Evaluation of silent period on masticatory cycles of different muscles in dentate oral contraceptives users and nonusers

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ABSTRACT

Objective: The aim of this study was to evaluate the influence of oral contraceptive use on the silent period (SP) of anterior temporal and masseter muscles during the menstrual cycle. **Materials and Methods:** Totally, 28 women on reproductive age were selected including 15 nonusers of any hormone and 13 contraceptive users. All patients were dentate without muscular temporomandibular disorders. Electromyography (SP test) of the anterior temporal and masseter muscles was conducted every week during three consecutive menstrual cycles at 1st day of menstruation (P₁), 7th day (P₂), 14th day (P₃) and 21st day (P₄). **Results:** The SP values in the anterior temporal and masseter muscles were measured at both sides. The SP values of the right side (13.49 ms) at P₂ were significantly different compared to the left side (12.28 ms). However, there was no significant difference on the interactions among the three factors. **Conclusion:** It can be concluded that the SP values in healthy women in reproductive age may not be influenced by the menstrual cycle with similar results for both muscles.

Key words: Computerized dentistry, menstrual cycle, electromyography, silent period, sexual hormones

INTRODUCTION

Electromyography (EMG) allows evaluation of the masticatory system through record of muscular electrical current during resting, chewing and tooth clenching.^[1] The silent period (SP) represents a reduced electromyographic activity that induces voluntary muscular activity through magnetic transcranial stimulation in the primary motor cortex analyzed by EMG.^[2,3] In the elevator muscles of the mandible, the SP begins after functional dental contact or stimulus in the oral and periodontal region during muscular contraction.^[4] Thus, the SP can be induced by the release of sudden stress in the muscle, percussion on the chin, voluntary and rhythmic dental contact, and electrical stimulus.^[5,6]

The SP might be influenced by patient's age as well as deficient occlusion, tooth loss, and emotional behavior.^[7] Some studies have suggested that the SP is an important factor for evaluation of neuropathies and motor deficiency.^[8] The researchers demonstrated that the SP could be an auxiliary exam for diagnosis of functional disturbance as a correlation between period length and symptoms severity.^[9] However, only one long SP should not be related to muscular pain.

Several etiological factors may be related to symptom of temporomandibular dysfunctions.^[10,11] Some previous studies^[12,13] also presented the influence of sexual hormones on the development of joint and muscular alterations. The effect of estrogen on the masticatory system depends on the amount of

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estrogen receptors present in muscles and joints.^[12] In this sense, some studies showed a difference on pain modulation during the menstrual cycle^[14] and in contraceptive users.^[13] Although there is a study about the electrical performance of masticatory muscles during the menstrual cycle was found,^[15] it did not mention SPs of the muscles.

Thus, the aim of this study was to evaluate the SP on masticatory cycle of two elevator muscles of the mandible during the menstrual cycle of dentate female patients using oral contraceptive (OC) or not. The research hypothesis assumed that the SP on masticatory cycles would be different regarding muscles, OCs users and nonusers, and menstrual period.

METHODS

Sample

Amongst 342 female students of the Araçatuba Dental School-São Paulo State University-Brazil, 30 women were selected after initial exam including anamnesis and clinical evaluation. Half of the sample was not taking any sexual hormone (G1) while the other half (G2) was using some type of hormone. Two women of G2 did not return to the appointments, totalizing 28 patients. The inclusion criteria assumed female patients in reproductive age between 18 and 32 years old and also users and nonusers of some OC (estrogen and/or progesterone hormones) to avoid pregnancy, as confirmed by the volunteer's physician. The G1 patients should present regular menstrual cycles (28 ± 3 days) without taking OCs for at least 3 months. The G2 patients were taking low-dose pills for at least 3 months in a cycle of 21 days for active medication followed by 7 days without active medication. The exclusion criteria were signs and symptoms of temporomandibular disorders (TMD), as confirmed by the Dworkin and Leresche Research Diagnostic Criteria for TMD;^[16] presence of systemic pathology; pregnancy during the study; more than one tooth loss in each quadrant (excluding third molar); orthodontic treatment; presence of occlusal interference on the opposite side of mastication; symptoms on muscle palpation; and patients submitted to dental treatment including alteration of occlusion as well as prostheses and restorations.

The patients were informed about the experiment and signed an informed consent form in accordance with the recommendations of the Committee on Ethics in Human Research, which approved the study.

Menstrual cycles monitoring

All female students were observed every week during three consecutive menstrual cycles and the exams initiated on the 1st day of the cycle of each month. The menstrual cycle was divided into four distinct phases at 1st day of menstruation to 2 days after (P_1), 7th day ± 2 days (P_2), 14th day ± 2 days (P_3) and 21st day ± 2 days (P_4).^[17] The patients were instructed to avoid any analgesic medication before exams. Women who were not on OC group predicted their ovulation using the ClearPlan ovulation prediction kit (ClearPlan Easy, Unipath Research, Princeton, NJ, US). The patients received enough kits for the first cycle.

Electromyography measurements

The EMG was measured every week according to the menstrual cycle phases using bipolar surface electrodes (Bio Research, Redmond, WA, US) with 10 mm in diameter. A 21 mm interelectrode distance was determined and the electrodes were connected to an amplifier by cables that were attached to a computer using the Bio Research "Bio EMG" software to capture the electrical signal and analyze the muscular electrical activity. The test was conducted at sampling rate of 1000 Hz during 15 s using 1X amplification. The electrodes were positioned based on palpation of the muscles at 1.5–2.0 cm superior to the zygomatic arch, immediately behind the frontal process of the zygomatic bone.^[18] The evaluation was conducted bilaterally over dry skin parallel to the masseter and temporal muscle fibers. The center part of the masseter muscle was localized at the point equidistant to the upper and lower insertions of the muscle with teeth in occlusal contact. For the temporal muscle, the anterior border was located during mandibular movement to place the electrode perpendicular to the sagittal plane.

The SP values were obtained in open-close-clench-chewing cycles. This cycle is defined as a cycle when a subject firmly closes the mouth from the open mouth position with teeth in the habitual occlusal position during the chewing cycle. Masticatory performance was carried out using a raisin.^[19] Each subject chewed the food for 35 cycles, which is very close to the moment of deglutition, and each cycle corresponded to the maximum bite chewing performed for each subject to grind the food. Eight subsequent cycles of SP were recorded for each subject among the 35 cycles.^[20,21] SP measurements were obtained 3 times in each subject for each chewing cycle and the mean was calculated in milliseconds (ms). The SP length was measured using the double-zoom option with error about 1.2 ms. The measurement considered the peak of the last significant spike preceding the inhibition to the peak of the first significant spike being part of the ongoing muscle activity. In case of SP with two or three inhibitory pauses (double or triple SP) or depressed muscle activity after complete inhibition of motor neurons, the length of all inhibitory pauses was measured or added to the duration of a depressed activity after the last inhibitory pause.^[4] During the SP records, the subjects were kept in a comfortable sitting position without headrest and with the Frankfort horizontal plane parallel to the floor.

Statistical analyses

All statistical analyses were conducted using the SPSS version 19.0 statistical software (SPSS Inc., Chicago, USA). The normality of SP values was assessed by Shapiro-Wilks' W-test. A four-way repeated measures analysis of variance (ANOVA) was performed to verify any significant difference regarding muscle, side, group, and period. The means were compared by Tukey Honestly Significant Difference test (α =0.05) *post hoc* test.

RESULTS

Table 1 shows the means and standard deviations of the SP values for each period of analysis. All groups exhibited similar SP value over time [Table 1]. Table 2 shows the results of four-way repeated measures ANOVA. Although side and period statistically affected the SP values (P = 0.03, ANOVA) [Table 2], there was no statistically significant difference on the interaction among the factors (muscle × side × group × period) (P > 0.05, ANOVA) [Table 2]. For the interaction between side and period, the SP values of the right side (13.49 ms) were different from the left side (12.28 ms) at $P_{2'}$ regardless group and muscle (P < 0.05) [Table 3].

DISCUSSION

Electromyography has been used to evaluate muscular alterations considering that SP represents the functional state of the neuromuscular system. The research hypothesis was partially accepted assuming the difference in SP between side and period of analysis. Celebic *et al.*^[4] found mean SP values between 12.5 and 12.9 ms in healthy individuals during tooth clenching. McNamara *et al.*^[22] showed mean SP from 9.21 to 11.5 ms in healthy patients during dental percussion and chewing while Watt *et al.*^[23] reported higher SP during occlusion.

Chewing is a complex and rhythmic physiological process of mandible that requires interaction between receptors and nerves to control bone, joints and muscles of the stomatognathic system.^[21] In the present study, the SP was evaluated through tooth contact during chewing considering the three steps of the chewing cycle (open-close-clench). It has been suggested that masseter presents the highest strength and electrical activity during food grinding.^[24,25] On the other hand, the temporal muscle is effective for the most accurate mandibular movements, such as laterality.^[22,23] However, the present study did not find a significant difference between those muscles since the structural composition and functional characteristics may change as an adaptation to functional requirements.^[24] In this sense, some authors confirmed that strengthening and stretching exercises might cause alterations in muscular properties^[20,26,27] because of higher tolerance to stretching or biomolecular structural alterations that may change with gender.^[28]

It is important to emphasize that mastication may occur on both sides simultaneously or alternately, but it is important to consider that the majority of people chew more on one particular side that is, they have a preferred chewing side (PCS).^[29-31] This preference may be influenced by occlusion, pain, hemispheric

Group	Muscle	Side	Period				
			P ₁	P ₂	P ₃	P ₄	
1	Temporal	Right	12.84 (2.33)	13.36 (2.57)	12.81 (1.84)	12.86 (2.03)	
		Left	12.62 (1.99)	12.21 (1.42)	12.53 (1.91)	12.56 (1.96)	
	Masseter	Right	12.61 (2.19)	13.30 (2.45)	12.46 (1.87)	14.50 (6.41)	
		Left	12.46 (2.15)	12.13 (1.45)	13.12 (2.83)	12.70 (2.08)	
2	Temporal	Right	13.26 (3.12)	13.66 (2.64)	12.98 (3.29)	14.14 (3.96)	
		Left	13.19 (2.60)	12.54 (2.36)	13.06 (2.64)	13.11 (2.40)	
	Masseter	Right	13.27 (2.91)	13.70 (2.72)	12.83 (2.76)	14.19 (4.07)	
		Left	12.94 (2.28)	12.28 (2.31)	13.05 (2.60)	13.10 (2.45)	

SP: Silent period, SD: Standard deviation

Table 2: Four-way repeated measures ANOVA									
Source	df	SS	MS	F	Р				
Muscle	1	0.379	0.379	0.022	0.882				
Side	1	36.579	36.579	2.121	0.148				
Group	1	16.890	16.890	0.979	0.325				
Muscle×side	1	0.452	0.452	0.026	0.872				
Muscle×group	1	1.824	1.824	0.106	0.746				
Side×group	1	0.050	0.050	0.003	0.957				
Muscle×side×group	1	0.001	0.001	0.000	0.994				
Between subjects	104	1793.815	17.248						
Period	3	21.907	7.302	1.764	0.153				
Period×muscle	3	6.364	2.121	0.513	0.674				
Period×side	3	37.408	12.469	3.013	0.030*				
Period×group	3	1.584	0.528	0.128	0.944				
Period×muscle×side	3	6.056	2.019	0.488	0.691				
Period×muscle×group	3	3.756	1.252	0.302	0.824				
Period×side×group	3	0.046	0.015	0.004	1.000				
Period×muscle×side×group	3	5.137	1.712	0.414	0.743				
Within subjects	312	1291.306	4.139						

*P<0.05 represents statistically significant difference. ANOVA: Analysis of variance, SD: Standard deviation, df: Degrees of freedom, SS: Sums of squares, MS: Mean squares

 Table 3: Mean values (SDs) of SP (ms) for each side and period, regardless group and muscle

Side	Period						
	P ₁	P ₂	P ₃	P ₄			
Right	12.97 (2.58) ^A	13.49 (2.53) ^A	12.76 (2.41) ^A	13.91 (4.36) ^A			
Left	12.78 (2.21) ^A	12.28 (1.86) ^B	12.93 (2.45) ^A	12.85 (2.17) ^A			
Different letters denote statistically significant difference between the sides (P <0.05). SP: Silent period, SD: Standard deviation							

laterality and other factors.^[32,33] Chewing is an exercise for the muscles of mastication, and the preference for one side may stimulate this side more intensively. Although in the present study, preferred side of chewing of the voluntaries has not been determined, we could speculate that the difference between sides at P_2 could be probably due to a stimulus generated by periodontal or muscular receptors important for SP.^[34,35] It should be necessary more studies to investigate this subject.

Furthermore, we can suppose that the hormonal alterations at P_2 probably influenced the muscular activity during SP, the OCs did not affect the muscular function [Tables 2 and 3]. In addition, it is important to consider the direct influence of occlusion on the masticatory standard considering the alterations in muscular contraction during food grinding.^[36]

For the OCs, previous studies^[12,13,37] observed that hormonal alterations in women change muscular physiology. Although the level of sexual hormones reduces during the menstrual cycle,

especially estrogen,^[38] there was no significant difference between the group and the menstrual period [Table 2]. In addition, additional factors such as stress, structural alterations in muscles and joint, occlusion and parafunctional habits also influence the physiopathology of the stomatognathic system in female patients.

CONCLUSION

According to the results of the present study, it was concluded that the physiological, hormonal alterations during the menstrual cycle in OC users and nonusers do not affect the SP in healthy patients. There was no significant difference in the SP on masticatory cycle of muscles in contraceptive users and nonusers during the menstrual cycle of healthy patients.

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