respect to MVC on natural dentition: the overall calculated median differences ranged from -4.37 to -7.83.

Both 40 μm and 200 μm occlusal papers did not significantly affect APC values ($P=0.869$); however the total standardized muscular activity (IMP index) statistically decreased ($P=0.004$) when placing articulating papers between the arches (median IMP values on natural dentition: 91.5; 40 μm paper: 80.5; 200 μm paper: 75.25).

Despite the use of tested occlusal indicators statistically changed two (POC and IMP) out of three investigated SEMG indices, no difference was found comparing paper thicknesses (40 μm Vs 200 μm ; $P>0.05$) or the side where the indicator was placed (Right Vs Left; $P>0.05$).

Fig.1 and Fig.2 (contingency tables) allow to see at a glance the proportion of subjects with a relative prevalence of TA/MS right or left activity during natural MVC or using a paper indicator between the dental arch (considering both 40 μm and 200 μm).

When occlusal paper was positioned to the right side of the mouth, 80% of the subjects presented a relative prevalence of the activity of right TA with respect to the activity of left TA while 61.7% of the subjects presented a relative prevalence of the activity of right MS with respect to the activity of left MS. Equally, if the occlusal paper was applied to the left side of the arch, 91.7% and 85% of the subjects presented a relative

prevalence of left TA and left MS muscles, respectively. All indices of muscular activity did not differ between sexes ($P > 0.05$).

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4. DISCUSSION

Surface electromyography (SEMG) has been suggested in dentistry to evaluate neuromuscular activity before and after several interventions to the masticatory system; for this reason, prosthodontics and orthodontics have been the main fields of application of SEMG (Hugger et al. , 2008; van der Bilt, 2011; Wozniak et al. , 2013). This technique is regarded as an objective and reliable method for evaluating muscle function and efficiency by detecting electrical potentials; the analysis of the recordings allows the assessment of general muscle activity, the cooperation of different muscles, and the variability of their activity over time (Hugger et al. , 2008; van der Bilt, 2011; Wozniak et al. , 2013). In our study we followed the EMG protocol proposed by Ferrario et al. which is based on a starting normalization record: an MVC is performed on cotton rolls just before the recording of the actual test, with the same electrodes and EMG apparatus (Ferrario et al. , 2000; Ferrario et al. , 2006). That protocol have been shown to be highly repeatable, allowing the standardization of EMG indices (like POC% and IMP $\mu\text{V}/\mu\text{S}\%$) (Ferrario et al. , 2000); moreover, it has been used successfully to discover EMG differences between healthy subjects, patients with temporo-mandibular disorders (TMD) or among patients belonging to different Research Diagnostic Criteria for TMD (Tartaglia et al. , 2008).

Variations of electromyographic signal recordings have been reported both in physiological daily activities or para-physiological conditions; for example, it is known that food interposition between the arches, and some particular specific bolus properties like size or hardness are able to influence temporal and masseter profiles during chewing (Grigoriadis et al. , 2014). On the other hand, the presence of a skeletal malocclusion (Farronato et al. , 2012), occlusal discrepancies or experimental balancing interferences produced variations of measured muscular activity with respect to control groups or between working and non-working sides (Ferrario et al. , 1999; Learreta et al. , 2007; Hugger et al. , 2013).

Jaw kinematics, that is a result of neuromuscular activation, might be altered by the presence of solid-state materials between the arches.

Interocclusal indicators are frequently employed both in clinical and laboratory environments for tooth contacts analysis; they can return qualitative (i.e: mark deepness/darkness) or quantitative (i.e: pressure values and sequence/timing of contacts) information to the operator (Millstein, 1983; Sharma et al. , 2013).

Regardless its nature or classification, an occlusal indicator is essentially like a solid-state material located between the arches: its physical properties like size/thickness, hardness or rough features, along with a potential slight impingement on intraoral mucosal surfaces, may represent a source of alteration for jaw

kinematics or neuromuscular activation. It is also known that periodontal mechanoreceptors located around the roots of the teeth signal rich information to the CNS (Central Nervous System) about tooth load and are involved in the control of the jaw muscles during biting and chewing (Trulsson, 2006; Trulsson et al. , 2010). Occlusal indicators, along with their physical properties, are certainly a source of stimulation (when subjects are biting on them) of the periodontal receptors, which play a major role in masticatory muscle reflexes (Brodin et al. , 1993). The influence of some types of occlusal indicators on mechanoreceptors – and the nature of this interaction - should be further explored. In our study maximum biting on cotton rolls and natural dentition were necessary for calibration of the electromyographic apparatus (BTS TMJOINT®): a wide range of nociceptors are excited during the above-mentioned tasks, with potential consequences on subsequent trials: while a rest period was set between trials during the experimentation (also for limiting fatigue of patients' musculature), a specific time interval to reduce bias regarding previous stimulation of nociceptors should be identified. A common clinical choice for qualitative chair-side occlusal analysis and checkings, due to low cost and ease of application, is the articulating paper (which is the specific indicator evaluated in the present study) (Millstein, 1983; Sharma et al. , 2013): its mode of action is represented by ink-release on occlusal surfaces, thus revealing upper and lower tooth contacts during clenching.

The potential influence of articulating paper on SEMG activity has been confirmed in our research: in particular, significantly reduced POC_TA and POC_MS values in comparison to MVC on natural dentition have been found, both for 40µm and 200µm paper thicknesses; in other words, a pronounced asymmetric muscular activity has been recorded with the introduction of an interocclusal media. A decrease from values obtained during MVC has been found for TA and MS muscles respectively, bringing the POC values outside the physiological standards reported by Ferrario et al. (Ferrario et al. , 2000) most of the times. While the median variations from MVC on natural dentition are quite moderate (-5.7 to -7.8 and from -4.3 to -5.8, for TA and MS muscles respectively), in some cases greater negative changes of POC values up to -25/-30 (see percentiles) were detected; they highlight an effect of articulating paper on the stomatognathic muscular system. The null-hypothesis initially formulated has been accepted: two specific SEMG parameters (POC and IMP) that are useful to assess muscle co-ordination and global standardized activity were significantly affected by the interposition of the articulating paper. Regarding the total muscular activity values on natural MVC (Median IMP on MVC: 91; IQR: 64.75-118.50), our results were in agreement with those obtained by De Felicio et al.: (Mean IMP: 110 ± 23.69) (De Felicio et al. , 2009). While slight variations (decrease) of IMP values has been reported for patients affected by TMD with respect to healthy subjects (Tartaglia et al. , 2011; De Felicio et al. , 2012;), that information is not available for comparison when placing occlusal indicators: in our study, significantly lower medians of this index were obtained with articulating papers; they

might potentially decrease the overall functional performance of the elevator masticatory muscles through different biological mechanisms (i.e: interactions with periodontal receptors) to be explored. Finally, APC values were not significantly influenced by the interposition of articulating papers ($P=0.8698$); it is possible that the specific positioning of the occlusal indicators (covering of an entire semi-arch, extending from last molar to canine) might have produced that result. When considering the sagittal plane (antero-posterior direction), the application of the paper on just few or groups of teeth - instead of the entire semi-arch - may potentially alter muscular activity.

Our findings are substantially in agreement with the investigations of Forrester et al. (Forrester et al. , 2011): to the best of our knowledge, this is the only study that applied SEMG to reveal whether occlusal media might affect neuromuscular activity. According to the above-mentioned research, articulating paper of 200 μm and T-Scan® sensor (a pressure measurement system with a thickness of 96 μm) have influenced measured SEMG activity of the masticatory muscles (MS and TA) during maximum clenches; on the other hand, AccuFilm II® (Parkell inc.) ultrathin strips (24 μm) and articulating silk (60 μm) showed no significant differences in EMG parameters when compared to natural dentition.

Forrester et al. also suggested that some specific material properties, like plasticity or stiffness, are able to modify the neuromuscular activity (Forrester et al. , 2011): the validity of tooth contact registration appears to be affected not simply by indicator thickness but also by several other physical variables, or by the ability of the media to conform/adapt to the occluded surfaces. In our experiment a change from 200 μm to 40 μm , in fact, did not produce a significant effect on masseter or temporalis POC median values. In our study, according to manufacturer safety data sheets, the chemical composition was similar between the two tested articulating papers: they were made by same components of cellulose, colored pigments and paraffin or vegetable oil.

While our study clearly pointed out that the analyzed occlusal indicators are an important factor in the modification of SEMG activity, the meaning of this change should be interpreted with caution: according to some investigations, a significant correlation exists between occlusal parameters (i.e: number/distribution of tooth contacts) and EMG parameters, including amplitude of the electric potentials and duration of contractile activity (Naeije et al. , 1989; Bakke et al. , 1992; Ferrario et al. , 2002; Trovato et al. , 2009). This way, and whether this assumption should be further confirmed, SEMG variations with respect to MVC on natural dentition might also suggest alterations in the pattern of tooth contacts or in the occlusal relationships: clinicians should be aware that using an articulating paper might influence or preclude the visualization of actual occlusal patterns (i.e: those developed without an occlusal indicator). For some patients the articulating media located in the mouth might produce unconscious movement of the lower jaw to the side

with the paper; this is a recognized clinical problem that adds up to the issues of some traditional occlusal indicators. Throughout the experimentation the movement of the lower jaw was carefully inspected: in particular, visual observation of the patient's dental midlines and proper intercuspation of upper/lower teeth during trials allowed to control mandibular deviations.

Imperfection of contemporary occlusal analysis, or at least some limitations of certain materials adopted for it, is not surprising (Millstein, 2008). For example, specific drawbacks have been described in the literature for articulating papers: they range from poor association between mark size and occlusal load (Qadeer et al., 2012), to low-reproducibility of contacts or operator-variability (Kerstein, 2008). The moisture (saliva) of intraoral environment, or some smooth materials adopted for tooth restorations (i.e: gold, metal alloys or ceramics), along with a repeated use of the indicator, are also able to affect the ink-release from articulating paper and thus impair reproducibility of markings (Saracoglu and Ozpinar, 2002). Moreover, despite a wide range of materials is available, no clear guidelines or standards have been described for proper use or for specific clinical scenarios (Millstein, 2008); we may speculate that there is still room for improvement (i.e: focusing on new occlusal indicators which should ideally have no or minimal influence on the occlusion parameters and hence on tooth contacts measured) .

Further exploration of alternative occlusal indicators should be accomplished; for example, 8 or 12 μ m ultra thin films are also available on the market even if they are recommended after a preliminary occlusal evaluation with greater thicknesses: their composition is also different, since they are made of a metallic-polyester core. Apart from chair-side evaluation and tooth contacts adjustments, a wide range of products are used for occlusal recordings and cast mountings. The influence of all the above mentioned materials on neuromuscular activity is largely unknown with potential considerable clinical consequences.

5. CONCLUSIONS

- 1) The selection and application of an occlusal indicator to support dental treatment requires careful consideration and interpretation; a choice towards media producing minimal or no change in sEMG activity might be suggested.
- 2) The examined articulating papers affected two specific SEMG parameters (POC and IMP), but did not significantly modify the APC index.
- 3) SEMG activity recorded with the occlusal indicator varied regardless left or right side positioning, and independently from tested paper thicknesses.

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6. LEGENDS

Table 1. Descriptive statistics of the electromyographic indices of the samples. All values are medians (first and third quartiles included in parentheses); for each row, p-value refers to the comparison of the three median values (entries: MVC; 40 μ m; 200 μ m) by using the Kruskal-Wallis test (level of significance was set at 5%). The statistical comparison limited to paper groups only (40 μ m Vs 200 μ m, within each row) was always not significant ($P>0.05$).

Figure 1. The contingency table illustrates the proportion of the subjects with a relative prevalence of RIGHT (Vertical legend, Red box) or LEFT (Vertical legend, Blue box) TA activity. The horizontal legend "side" indicates: MVC = a clenching on natural dentition (MVC with no paper); side *right* = a clenching with the articulating paper located on right emiarch; or side *left* = a clenching with the articulating paper located on left emiarch.

The middle column indicates the proportion during MVC on natural dentition. When occlusal paper was positioned to the right side of the mouth, 80% of the subjects presented a relative prevalence of the activity of right TA with respect to the activity of left TA. Equally, if the occlusal paper was applied to the left side of the arch, 91.7% of the subjects presented a relative prevalence of the activity of left TA muscle.

Figure 2. The contingency table illustrates the proportion of the subjects with a relative prevalence of RIGHT (Vertical legend, Red box) or LEFT (Vertical legend, Blue box) MS activity. The horizontal legend "side" indicates: MVC = a clenching on natural dentition (MVC with no paper); side *right* = a clenching with the articulating paper located on right emiarch; or side *left* = a clenching with the articulating paper located on left emiarch.

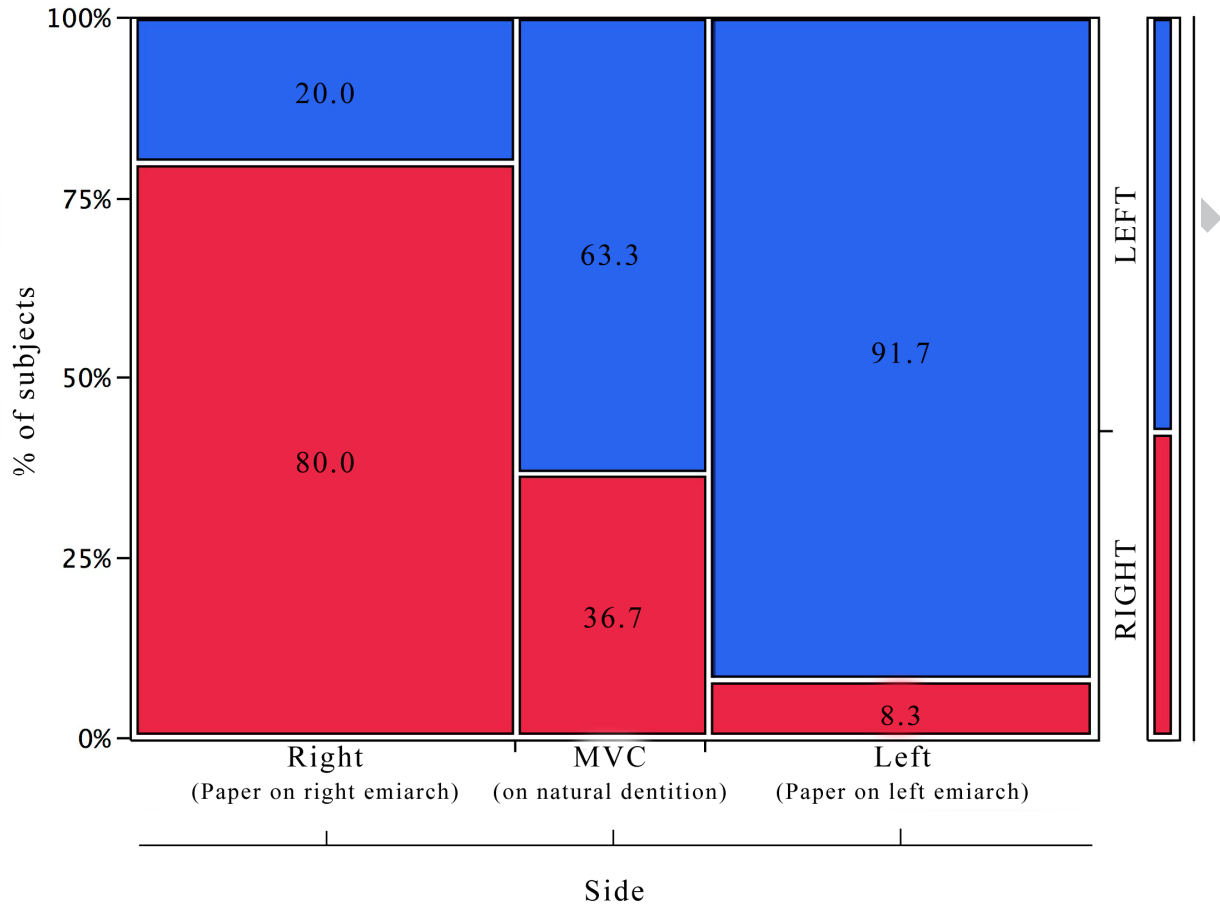
The middle column indicates the proportion during MVC on natural dentition. When occlusal paper was positioned to the right side of the mouth, 61.7% of the subjects presented a relative prevalence of the activity of right MS with respect to the activity of left MS. Equally, if the occlusal paper was applied to the left side of the arch, 85% of the subjects presented a relative prevalence of the activity of left MS muscle.

7. REFERENCES

- Baba K, Akishige S, Yaka T, Ai M. Influence of alteration of occlusal relationship on activity of jaw closing muscles and mandibular movement during submaximal clenching. *J Oral Rehabil* 2000;27:793-801.
- Bakke M. Mandibular elevator muscles: physiology, action, and effect of dental occlusion. *Scand J Dent Res* 1993;101:314-31.
- Bakke M, Michler L, Moller E. Occlusal control of mandibular elevator muscles. *Scand J Dent Res* 1992;100:284-91.
- Belser UC, Hannam AG. The influence of altered working-side occlusal guidance on masticatory muscles and related jaw movement. *J Prosthet Dent* 1985;53:406-13.
- Brodin P, Turker KS, Miles TS. Mechanoreceptors around the tooth evoke inhibitory and excitatory reflexes in the human masseter muscle. *J Physiol* 1993;464:711-23.
- Christensen LV, Rassouli NM. Experimental occlusal interferences. Part II. Masseteric EMG responses to an intercuspal interference. *J Oral Rehabil* 1995;22:521-31.
- De Felicio CM, Ferreira CL, Medeiros AP, Rodrigues Da Silva MA, Tartaglia GM, Sforza C. Electromyographic indices, orofacial myofunctional status and temporomandibular disorders severity: A correlation study. *J Electromyogr Kinesiol* 2012;22:266-72.
- De Felicio CM, Sidequersky FV, Tartaglia GM, Sforza C. Electromyographic standardized indices in healthy Brazilian young adults and data reproducibility. *J Oral Rehabil* 2009;36:577-83.
- Dittmer MP, Kohorst P, Borchers L, Schweska-Polly R, Stiesch M. Stress analysis of an all-ceramic FDP loaded according to different occlusal concepts. *J Oral Rehabil* 2011;38:278-85.
- Dworkin SF, LeResche L. Research diagnostic criteria for temporomandibular disorders: review, criteria, examinations and specifications, critique. *J Craniomandib Disord* 1992;6:301-55.
- Farronato G, Giannini L, Galbiati G, Sesso G, Maspero C. Orthodontic-surgical treatment: neuromuscular evaluation in skeletal Class II and Class III patients. *Prog Orthod* 2012;13:226-36.
- Ferrario VF, Serrao G, Dellavia C, Caruso E, Sforza C. Relationship between the number of occlusal contacts and masticatory muscle activity in healthy young adults. *Cranio* 2002;20:91-8.
- Ferrario VF, Sforza C, Colombo A, Ciusa V. An electromyographic investigation of masticatory muscles symmetry in normo-occlusion subjects. *J Oral Rehabil* 2000;27:33-40.
- Ferrario VF, Sforza C, Serrao G, Colombo A, Schmitz JH. The effects of a single intercuspal interference on electromyographic characteristics of human masticatory muscles during maximal voluntary teeth clenching. *Cranio* 1999;17:184-8.
- Ferrario VF, Tartaglia GM, Galletta A, Grassi GP, Sforza C. The influence of occlusion on jaw and neck muscle activity: a surface EMG study in healthy young adults. *J Oral Rehabil* 2006;33:341-8.
- Forrester SE, Presswood RG, Toy AC, Pain MT. Occlusal measurement method can affect SEMG activity during occlusion. *J Oral Rehabil* 2011;38:655-60.
- Gore E, Evlioglu G. Assessment of the effect of two occlusal concepts for implant-supported fixed prostheses by finite element analysis in patients with bruxism. *J Oral Implantol* 2014;40:68-75.
- Grigoriadis A, Johansson RS, Trulsson M. Temporal profile and amplitude of human masseter muscle activity is adapted to food properties during individual chewing cycles. *J Oral Rehabil* 2014;41:367-73.
- Halperin GC, Halperin AR, Norling BK. Thickness, strength, and plastic deformation of occlusal registration strips. *J Prosthet Dent* 1982;48:575-8.
- Helkimo E, Carlsson GE, Helkimo M. Bite force and state of dentition. *Acta Odontol Scand* 1977;35:297-303.
- Hugger A, Hugger S, Schindler HJ. Surface electromyography of the masticatory muscles for application in dental practice. Current evidence and future developments. *Int J Comput Dent* 2008;11:81-106.
- Hugger S, Schindler HJ, Kordass B, Hugger A. Surface EMG of the masticatory muscles (part 2): fatigue testing, mastication analysis and influence of different factors. *Int J Comput Dent* 2013;16:37-58.
- Kerstein RB. Combining technologies: a computerized occlusal analysis system synchronized with a computerized electromyography system. *Cranio* 2004;22:96-109.

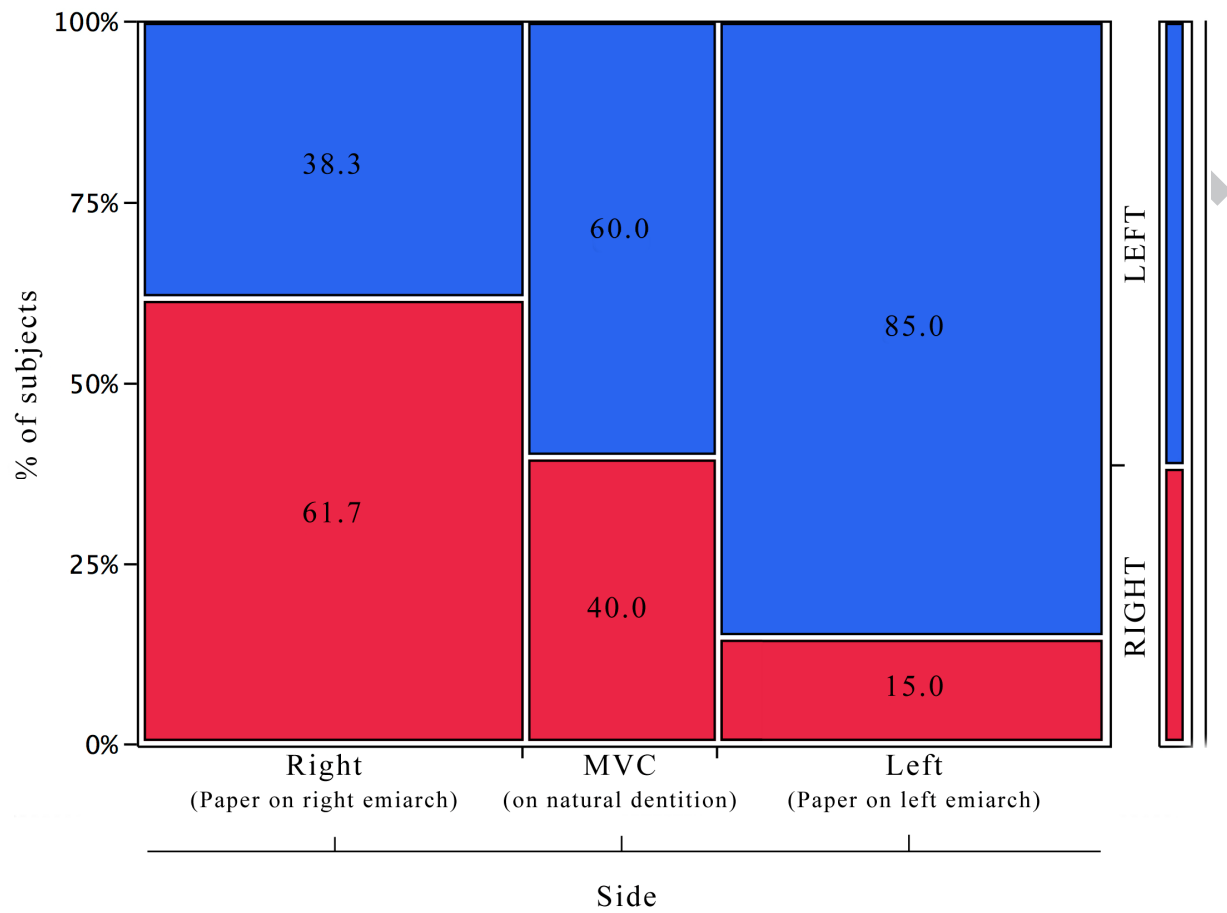
- Kerstein RB. Articulating paper mark misconceptions and computerized occlusal analysis technology. *Dent Implantol Update* 2008;19:41-6.
- Learreta JA, Beas J, Bono AE, Durst A. Muscular activity disorders in relation to intentional occlusal interferences. *Cranio* 2007;25:193-9.
- Millstein P. Know your indicator. *J Mass Dent Soc* 2008;56:30-1.
- Millstein PL. An evaluation of occlusal contact marking indicators: a descriptive, qualitative method. *Quintessence Int Dent Dig* 1983;14:813-36.
- Myers GE, Anderson JR, Jr. Nature of contacts in centric occlusion in 32 adults. *J Dent Res* 1971;50:7-13.
- Naeije M, McCarroll RS, Weijs WA. Electromyographic activity of the human masticatory muscles during submaximal clenching in the inter-cuspal position. *J Oral Rehabil* 1989;16:63-70.
- Owens S, Buschang PH, Throckmorton GS, Palmer L, English J. Masticatory performance and areas of occlusal contact and near contact in subjects with normal occlusion and malocclusion. *Am J Orthod Dentofacial Orthop* 2002;121:602-9.
- Parker MW. The significance of occlusion in restorative dentistry. *Dent Clin North Am* 1993;37:341-51.
- Qadeer S, Kerstein R, Kim RJ, Huh JB, Shin SW. Relationship between articulation paper mark size and percentage of force measured with computerized occlusal analysis. *J Adv Prosthodont* 2012;4:7-12.
- Saad MN, Weiner G, Ehrenberg D, Weiner S. Effects of load and indicator type upon occlusal contact markings. *J Biomed Mater Res B Appl Biomater* 2008;85:18-22.
- Saracoglu A, Ozpinar B. In vivo and in vitro evaluation of occlusal indicator sensitivity. *J Prosthet Dent* 2002;88:522-6.
- Sharma A, Rahul GR, Poduval ST, Shetty K, Gupta B, Rajora V. History of materials used for recording static and dynamic occlusal contact marks: a literature review. *J Clin Exp Dent* 2013;5:e48-e53.
- Tartaglia GM, Lodetti G, Paiva G, De Felicio CM, Sforza C. Surface electromyographic assessment of patients with long lasting temporomandibular joint disorder pain. *J Electromyogr Kinesiol* 2011;21:659-64.
- Tartaglia GM, Moreira Rodrigues da Silva MA, Bottini S, Sforza C, Ferrario VF. Masticatory muscle activity during maximum voluntary clench in different research diagnostic criteria for temporomandibular disorders (RDC/TMD) groups. *Man Ther* 2008;13:434-40.
- Trovato F, Orlando B, Bosco M. Occlusal features and masticatory muscles activity. A review of electromyographic studies. *Stomatologija* 2009;11:26-31.
- Trulsson M. Sensory-motor function of human periodontal mechanoreceptors. *J Oral Rehabil* 2006;33:262-73.
- Trulsson M, Francis ST, Bowtell R, McGlone F. Brain activations in response to vibrotactile tooth stimulation: a psychophysical and fMRI study. *J Neurophysiol* 2010;104:2257-65.
- van der Bilt A. Assessment of mastication with implications for oral rehabilitation: a review. *J Oral Rehabil* 2011;38:754-80.
- Wozniak K, Piatkowska D, Lipski M, Mehr K. Surface electromyography in orthodontics - a literature review. *Med Sci Monit* 2013;19:416-23.

Fig. 1



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Fig. 2



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Table 1

N=30	MVC (Natural dentition)	40µm (MVC on 40µm paper)	200µm (MVC on 200µm paper)	P-value
POC_TA (%)	86.76 (84.17; 88.47)	81.05 (68.08; 86.69)	78.93 (62.78; 84.53)	<i>0.0001</i>
POC_MS (%)	84.78 (79.85; 87.60)	80.41 (73.24; 84.59)	78.94 (70.07; 83.74)	<i>0.0002</i>
APC (%)	84.40 (78.70; 89.54)	84.77 (74.75; 88.58)	83.64 (78.12; 88.58)	<i>0.8698</i>
IMP	91.5 (64.75; 118.50)	80.5 (61.25; 90.5)	75.25 (62; 90.25)	<i>0.0048</i>

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Biography

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Keywords: surface electromyography, temporalis anterior, superficial masseter, articulating paper, occlusal analysis

Abstract: This study evaluated the influence of two different occlusal indicators (articulating papers of 40 μ m and 200 μ m) on muscular activity of the temporalis anterior (TA) and superficial masseter (MS) during maximum voluntary clenches (MVC), using surface electromyography (SEMG). It was hypothesized that an articulating paper positioned between dental arches during MVC elicits a different muscular activity compared with the occlusion on natural dentition (without the occlusal indicator).

30 healthy adult subjects with a complete, natural dentition were recruited; SEMG activity was recorded in the following experimental conditions: MVC with cotton rolls for standardization purposes; MVC on natural dentition; MVC onto the 40 μ m or 200 μ m paper indicator positioned on right or left side of the dental arch. Percentage Overlapping Coefficient (POC; separate values obtained for TA and MS), antero-posterior coefficient (APC) and total muscle activities (IMP) were the analyzed SEMG parameters.

The use of an occlusal indicator statistically changed POC_TA, POC_MS and IMP median values ($p < 0.05$). Both 40 μ m and 200 μ m occlusal papers did not significantly affect APC values ($P=0.86$). A pronounced asymmetric muscular activity has been recorded with the introduction of an interocclusal media. All indices of muscular activity did not differ between sexes (Kruskal Wallis test, $P > 0.05$). In conclusion, the examined articulating papers affected two specific SEMG parameters (POC and IMP); the recorded muscular activity with the occlusal indicator varied regardless left or right side positioning, and independently from tested paper thicknesses.