43

Original article

Masseter muscle thickness and elasticity in periodontitis

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Abstract: This study aimed to compare the thickness and elasticity of the masseter muscle between patients with gingivitis and patients with periodontitis. A total of 124 patients (63 gingivitis, 61 chronic periodontitis) were recruited at the start of the study, but only 84 patients were declared as final participants. Patients were divided into two groups: (1) patients suffering from gingivitis and (2) those with generalized chronic periodontitis. Clinical (PI, plaque index; GI, gingival index; PD, probing depth; CAL, clinical attachment loss; and BOP, bleeding on probing scores) and ultrasonographic (thickness and elasticity of the masseter muscle) measurements of periodontitis were performed. There were no significant differences in gender, age, body mass index, education status, income level, or marital status between the two groups (P > 0.05). The mean age \pm SD for the gingivitis and periodontitis groups was 39.5 ± 10.8 years and 44.8 ± 8.8 years, respectively. There were significant differences between the two groups in the number of PI, GI, PD, CAL, and BOP scores. There were significant differences between the two groups when thickness of masseter during contraction and at rest was taken into account. The gingivitis group had significantly thicker masseter during both contraction and rest. On the other hand, when the elasticity of the masseter was evaluated, there were no significant differences found between the two groups and two sides for each group. Masseter muscle thickness in the periodontitis group was found to be decreased compared with that in the gingivitis group. Furthermore, loss of periodontal tissues due to periodontitis reduces the masticatory ability.

Keywords; periodontitis, public health, radiology

Introduction

Periodontitis is an inflammatory disease of the supportive tissues surrounding the teeth. It is characterized with gingival bleeding, pocket formation, periodontal attachment loss, tooth mobility, oral function loss, and, eventually, tooth loss. Chronic periodontitis is one of the most common chronic diseases and a health problem faced all around the world [1,2].

Loss of oral function due to periodontitis may affect occlusal balance and mastication [3]. Mastication is an indispensable oral function related to an individual's physical, social, and mental health. It is a complex process that involves a group of muscles, the masseter being one of which. Its structure and thickness are vital to oral functions [4,5].

Masseter muscle thickness can be determined through several imaging modalities, including magnetic resonance imaging, computed tomography, and ultrasonography, which provides a straightforward examination of the masseter muscle due to its superficial anatomical position [6]. Ultrasonography poses several advantages such as real-time imaging, easy applicability, low cost, non-invasiveness, and the use of no radiation [7]. It can also provide information about tissue stiffness with ultrasound elastography. Elasticity describes the quality and condition of a muscle. Moreover, elasticity provides knowledge about muscle contraction [8].

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An overview of the literature revealed that there are many previous studies evaluating the relationship between periodontitis and biting abilities [9,10]. However, as far as researchers know, no study has ever investigated the effect of thickness and elasticity of the masseter muscle on biting ability of patients with periodontitis. The aim of the present study was to evaluate thickness and elasticity of the masseter muscle with the aid or ultrasonography in patients with periodontitis.

Materials and Methods

Ethical consideration

Witten informed consent was obtained from each respondent at the point of study entry. The study was conducted in full accordance with the applicable ethical principles, including the Declaration of Helsinki in 1964 by the World Medical Association and later versions. This study was independently reviewed and approved by the Ethical Committee of the Faculty of Medicine, Erciyes University (meeting date 06.10.2016 decision number:2016/363).

Study population

A total of 124 patients (63 gingivitis, 61 chronic periodontitis) were recruited at the Department of Periodontology, Faculty of Dentistry, Erciyes University, Kayseri, Turkey. Thirty-six patients did not meet some of the inclusion criteria, and four patients refused to participate in the study. As a result, 84 patients made it to the final number of respondents in the study. Patients were divided into two groups: (1) patients with gingivitis and (2) those with generalized chronic periodontitis.

Case definition

Chronic periodontitis and gingivitis cases were diagnosed according to the 1999 classification developed by Armitage [11]. In addition, if more than 30% of sites are affected, the patient was diagnosed with generalized periodontitis.

- The exclusion criteria were as follows:
- Periodontal treatment within the last year
- Systemic disease
- Prosthesis (except fixed prosthesis installed at least 3 years prior)
- Self-reported psychiatric disorder
- Antibiotic use within the 6 months prior
- Use of nonsteroidal anti-inflammatory drug within 6 months prior
- Prescribed steroids
- · Immunosuppressive and psychiatric drugs
- Symptoms of acute dental problems
- Any apparent oral infection
- Aged >65 or <18 years
- Pregnancy or breastfeeding
- Smoking and alcohol consumption
- Temporomandibular joint disorder
- Bruxism
- Missing teeth which had not been restored with fixed prostheses for at least 3 years
- Unilateral chewing habit

Clinical periodontal examination

All participants were assessed both clinically and radiographically with regard to their oral health. To avoid bias and for consistency of results, the

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Fig. 1 Ultrasonographic thickness measurements in a transverse view of the masseter muscle. (a) Represents the maximum contraction. (b) Represents at rest position

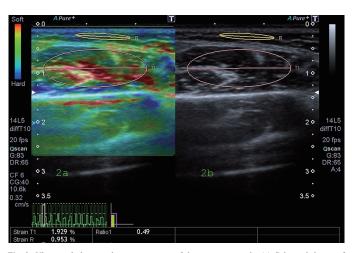


Fig. 2 Ultrasound elastography measurements of the masseter muscle. (a) Color-coded map of the strain elastography in the region of interest ranging from red to blue. Blue areas represent harder tissues, while red areas represent softer tissues. (b) The same region of interest in B-mode ultrasonography. Upper circle selects subcutaneous fat tissue as the reference area, and lower circle includes the masseter muscle region.

 Table 1
 Demographic values of the study groups

	Gingivitis	Periodontitis	P values
Age	39.5 ± 10.8	44.8 ± 8.8	0.121
Gender			
Male	12 (28.6%)	14 (33.4%)	0.221
Female	30 (71.4%)	28 (66.7%)	
Height (cm) (mean ± SD)	164 ± 6	160 ± 8	0.191
Weight (kg) (mean ± SD)	68 ± 10	71 ± 12	0.080
BMI (kg/m ²) (mean \pm SD)	25.4 ± 4.1	26.2 ± 5.2	0.178
Education level			
Primary education	10 (23.8%)	15 (35.7%)	
Secondary education	6 (14.3%)	6 (14.3%)	0.129
High school	15 (35.7%)	10 (23.8%)	
University	7 (16.7%)	7 (16.7%)	
Postgraduate/doctorate	4 (9.5%)	4 (9.5%)	
Income level			
<2,500 TL	17 (40.5%)	15 (35.7%)	0.369
>2,500 TL	25 (59.5%)	27 (64.3%)	
Marital status			
Married	25 (59.5%)	28 (66.7%)	0.489
Single	17 (40.5%)	14 (33.4%)	

TL, turkish lira (1 euro = 6.3 turkish liras) (t-test, chi-square test)

clinical examination was performed only by a single examiner (O.K). Full clinical periodontal measurements were recorded at six sites (mesiobuccal, mid-buccal, distobuccal, medio-lingual, mid-lingual, and distolingual) per tooth, which included the plaque index (PI) [12], gingival index (GI) [13], bleeding on probing (BOP), probing depth (PD), and clinical attachment loss (CAL). BOP percentage was calculated by dividing the bleeding sites by the total sites examined for each subject. CAL refers to the distance from the cemento-enamel junction in an apical direction to the base of the pocket/sulcus. A Williams probe (Hu-Freddy, Chicago, IL, USA) was used for the periodontal measurements. Additionally, the number of missing teeth and teeth with mobility (at each level) was also recorded. The mouth was evaluated separately (right mandibular and maxillary jaws and left mandibular and maxillary jaws).

In order to estimate the reliability of the measurements during the study period, six randomly selected patients were referred to the periodontology clinic for reevaluation. The PD and CAL scores were evaluated; intraexaminer reproducibility was >0.91 for both parameters.

Ultrasonography procedures

All ultrasound imaging and measurement procedures were conducted at Erciyes University, Faculty of Dentistry, Department of Oral and Maxillofacial Radiology. Measurements of muscle thickness were performed simultaneously for the right and left sides using the B-Mode ultrasonography and high frequency linear scanning ultrasound probe (7-18 Mhz, Aplio 500; Toshiba Medical Systems Corporation, Otawara, Japan).

To avoid artifacts and oblique views, the probe was positioned perpendicular to the skin surface. A water-based gel was applied to remove and help sound waves travel efficiently between the probe and the skin. The distance between the hyperechoic linear image of the mandible and muscle fascia was measured.

Measurements were obtained while the participants were positioned on the chair, with Frankfort's horizontal plane parallel to the floor. The measurements were taken by placing a transversely positioned transducer on the thickest part of the masseter muscle without applying pressure to the skin. Muscle thickness was measured on images of the masseter muscle at rest and at maximum contraction with the dual display feature of the device on the same screen (Fig. 1). By activating the elastography feature of the device, the transverse plane was subjected to compression and decompression of the muscles with the probe at about the same intensity and time intervals. Compression-decompression intensity and time interval were monitored as sinusoidal waves on the USG monitor. By selecting the images in the optimum range determined by the manufacturer, measurement areas were created within the muscle and reference tissue. The values measured in the designated areas were converted into ratios by the device (Fig. 2)

Reproducibility of the ultrasonographic measurements

All ultrasonographic measurements were performed by a calibrated examiner (M.O.) The reproducibility of the measurements was tested on 10 volunteers before the onset of the study. Ultrasonographic measurements were recorded with 1-week intervals. A paired-samples *t*-test revealed that no significant differences existed between the measurements (P = 0.632).

Table 2 Clinical evaluations of the study groups

	Gingivitis Perio			lontitis	Developer (m)		
	Right	Left	P values (\mho)	Right	Left	P values (O)	P values (Ψ)
Number of missing tooth	$0.57\pm0.6^{\rm a}$	$0.50\pm0.5^{\rm a}$	0.452	$0.67\pm0.6^{\rm b}$	$0.68\pm0.4^{\rm b}$	0.440	0.122
							0.092
PI overall (mean \pm SD)	0.79 ± 0.3	0.82 ± 0.3	0.511	1.72 ± 0.6	1.69 ± 0.7	0.578	< 0.001
PI posterior (mean ± SD)	0.83 ± 0.4	0.84 ± 0.4	0.723	1.92 ± 0.5	1.84 ± 0.6	0.684	< 0.001
P values (**)	0.494	0.856		0.357	0.398		
GI overall (mean ± SD)	0.71 ± 0.4	0.73 ± 0.3	0.634	1.76 ± 0.5	1.74 ± 0.6	0.674	< 0.001
GI posterior (mean ± SD)	0.81 ± 0.4	0.85 ± 0.4	0.701	1.84 ± 0.6	1.81 ± 0.6	0.314	< 0.001
P values (**)	0.178	0.341		0.368	0.269		
PD overall (mean ± SD)	$1.66 \pm 0.4^{\mathrm{a}}$	$1.60 \pm 0.3^{\mathrm{a}}$	0.251	$3.56\pm1.3^{\rm b}$	$3.31\pm0.8^{\rm b}$	0.112	< 0.001
PD posterior (mean ± SD)	1.69 ± 0.5	1.65 ± 0.4	0.221	$3.72 \pm .4$	3.52 ± 0.7		< 0.001
P values (**)	0.678	0.545		0.358	0.257		
CAL overall (mean ± SD)	0.57 ± 0.7	0.61 ± 0.7	0.602	4.82 ± 1.0	4.59 ± 1.1	0.220	< 0.001
CAL posterior (mean ± SD)	0.62 ± 0.8	0.66 ± 0.6	0.598	4.91 ± 0.9	4.72 ± 1.0	0.312	< 0.001
P values (**)	0.567	0.601		0.422	0.512		
BOP % overall (mean ± SD)	8.4 ± 3.3	7.3 ± 2.4	0.478	81 ± 11.2	78.4 ± 9.7	0.356	< 0.001
BOP % posterior (mean ± SD)	10.2 ± 2.9	10.3 ± 2.5	0.572	86 ± 9.8	84 ± 9.2	0.256	< 0.001
P values (**)	0.371	0.424		0.547	0.612		

One-way ANOVA (analysis of variance). Ψ , *P* values (Ψ) show gingivitis/periodontitis comparisons (upper *P* values for right, lower for left sides). \mho , *P* values (\mho) show right and left comparisons for healthy periodontium/periodontitis. **, *P* values show overall vs posterior comparisons.

Table 3 Comparison of the ultrasonographic measurements between study groups

		Ging	ivitis	Periodontitis			P values (Ψ)	
		Right	Left	P values (\mho)	Right	Left	P values (\mho)	
Thickness of the masseter	Contraction	14.3 ± 2	14.2 ± 1	0.278	11.9 ± 1	11.8 ± 1	0.643	< 0.001
								< 0.001
	Relax	13.9 ± 2	13.8 ± 2	0.345	11.9 ± 1	11.6 ± 1	0.119	0.001 <0.001
P values (‡)		< 0.001	0.001		0.674	0.128		
Elasticity of the masseter Contra Relax	Contraction	1.9 ± 2	1.9 ± 1	0.248	1.6 ± 1	1.7 ± 1	0.478	0.142 0.271
	Relax	1.5 ± 1	1.6 ± 1	0.377	1.2 ± 1	1.3 ± 1	0.443	0.241 0.227
P values (‡)		0.016	0.040		0.021	0.037		

One-way ANOVA (analysis of variance), t-test. ‡, P values (§) show contraction vs relaxation. Ψ , P values (Ψ) show gingivitis/periodontitis comparisons (first P values for contraction, second P values for relaxation). \Im , P values (\Im) show right and left comparisons for gingivitis/periodontitis contraction and relaxation.

Table 4 Relationship between parameters for the study population

Parameter 1	Parameter 2	P values	Correlation coefficient
Probing depth, right side of the jaws (mean)	Right-side masseter thickness (contraction)	< 0.001	-0.378
Probing depth, right side of the jaws posterior (mean)	Right-side masseter thickness (contraction)	< 0.001	-0.623
Probing depth, left side of the jaws (mean)	Left-side masseter thickness (contraction)	0.021	0.524
Probing depth, left side of the jaws posterior (mean)	Left-side masseter thickness (contraction)	0.001	-0.721
Probing depth, left side of the jaws anterior (mean)	Left-side masseter thickness (contraction)	0.040	-0.209

Pearson correlation

Statistical analysis

A pilot study involving varied samples (five periodontitis and five gingivitis) was used for sample size calculation. Preliminary statistical analysis revealed that ≥ 11 participants in each study groups would be necessary to provide 80% power to detect a difference at the 0.05 significance level.

The Kolmogorov-Smirnov test was used to assess the normality of the data. The Student's *t*-test was used for the two independent samples that follow a normal distribution. The categorical variable samples' intergroup comparisons were performed by the chi-square analysis with exact method. One-way ANOVA ("analysis of variance") was used to compare the means of two or more independent groups. For the post hoc comparisons, LSD correction was used. All analyzes were conducted using statistical software (IBM, SPSS Statistics 21, Chicago, IL, USA) with significance level set to 0.05.

Results

Demographic data for the two groups of participants are shown in Table 1. There were no significant differences between the two groups with respect to gender, age, body mass index, education, income level, and marital status (P > 0.05). The mean age \pm SD was 39.5 \pm 10.8 years and 44.8 \pm 8.8 years for gingivitis and periodontitis groups, respectively; 71.4% and 66.7% of the participants were female for the gingivitis and periodontitis group, respectively.

There were no significant differences between the two groups in terms of the number of missing tooth. Although there were no significant differences with the intra-group comparisons for the left/right sides for both groups, there were significant differences for the periodontal parameters (both right and left sides) when intergroup comparison was performed, including PI, GI, PD, CAL, and BOP scores (Table 2).

For the thickness of the masseter during contraction and at rest, there were significant differences between the two groups. The gingivitis group had significantly thicker masseter both during contraction and at rest. Additionally, in the gingivitis group, the masseter thickness during contraction was significantly higher than at rest. However, there were no significant differences between the right and left sides for either group. In addition, there were no significant differences between the two groups and two sides of each group when masseter muscle elasticity was measured. Contraction masseter thickness was significantly higher than at rest for both groups. Moreover, there were no significant differences for masseter elasticity level between each side for either group (Table 3).

Mean PD of the right and left sides was negatively correlated with right and left side masseter thickness, respectively. In addition, the mean PD of the posterior teeth on the right side was significantly correlated with the masseter thickness on the right side, and its correlation coefficient was higher. Similarly, mean PD of the posterior teeth on the left side was significantly correlated with left side masseter thickness, and its correlation coefficient was higher, too. Although the mean PD of the anterior teeth on the left side was significantly correlated with the left side masseter thickness, this correlation was lower than mean PD of the posterior teeth (Table 4). As the largest masticatory muscle, the masseter muscle has interested many researchers. Previous studies have evaluated the thickness of the masseter with regard to many disorders such as submucous fibrosis, facial myalgia, and bruxism [14-16]. From what researchers know, no study has been found in the literature which has investigated the relationship between chronic periodontitis and masseter thickness.

Changes in the stomatognathic system may affect the whole masticatory system including the masticator muscles. Morphologic and functional analysis of the masseter had been studied several times since it is one of the most essential components of the masticatory system. Several studies have investigated morphology [17,18], bite strength [21], electromyographic activity [19-21], and histomorphology of this muscle using various methods. It is well known that periodontitis causes mobility and loss of oral functions; therefore it could affect masticatory capacity, which can be measured with bite force and masseter muscle thickness [22]. In the present study, masseter muscles were evaluated in terms of thicknesss and elasticity between the gingivitis group and periodontitis group as an indicator of masticatory functions with the use of B-mode ultrasonography.

A few studies investigated the relationship between periodontitis and biting abilities [19,20,23]. Alkan et al. used pressure-sensitive sheets to evaluate biting abilities in their study [9]. They reported that bite force was significantly higher in subjects with healthy periodontium compared to patients with periodontitis. Morita et al. also used a similar method with pressure-sensitive sheets to measure their subjects' biting abilities [24]. They investigated associations of periodontal status and biting abilities in Chinese population and did not find a significant relationship between periodontal status and biting ability. Although they used similar methods, their findings were contradictory. In the present study, the masseter thickness was higher in the gingivitis group when compared to the periodontitis group, consistent with the findings of Alkan et al. [9].

This decrease in masseter muscle thickness in the periodontitis group could be attributed to alterations of masticatory ability. Masticatory forces, which are generated by the masticatory muscles, are controlled by the mechanoreceptors of the periodontal ligament and the alveolar bone [25]. Johansson et al. stated that loss of periodontal ligament due to periodontitis can affect the neural control of masticatory actions; therefore, it can result in reduced masticatory abilities [3]. These results are consistent with the present findings.

A previous study reported that chronic diseases contribute to the loss of muscle mass and decrease in muscle strength along with several factors including age, genetics, and diet [26]. Age and gender did not affect the results because these were well-balanced in study groups. Therefore, a decrease in masseter thicknesses in the periodontitis group cannot be associated with advancing age.

Some previous studies on masseter muscles of healthy volunteers did not report a statistically significant difference between the left and right sides for the elasticity of the muscle [27,28]. Thus, findings were consistent with the previous literature: there was no statistical difference between the right and left sides of patients in either group with regard to both thickness and elasticity of the masseter muscle.

Many different methods have been used to assess muscle elasticity, such as fine-needle electromyography, magnetic resonance elastography, and ultrasonographic elastography. Ultrasonographic elastography is known as a noninvasive, accessible, and real-time imaging method [8] and has been reported to be feasible and effective in evaluating the masseter muscle [18]. It provides a color-coded elasticity map of strain distribution within tissues. Sonographic elastography is generally divided into two groups according to type of tissue displacement. Strain elastography was the first developed technique for use in clinical practice. This can be performed with conventional ultasonography and softwares. Additionally, the elasticity of the masseter between the gingivitis group and periodontitis group was assessed with strain elastography.

In the study, there was no significant difference for elasticity between gingivitis group and periodontitis group. This could be explained by the use of strain-type elastography. Strain elastography requires a reference tissue to obtain a relative strain value. In the present study, subcutaneous fat tissue was considered as reference tissue to obtain strain ratio. Some disadvantages of strain-type sonoelastography have been reported such as operator dependency and relative evaluation [29].

Recent studies [29,30] have shown that shear wave elastography may be less operator dependent than strain elastography because it does not require "freehand" compression. Ariji et al. [28] compared shear wave elastography with strain elastography to evaluate reliabilities on measuring hardness. They concluded that shear wave elastography could be suitable for use in evaluating the masseter muscle. They also stated that use of strain elastography with reference tissue phantoms can provide data consistent with those measured using shear wave elastography. Therefore, lack of difference between the periodontitis group and gingivitis group in terms of masseter elasticity can be attributed to those limitations.

Borges et al. [31] evaluated masticatory performance of patients with chronic periodontitis in their study using a special device called biocapsule. They concluded that the loss of periodontal structures can negatively affect the masticatory function. Their results were consistent with the present findings. None of the studies mentioned previously evaluated the relationship between periodontitis and the masticatory capacity in terms of the masseter muscle thickness. In the study by Koc et al. [20], they mentioned that periodontal therapy improves masticatory functions. However, in this present study, the changes in masseter thickness and elasticity after periodontal therapy were not evaluated.

Factors believed to influence chewing ability indirectly were masseter muscle thickness, number of remaining teeth, and whether the remaining teeth comprise 20 or more natural teeth [32-35]. Some studies indicated that the number of posterior teeth is a key factor impacting this situation [33,35]. In the present study, all respondents had >20 natural teeth. In addition, the researchers wanted to evaluate and compare their condition in posterior teeth independently of the anterior teeth means and found results were no different from full-mouth clinical measurement means. This outcome may have originated from the fact that the periodontal conditions did not differ significantly between the posterior and anterior segments.

The present study has some limitations. First, operator dependency of strain elastography may affect the elasticity values. Second, more precise inclusion criteria could be defined for age, sex, and body mass index because all these factors may indirectly affect structure of the masseter muscle. As many clinicians would appreciate, it is quite difficult to create such a study population. However, there were no significant differences between study groups for demographic parameters. Thus, this limitation can be remissible. Third, although the periodontitis group consisted of generalized chronic periodontitis, this study could be performed in individuals with worse clinical periodontal statuses. However, this was the only population that could be studied due to the very precise inclusion criteria. Finally, patients with all teeth in the mouth could be included, so that impact of fixed prosthesis, if there is, could be prevented.

Within the limitation of the present study, it can be concluded that thickness of the masseter muscle in the periodontitis group was found lower compared to the gingivitis group. This might be due to loss of oral functions and chewing abilities of patients with periodontitis. In addition, results suggest that loss of periodontal support tissues due to periodontitis reduces masticatory ability. No significant difference was found between the gingivitis group and periodontitis group for elasticity of the masseter. Different elastography modalities can be used to evaluate morphology of the masseter. Further studies that evaluate changes in masseter muscles after periodontal therapy should be conducted in order to understand the possible key role of periodontitis in masticatory capacity. There is also a need for further studies with a more defined inclusion and exclusion criteria and larger study population.

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Conflict of interest

The authors declare that there is no conflict of interest concerning the contents of the study.

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