Standardised indexes reduce the variability of masticatory muscles electromyographic values

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Abstract

In dentistry, Surface Electromyography (sEMG) is currently used in both clinical and research fields to study the electrical activity of the masticatory muscles, such as the anterior Temporals and Masseters, which are easily accessible. Due to biological and technical factors that can alter the capture of the sEMG signal, comparing recordings obtained from the same subject at different days, with electrode replacement, can be challenging, thus complicating inter- and intra-subject sEMG comparisons. In the present study, sEMG activity of the masticatory muscles during maximum teeth clenching was simultaneously recorded using two different electrode configurations (A and B) to simulate different electrodes positioning as may occur between appointments. Raw sEMG signals and standardised indexes based on acquisitions from configurations A and B were compared. The position of the electrodes significantly affected the sEMG raw potentials of the masticatory muscles during maximum teeth clenching, while standardised indexes were not influenced by electrode positioning.

Key Words: standardised surface electromyography, masticatory muscles electromyography, healthy subjects, teeth clenching.

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Surface Electromyography (sEMG), thanks to its accessibility, is a non-invasive technique that provides information about muscles function as the sum of the electrical contributions of their active motor units and thus reflects both the muscle membrane properties and the control strategies of the central nervous system.[1] In dentistry sEMG is currently used in both clinical and research fields to study the electrical activities of the masticatory muscles like anterior Temporals and Masseters.[2-4] From a clinical point of view it represents an instrumental support for the diagnosis of temporomandibular disorders, the assessment of the effectiveness of orthodontic and restorative treatments and the monitoring of patients.[2-4] It is generally accepted that sEMG variables are highly affected by technical and biological factors influencing its signal capture,[5] such as electrode location and soft tissues characteristics, *i.e.* with respect to innervation zones and tendons.[6-10] Hence, the comparison of recordings obtained from the same subject at different timepoints, that may simulate position variability between appointments, may be critical,[11] thus precluding inter- and intra- subject sEMG comparison. Nevertheless, low sensitivity of the tests obtained using this methodology has been advocated, thus questioning its clinical use.[12,13] In order to reduce the 'biological noise' and allow useful comparisons between different subjects and different studies, a standardization of the procedures has been proposed.[1,14] According to the standardization protocol described by Ferrario *et al.*,[1,14] when a clinical condition such as chewing, clenching in maximal intercuspidation, swallowing or wearing an oral device like an occlusal splint needs to be tested, the electrical activities of the masticatory muscles are normalised - *i.e.* expressed as a percentage of their electrical potentials recorded during a reference acquisition represented by a maximum voluntary teeth clenching on cotton rolls placed between antagonist molars. Cotton rolls are supposed to minimize the activation of periodontal receptors so that the difference between sEMG values of the masticatory muscles during the tested condition and the corresponding values during teeth clenching with the

interposition of cotton rolls may represent a neuromuscular evaluation of dental occlusion effect on the tested condition. A further implementation of the standardization protocol includes the computation of indexes that combine with each other the sEMG normalised potentials of each masticatory muscle to evaluate their contribution to the overall masticatory function such as symmetry, synchrony, global activity and torsional effect. The normalization process with cotton rolls together with indexes computation goes under the name of standardized sEMG (ssEMG).
ssEMG data are repeatable[15] and allow an objective differentiation among several diagnostic categories defined in accord with the Research Diagnostic Criteria for Temporomandibular Disorders (RDC/TMD).[16]
Considering the scientific knowledge from several research and clinical studies based on this methodology over more than 20 years, sEMG is now considered an effective and trustworthy system that provides quantitative data on the functional condition of the masticatory muscles as a 'neuromuscular functional analysis'.[17]
However, the actual effect of the standardised procedures to overcome biological and technical critical factors of sEMG recordings has never been measured.
The aim of the present study was to quantify the effect of electrodes' positioning on the raw (non-normalised) sEMG signals and on the normalised indexes of the masticatory muscles by testing two different electrodes positions in healthy young subjects.
The experimental hypothesis is that the position of the electrodes affects the non-normalised sEMG data recorded during maximum voluntary teeth clenching, whereas the normalised indexes are less influenced by the electrodes' position.

Study design

sEMG activity of the masticatory muscles during maximum teeth clenching was simultaneously recorded with two different configurations (type A and B) of the electrodes that may simulate position variability between appointments. In type A, the electrodes were positioned on the skin area corresponding to the muscle belly of the anterior Temporals (TA) and superficial Masseters (MM) in a posterior position while in type B, the electrodes were positioned more anteriorly, as described below. In each configuration, maximal voluntary contraction (MVC) data were normalised using the cotton rolls reference acquisition values, then the standardized sEMG indexes (ssEMG) were computed. All non-normalised values and ssEMG indexes were compared between type A and B electrodes positions.

Materials and Methods

Subjects

The study protocol was approved by the local ethic committee (Università degli Studi di Milano, code DG-EMG-2016). The population was recruited among Fellow under- and post-graduate students and staff at our "Thin Sections Laboratory". According to the inclusion criteria all subjects had to i) be in good physical health, ii) be free from pathologies of the neck and stomatognathic apparatus, iii) be pain-free during clenching and iv) own a complete dentition up to the second upper and lower molar. The exclusion criteria were the following: presence of periodontal disease, partial or total removable dental prosthesis and not enough skin surface around the Temporals area to apply a pair of electrodes on each side. The experimental procedures and possible risks that could occur during the instrumental examinations were explained to all subjects. Experimental procedures and potential risks related to instrumental examinations were explained by means of an informed consent.

Electrode type and positioning

The Masseter and anterior Temporal muscles of both sides (left and right) were examined. Disposable pre-gelled silver/silver chloride bipolar surface electrodes (rectangular shape, 21x41 mm, 20 mm inter-electrode distance) (F3010, Fiab, Firenze, Italy) were used.
In order to standardize the position of the electrodes in the two configurations on each muscle, the line described by Ferrario *et al.*[1,14] was taken as a reference as follows: i) MM: the operator, standing in front of the seated subject, palpated the muscular belly while the subject clenched his/her teeth. The electrodes were fixed parallel to the exocanthion-gonion line and with the upper pole of the electrode under the tragus-labial commissural line; ii) TA: the muscular belly was palpated during tooth clenching and the electrodes were fixed parallel to the exocanthion-gonion line along the anterior margin of the muscle (corresponding to the fronto-parietal suture).
For each muscle two parallel bipolar electrodes were positioned so that the electrodes in configuration type A had their lateral margin located posteriorly to the reference lines described above, while in B the electrodes had their lateral margin located anteriorly to the reference line with a 1 mm distance to each other (Figure 1).
Therefore, a total of eight electrodes was applied to each patient to analyse the four muscles with the electrodes in the two configurations previously described. Recordings were performed in the same pattern on both sides.
To reduce skin impedance, the skin was carefully cleaned prior to electrode placement, and recordings were performed 5 min later, allowing the conductive paste to adequately moisten the skin and a disposable reference electrode was applied to the forehead.

sEMG recordings and measurements

Instrumentation

Surface EMG activity was recorded using a computerized instrument (EasyMYo, 3 Technology S.r.l., Udine, Italy). The analog sEMG signal was amplified (gain 100, bandwidth 0–1000 Hz, peak-to-peak input range from 0 to 3600 mVpp) using a differential amplifier with a high common mode rejection ratio (CMRR¼115 dB typ. in the range 0–60 Hz, input impedance 100Gohm), digitized (24 bit resolution, 4000 Hz A/D sampling frequency), and digitally filtered (high-pass filter set at 30 Hz, low-pass filter set at 400Hz, band-stop for common 50–60 Hz noise). The signals were averaged over 25 ms, with muscle activity assessed as the root mean square (RMS) of the amplitude (μV). SEMG signals were recorded for further analysis. Before acquisition session the subjects were properly trained to elicit true teeth maximal voluntary contraction using an on-time sEMG signal visualization.

Normalisation procedure

Two 10-mm thick cotton rolls were positioned on the mandibular mandibular second premolars/first molars of each patient and a 5-s MVC was recorded. For each muscle, the mean sEMG potential obtained in that first acquisition was set at 100%, and all further sEMG potentials were expressed as a percentage of this value (μV/μVx100).[14]

Analysed task

sEMG activity was recorded during a 5-s MVC test in dental intercuspation: the subject was invited to clench as hard as possible, and to maintain the same level of contraction for the whole test. For each subject, the central 3s of the MVC test were then analysed, and the sEMG potential was normalised as detailed before.

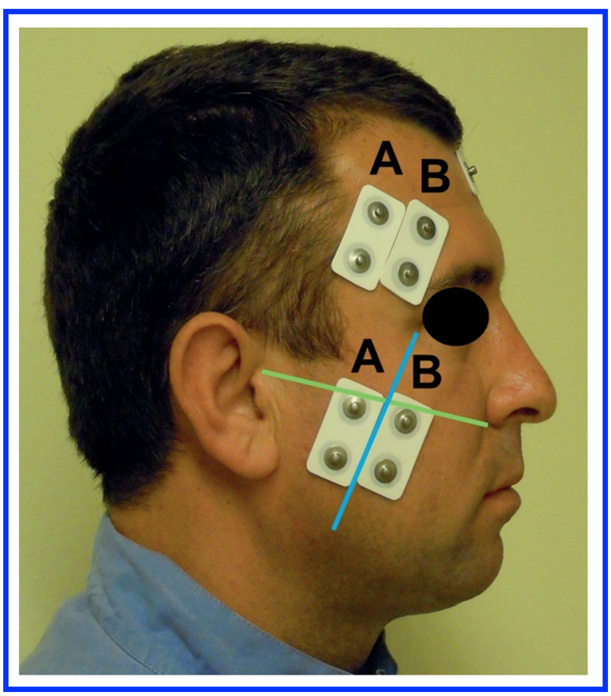

Figure 1. *Positions of the electrodes on the cutaneous area overlying the masticatory muscles of the right side in the two tested configurations: A (posterior) and B (anterior). Electrodes on the Masseter were fixed parallel to the exocanthion-gonion line (blue) and with the upper pole of the electrodes under the tragus-labial commissural line (green). Electrodes on the Anterior Temporal were placed parallel to the exocanthion-gonion line.*

sEMG data analysis

Descriptive statistics (mean and Standard Deviation, SD) were computed for RMS obtained in the two configurations. Separately from each configuration of the electrodes (A and B) normalised sEMG waves were compared by computing a series of ssEMG indexes using the instrument software tools as follows: i) the Percentage Overlapping Coefficient (POC, %),[1] an index of symmetric muscular contraction. The index ranges between 0% and 100%: when two paired muscles contract with perfect symmetry, a POC of 100% is obtained. Masseter and Temporal POCs were obtained for each subject; ii) the Torque coefficient (TC, %),[1] was assessed to evaluate if an unbalanced contractile activity of contralateral Masseter and Temporal muscles, such as that of right Temporals and left Masseter, might give rise to a potential lateral displacing component. TC ranges between 100% (complete prevalence of right Temporal and left Masseter) and -100% (complete prevalence of left Temporal and right Masseter); iii) the Asymmetry index (Asim, %),[1] was assessed to evaluate the presence of an unbalanced contractile activity of right-side muscles. Asim index ranges between 100% (complete prevalence of right-side muscles) and -100% (complete prevalence of left side muscles); iv) the Activity index (Attiv, unit %),[18] was obtained as the percentage ratio of the difference between the mean Masseter and Temporals standardised potentials and the sum of the same standardised potentials, to individuate the most prevalent pair of masticatory muscles. The index is positive (up to 100%) when the Masseter muscle standardised potentials are larger than those of the Temporals muscles, negative (up to 100%) when the Temporals muscle potentials are larger and null when they are equal; v) the Standardised activity index (Impact, %) was calculated to quantify the total muscular activity performed during MVC in respect to the standardization clenching on cotton rolls; Impact was estimated computing the mean (Masseter and Temporal) total muscle activities as the integrated areas of the sEMG potentials over time.[18]

Statistical evaluation

To quantify the effect of the electrodes position on the sEMG signals and ssEMG indexes, all values obtained from two configurations were compared. For all ssEMG indexes (unit %) and the non-normalised sEMG RMS values (unit μV), descriptive statistics (mean and standard deviation) were calculated in each configuration. Student's T tests ($p < 0.05$) were used to evaluate the systematic error and the Mean Absolute Difference (MAD) between the values of 2 sets of measurements was computed. The Technical Error of Measurement (TEM) was used to evaluate the random error.[19,20] The TEM, or Dahlberg's error, is calculated as follows:

$$TEM=\sqrt{[(\sum D^2)/2n]}$$

where D is the difference between each couple of replicate measurements and n is the number of couples. Furthermore, for indexes that can only have positive values (RMSs, POCs and Impact), the Relative Error of Measurement

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(REM) was computed as the ratio between the mean and MAD for each value.

Results

Twelve subjects (6 males, 6 females, age range 19-35 years, mean 25±5) were included in the study. All results are reported in Table 1.
Considering non-normalised sEMG values, the electrical activity of both right and left TA muscles during MVC showed a statistically significant difference between the two electrodes configurations. Non-normalised sEMG values were always higher in type A than in B for both sides; the electrical activity of MM was statistically higher in configuration type A than in B only for the left side.

Regarding the normalised indexes, no statistically significant differences between the ssEMG data from two different electrodes positions emerged. Regarding the Asim, Attiv, Torque and Impact indexes, mean values in type A and B were comparable and methods errors were minimal.

Discussion

In the current study, the effect of electrodes position on the recorded signal amplitude and on normalised indexes were evaluated. Average values of non-normalised signals in µV were significantly different in 3 muscles out of 4, underlining that the electrical signal amplitude parameter seems to be significantly influenced by the electrode position. The absolute average difference among non-normalised values

Table 1. *Comparison between sEMG mean and standard deviation (SD) values obtained with electrodes configuration A* versus *B using non-normalised raw values (RMS in µV) and standardised indexes (ssEMG, in %).*

	Configuration	Mean	SD	T-test	MAD	TEM	REM
Non-normalised values							
Right Temporal RMS	A	172	97	p <0.01	65	58	38
	B	108	73				
Left Temporal RMS	A	160	91	p <0.05	62	57	39
	B	109	54				
Right Masseter RMS	A	122	80	N.S.	43	38	35
	B	146	71				
Left Masseter RMS	A	117	56	p <0.05	59	53	51
	B	163	98				
Standardised indexes (%)							
POC Temporal	A	83	3	N.S.	2	2	3
	B	83	3				
POC Masseter	A	81	4	N.S.	2	2	3
	B	82	4				
Impact	A	98	15	N.S.	3	3	3
	B	101	14				
Asim	A	-1	10	N.S.	4	3	
	B	-1	10				
Attiv	A	-2	9	N.S.	2	2	
	B	-1	8				
TC	A	0	6	N.S.	3	2	
	B	1	5				

Non-normalised values and standardised indexes obtained from different sEMG electrodes positions.
MAD, mean absolute difference (in µV for raw values and % for standardised indexes); TEM, technical error of measurement (in %); REM, relative error of measurement (in %).

obtained in type A and B positions ranged from 43 to 65 µV with the Relative Error of Measure (REM) ranging from 35 to 51% of the mean value.

By contrast, no significant differences in the normalised indexes elaborated in type A and B electrodes configurations were found, thus confirming the effectiveness of the standardization procedure. Also, the absolute average difference among standardised indexes in the two configurations ranged from 2 to 3% and the relative error of measurement was about 3% of the average. Such results suggest that the electrode position plays an influence on non-normalised sEMG values.

The average POC values of anterior Temporals and Masseters obtained in the current study were slightly lower than the ones reported by De Felicio *et al.*[19] and by Ferrario *et al.*[1,14] in healthy subjects, probably due to a less restrictive selection of our sample. In the present study, not all subjects had Angle class I and subjects that previously undergo orthodontic and/or fixed prosthodontic treatments weren't excluded from the sample.

Regarding the typology of standardisation of the sEMG signal of the masticatory muscles, different protocols have been suggested, like for example those using as reference test the rest position, maximal or submaximal teeth clenching, clenching with cotton rolls or wax sheet between dental arches.[15]

Our model, already used in several sEMG studies,[2,3,15,16,17,21-25] uses cotton rolls as reference and includes the computation of standardised indexes based on the whole EMG waveform, improving the previously used standardised EMG indexes proposed by Naeije *et al.* in 1989,[26] based on average sEMG values.

Reducing periodontal receptors feedback through cotton rolls may represent a limitation of the standardisation procedure if the evaluation has objectives other than evaluating the role of dental occlusion in masticatory muscles recruitment. For example, other stimuli that can influence the muscular activity in both reference and tested recordings, such as intramuscular pain, cannot be discriminated by standardised indexes. Regarding clinical aspects, subjects undergoing dental therapy or suffering from temporomandibular disorders might adopt different recruitment strategies of motor units;[22] instrumental information about the functional adaptation of muscles to dental contact, may be useful for the classification of painful patients and the functionalization of dental rehabilitations that might require muscular adaption to occlusal changes. In particular, this ssEMG evaluation may represent a helpful tool to test the capacity of a single individual to accept even large modifications of the anatomy of his/her masticatory apparatus thus reducing the risk to have non-reversible damages over time.[21,25] The ssEMG protocol may suggest a caregiver about the ability of the patient's masticatory muscles to adapt to the new occlusal condition, thus integrating the clinical evaluation and guiding the choice of the optimal treatment plan for that specific individual.

sEMG is currently used in dentistry to study the electrical activity of the masticatory muscles under several conditions. The myoelectric activity signal may vary due to many factors such as changes in electrode location, modifications of tissue properties, tissue temperature, muscle resting length, velocity of contraction and fibre type.[6,7,27,28]

In the last 30 years, the effect of electrode position on the estimates of Conduction Velocity (CV), amplitude and spectral variables of surface EMG has been addressed[29] taking into account several types of muscles (or groups of muscles) ranging from the masticatory muscles to the muscles of the shoulder, of the arm and leg. The most revealing standardization effort occurred in 1997–1999 within the European Project on "Surface EMG for Non-Invasive Assessment of Muscles" (SENIAM) [www.seniam.org] where a fine analysis of literature was presented for several muscles.

Armijo-Olivo *et al.*[30] reported that "anthropomorphic differences between different recording sites and between individuals make the comparisons of the EMG signal difficult" so that no accurate comparisons of the muscular functions can be obtained by using raw electrical sEMG potentials. Furthermore, the same Authors stated that "comparing the EMG activities between subjects and under different conditions requires a process known as normalization".[30]

The same review concludes that about 60% of the electromyographic studies did not apply a standardization procedure, thus making questionable the possibility to compare data among subjects under different conditions either the same subjects in a longitudinal evaluation or different groups of individuals.[30]

Translating the experimental findings of the present study into clinical or research settings where sEMG acquisitions might require to be repeated over time in the same patient, a small variability in the values of the standardised indexes due to the electrodes position is to be expected. Future studies may be designed to assess the contribution to sEMG variability caused by other factors like for example tissue thickness and electrical conductivity or hormonal state.[5]

Conclusions

In the present study, the experimental tested hypothesis was confirmed. The position of the electrodes significantly affected the sEMG non-normalised potentials of the masticatory muscles during maximum teeth clenching, while ssEMG indexes demonstrated to be not influenced by the electrodes positioning. It can be estimated that 3% of the variability of ssEMG indexes is due to electrodes location. sEMG standardisation procedures proved to reduce the technical related instrumental evaluation variability, thus producing more reliable measurements.

List of abbreviations

sEMG, Surface Electromyography
RDC/TMD, Research Diagnostic Criteria for Temporomandibular Disorders
MVC, Maximal Voluntary Contraction
MM, Masseter
TA, Anterior Temporal

RMS, Root Mean Square
POC, Percentage Overlapping Coefficient
TC, Torque Coefficient
Asim, asymmetry index
Attiv, activity index
Impact, standardised activity index
MAD, Mean Absolute Difference
TEM, Technical Error Of Measurement
REM, Relative Error Of Measurement
SENIAM, European project on surface EMG for non-invasive assessment of muscles
ssEMG, standardised surface electromyography

Contributions

Conception and design, CD, DC, and RR; analysis and interpretation of data, DH, MV, and RR; drafting the article or revising it critically for important intellectual content, DH, GP, MV, and RR; final approval of the version to be published; RR, DC, and CD.

Conflicts of interest

The authors declare no conflict of interest.

Ethics approval

The study protocol was approved by the local ethic committee (Università degli Studi di Milano, code DG-EMG-2016).

Availability of data and materials

All data generated or analyzed during this study are included in this published article.

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