



Original Article

## Quantitative and qualitative evaluation of the masseter muscle by ultrasonography and correlation with whole body health status

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**Abstract.** [Purpose] Ultrasonography can be used to non-invasively analyze any cross-section of the human body and to measure tissue elasticity, thickness, and brightness. This study was performed to examine the quantitative and qualitative changes in the masseter muscle at rest and at maximal occlusion, and to evaluate the relationship between these changes and the general health of the individual. [Participants and Methods] The study cohort comprised 30 healthy adults. Correlations between basic participant information (sex, age, height, body weight, body mass index, body fat, maximum bite force, handgrip strength, and tongue pressure) and masseter muscle ultrasonographic data were examined. [Results] Masseter muscle thickness was significantly greater in males than in females. Body weight and body mass index correlated positively with masseter muscle thickness. Body mass index and body fat percentage correlated positively with masseter muscle brightness. Tongue pressure correlated positively with handgrip strength. [Conclusion] Our analyses of muscle thickness and brightness suggest that ultrasonography may be useful in evaluating masseter muscle quantity and quality, and that the condition of the masseter muscle may correlate with the overall health status of the individual.

**Key words:** Ultrasonography, Masseter muscle, Muscle thickness

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### INTRODUCTION

It is well known that the morphogenesis of the maxillofacial cranium is associated with changes in the development and function of the surrounding soft tissue system<sup>1)</sup>. Furthermore, the masseter size is strongly related to the vertical maxillofacial dimensions<sup>2)</sup>, and the masseter muscle thickness is closely connected to the mandibular morphology<sup>3)</sup>. In 2002, Ogata et al. evaluated the quality of the masseter muscle and described the relationship between the action potential conduction velocity of the masseter and its vertical maxillofacial morphology<sup>4)</sup>. Since then, many studies have evaluated the correlation between the masticatory muscles, including the masseter, and maxillofacial morphology<sup>2-8)</sup>.

It is important to evaluate oral function to prevent oral frail, which has recently gained attention in the dental field. The strength of the masticatory muscles is one of the diagnostic criteria for oral dysfunction, and is related to the general muscular condition of each individual. In the field of physical therapy, skeletal muscle has been evaluated by various methods for a long time. Among these methods, ultrasonography is widely used for skeletal muscle analysis because it is non-invasive and convenient<sup>9-12)</sup>. In the maxillofacial region, many studies have reported the use of ultrasonographic devices to evaluate the masticatory muscles<sup>13-17)</sup>, but the methods and results have not yet been standardized<sup>15-17)</sup>. Furthermore, studies have

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reported that among the masticatory muscles, the masseter has the highest correlation with tooth presence and general muscular fitness<sup>18</sup>). We hypothesized that the state of the masseter muscle is related to the individual maximum occlusal force and whole-body muscle strength. We also hypothesized that the elasticity of the masseter would be increased during contraction compared with at rest.

The purpose of this study was to examine the quantitative and qualitative changes in the masseter muscle between rest and maximum occlusion using ultrasonography, and to evaluate the relationship between these changes and the general condition of individuals.

## PARTICIPANTS AND METHODS

The study cohort comprised 30 dentists (15 males and 15 females; average age  $28.0 \pm 2.3$  years, weight  $61.0 \pm 15.8$  kg, height  $167.0 \pm 9.5$  cm) working at the Department of Orthodontics, Showa University Dental Hospital. We excluded individuals undergoing orthodontic treatment, pregnant females, and those with congenital maxillofacial diseases. The details of the study were provided both verbally and in writing, and informed consent was obtained from all participants. This study was approved by the Research Ethics Review Board of Showa University (approval number: 22-212-A).

We collected baseline and masseter muscle data from each participant. Baseline data comprised gender, age, height, weight, body mass index (BMI), body fat, maximum bite force, handgrip strength, and tongue pressure<sup>19</sup>). Body composition, including height, weight, BMI, and body fat, was measured using a weight scale (RD-803L; Tanita Corporation, Tokyo, Japan). Maximum bite force was measured using an occlusal force-measuring device with a capacitive pressure-mapping sensor (OFMD-CPS; Yoshida, Tokyo, Japan). An ultrasound (US) system (ARIETTA 850SE; Fujifilm Healthcare Ltd., Tokyo, Japan) was used to measure the masseter muscle thickness and elasticity at rest and at maximum occlusion, and to measure the masseter muscle brightness at rest.

The bite force was measured while the participant was in a relaxed sitting position. We inserted a U-shaped sensor into the participant's mouth, pressed a measuring button, and then asked the participant to perform maximum clenching in the intercuspal position for 3 seconds. The measured bite force was immediately displayed on the monitor and the maximum values were recorded. Measurements were taken twice with a 10-second rest period between measurements. The average of the two measurements was taken as the maximum occlusal force for each participant.

Handgrip strength was measured using a digital hand dynamometer (Jamar Plus+ Digital Hand Dynamometer; Performance Health International Ltd., Nottingham, UK) with the participant in a seated position with the forearms extended straight forward. The average of two measurements of the dominant hand was used as the handgrip strength for each participant.

A tongue pressure manometer (TPM-02E; JMS Co. Ltd., Hiroshima, Japan) was used to measure the maximum tongue pressure while the participants were in a relaxed sitting position. The participants were asked to place a balloon on the anterior part of their palate and to close their lips by biting on a hard ring with their upper and lower incisors. They were then asked to lift their tongue and press the balloon against the palate with maximum voluntary muscle effort for approximately 7 seconds. Measurements were taken twice with a rest period of approximately 30 seconds between measurements, and the average of the two measurements was used as the tongue pressure for each participant.

The thickness of the masseter muscle was measured by B-mode US. To provide a good acoustic transition environment, ultrasonic measurement jelly (Logiclean; GE Healthcare Pharma Ltd., Tokyo, Japan) was applied to the tip of the 10-MHz linear probe (L64 K; Fujifilm Healthcare Ltd.) during the measurement. Measurements were taken while the participants were sitting with the Frankfurt plane parallel to the floor. The probe was placed perpendicular to the masseter muscle and applied to the skin without pressure. The same examiner made three resting measurements, followed by three maximum clenching measurements on both sides. Maximum occlusion was confirmed using the OFMD-CPS. The masseter muscle thickness was defined as the length of the widest part in the area surrounded by the outer and inner fascia.

The elasticity of the masseter muscle was measured using the Shear-Wave Measurement US mode, which quantitatively evaluates the tissue elasticity based on the shear wave velocity ( $V_s$  in m/s) generated *in vivo*; the higher the modulus of elasticity of an object, the faster the  $V_s$ . Data were collected when each reliability index (i.e., the percentage of the net effective  $V_s$ ) was equivalent. Three such measurements were taken, and the average of the three measurements was used as the measured value. The elasticity was measured bilaterally at rest and at maximum occlusion, similarly to the thickness. When measuring the muscle elasticity at maximum occlusion, the measurement site was positioned while the examiner reviewed the B-mode US image, and the participant was instructed to maintain the maximum occlusion state for at least 3 seconds.

The Shapiro–Wilk test was used to confirm the normality of the masseter muscle data (muscle thickness at rest, muscle thickness at maximum occlusion, muscle stiffness at rest, muscle stiffness at maximum occlusion). The intraclass correlation coefficient (ICC) of three repeated measurements was calculated. Correlation analysis and Pearson's correlation coefficient were used for statistical validation between each baseline value (gender, age, height, body weight, BMI, body fat, maximum bite force, hand strength, and tongue pressure) and each masseter muscle measurement. The level of significance was set at  $\alpha=0.05$  (two-tailed), and  $p<0.05$  was considered statistically significant. All data were analyzed using SPSS Statistics 26 (IBM Corporation, Armonk, NY, USA).

## RESULTS

The demographic characteristics of the participants are shown in Table 1.

The ICCs showed moderate agreement for the measurements of resting muscle elasticity (right side), resting muscle elasticity (left side), and maximum occlusal muscle elasticity (left side), and showed substantial or almost perfect agreement for all other measurements<sup>20</sup>). Overall, the ICC values indicated that the measurements were reliable.

Correlation analysis was performed to evaluate the relationship between US-measured data and baseline data. The unpaired t-test was used to compare each US-measured value between genders, and showed that the masseter muscle thickness was significantly greater in males than females at rest and maximum occlusion (Table 2). Masseter muscle thickness was positively correlated with body weight and BMI. In addition, the masseter muscle thickness measurements at rest and during contraction was positively correlated with the handgrip strength and tongue pressure, except for the left-sided contracted muscle thickness. The masseter muscle brightness was significantly positively correlated with both the BMI and body fat (Table 3).

## DISCUSSION

This study quantitatively and qualitatively evaluated the masseter muscles in a healthy adult population with an average age of  $28.0 \pm 2.3$  years. Although US is a promising method for evaluating the masticatory muscles, it has been difficult to establish a protocol for objective evaluation due to differences in equipment, participant selection, and units of measurement. Many reports have emphasized the need for standardized US procedures to determine its accuracy<sup>15-17</sup>). The reported variations include differences in participant posture at the time of measurement, measurement equipment, and measurement area<sup>16</sup>). A previous study that measured the masseter in sections found no significant differences between the left and right

**Table 1.** Baseline information

Variable	Data
Gender, males/females	15/15
Age (years)	$28 \pm 2.3$
Height (cm)	$167.1 \pm 9.6$
Weight (kg)	$61.0 \pm 15.9$
Body mass index ( $\text{kg}/\text{m}^2$ )	$21.7 \pm 3.8$
Body fat (%)	$23.9 \pm 7.5$
Bite force (N)	$682.8 \pm 55.9$
Handgrip strength (kg)*	$34.4 \pm 10.7$
Tongue pressure (kPa)	$40.1 \pm 8.9$

Data are presented as mean  $\pm$  standard deviation.

\*Measured with the dominant hand (27 right, 3 left).

**Table 2.** Comparisons between males and females

	Males n=15	Females n=15	Unpaired t-test p-value
Right RMT (mm)	$13.9 \pm 2.2$	$11.5 \pm 1.4$	**
Right CMT (mm)	$15.5 \pm 2.1$	$12.8 \pm 1.6$	**
Left RMT (mm)	$14.2 \pm 2.3$	$11.7 \pm 1.4$	**
Left CMT (mm)	$15.6 \pm 2.3$	$13.1 \pm 1.8$	**
Right RME (kPa)	$4.6 \pm 1.2$	$4.1 \pm 1.2$	
Right CME (kPa)	$21.3 \pm 9.5$	$16.5 \pm 7.8$	
Left RME (kPa)	$5.2 \pm 2.4$	$5.1 \pm 2.2$	
Left CME (kPa)	$25.2 \pm 13.7$	$18.8 \pm 4.9$	
Right MB	$34.6 \pm 7.4$	$39.5 \pm 12.3$	
Left MB	$40.1 \pm 11.7$	$44.4 \pm 12.9$	

RMT: rested muscle thickness; CMT: contracted muscle thickness; RME: rested muscle elasticity; CME: contracted muscle elasticity; MB: muscle brightness.

\* $p < 0.05$ , \*\* $p < 0.01$  (n=30).

**Table 3.** Correlations between masseter muscle data and basic participant information

	Age	Height	Weight	Body mass index	Body fat	Bite force	Handgrip strength	Tongue pressure
Right RMT (mm)	-0.075	0.461*	0.735**	0.728**	0.091	0.183	0.562**	0.554**
Right CMT (mm)	-0.039	0.496**	0.712**	0.676**	0.015	0.177	0.645**	0.517**
Left RMT (mm)	-0.120	0.374*	0.684**	0.702**	0.010	0.277	0.481**	0.507**
Left CMT (mm)	-0.165	0.309	0.544**	0.560**	-0.083	0.355	0.338	0.305
Right RME (kPa)	0.166	0.475**	0.252	0.134	0.016	-0.321	0.117	-0.107
Right CME (kPa)	0.164	0.165	0.227	0.191	-0.181	-0.060	0.300	0.406*
Left RME (kPa)	0.017	-0.060	0.000	0.021	-0.051	0.305	-0.108	-0.016
Left CME (kPa)	0.202	0.297	0.232	0.139	-0.152	-0.013	0.244	0.259
Right MB	0.348	-0.176	0.166	0.367*	0.576**	0.028	-0.158	0.061
Left MB	-0.009	-0.006	0.312	0.433*	0.532**	-0.183	0.084	0.095

Pearson's correlation coefficient; \* $p < 0.05$ , \*\* $p < 0.01$  ( $n = 30$ ).

RMT: rested muscle thickness; CMT: contracted muscle thickness; RME: rested muscle elasticity; CME: contracted muscle elasticity; MB: muscle brightness.

sides or between genders<sup>17</sup>). In the present study, we used the methods of Strini et al.<sup>21</sup>, and all masseter muscles were measured by one examiner. The contact pressure of the probe was carefully controlled during the measurement. As the displayed image easily changed depending on the hand pressure, no pressure was applied while performing the measurements. When measuring the masseter muscle thickness during contraction, the OFMD-CPS was used to reproduce the maximum occlusion state. We considered that the measurement conditions were unified because the occlusal force was measured using a sensor sheet. In addition, participants with a strong pharyngeal reflex were handled with great caution.

The masseter muscle thickness significantly differed between genders and was significantly positively correlated with the body weight and BMI. These results were similar to a previous report that indicated that age and BMI may be associated with increased masticatory muscle thickness and that muscle thickness tends to be thinner in females than males<sup>13</sup>). In addition, the masseter muscle thickness was positively correlated with the handgrip strength and tongue pressure, indicating a relationship between the muscles of the oral cavity and the muscles of the whole body. In the future, we would like to use this method to evaluate the thickness of the masseter muscle in patients with jaw deformities such as facial asymmetry and congenital diseases. There was no correlation between the masseter muscle thickness and age in the present study, which we considered to be due to the small age range in the study population. In the future, we will evaluate the changes in the masseter muscle thickness with age by performing measurements in children and the elderly.

The tissue elasticity has been evaluated based on the Vs generated in the living body. Many studies have already evaluated the hardness of the masseter and temporal muscles using ultrasonography<sup>16</sup>). Previous studies have reported that the Vs is significantly higher in patients with arthrosis of temporomandibular joint than in a healthy population, and that the masseter elasticity is positively correlated with the characteristic pain intensity and negatively correlated with the maximum mouth opening and painless mouth opening<sup>22</sup>).

We hypothesized that the elasticity of the masseter would be increased during contraction compared with at rest, as the Vs and tissue stiffness were positively correlated; however, no significant correlation was found. Taking hepatitis as an example, the factors that affect tissue hardness are liver fibrosis, inflammation, and jaundice<sup>23</sup>), all of which can be said to be processes of internal degeneration. As the fibrosis progresses and the inflammation intensifies, the Vs becomes faster. However, as the present study evaluated healthy adults and did not assess individuals with tissue degeneration, we did not consider it appropriate to assess the muscle quality based on these results. Additionally, to measure the elasticity, unlike measuring the muscle thickness, the participants needed to maintain the clenched state for 2–3 seconds; therefore, the measurement results might have been influenced by fatigue. Furthermore, any movement of the probe during the measurement may reduce the effectiveness of the measurement and cause data variability, highlighting the need for skilled and consistent measurement techniques.

Muscle brightness represents non-contractile tissues such as fat and fibrous tissues within the muscle and enables the assessment of muscle quality. A previous study evaluating the quadriceps femoris muscle in older women found that both muscle thickness and muscle brightness are significantly correlated with muscle strength, and that muscle brightness is a measure of intramuscular fat<sup>24</sup>). Additionally, muscle brightness is not correlated with the BMI, body fat percentage, or subcutaneous fat thickness, indicating that individual qualitative changes in muscles cannot be predicted from obesity indicators<sup>24</sup>). Moreover, different muscle parts have different characteristics, and in healthy young individuals, quantitative indicators are more useful than qualitative indicators for evaluating knee extension muscle strength<sup>25</sup>). While there are some limited reports on limb muscle brightness, no studies have evaluated the brightness of the masseter muscle in young adults.

In the present study, we performed the brightness analysis using B-mode US images at rest. The results showed a positive correlation between the masseter brightness and the BMI and body fat. However, as the present study evaluated young adults with a narrow age range, there were no significant differences in muscle brightness in accordance with age or gender. In the future, these measurements need to be made in children and the elderly to determine whether muscle brightness evaluations could contribute to the evaluation of age-related changes in muscle quality.

Our hypothesis regarding the muscle elasticity was not fully proven. This may be because the method may not have been suitable and it was difficult to analyze intramuscular changes. In addition, contrary to our prediction, no correlation was found between the maximum bite force and masseter muscle data in this study. In the future, we would like to further explore this potential relationship by performing a skeletal evaluation using cephalometric radiographs of each individual.

In conclusion, this study indicated that there was a relationship between the muscles of the oral cavity and the muscles of the whole body; as the body weight and BMI increased, the masseter muscle became thicker. In addition, the masseter muscle brightness was positively correlated with the BMI and body fat percentage, suggesting that it may be possible to qualitatively evaluate the masseter muscle using ultrasonography. We believe that a method for qualitatively evaluating the masticatory muscles using ultrasonography will be established and may help in the evaluation of myofunctional therapy and the diagnosis of oral dysfunction in clinical practice.

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### *Conflict of interest*

None.

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