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Title

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A concise and informative title

Age-related changes in swallowing muscles

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Key summary points

Aim: To measure age-related changes in swallowing muscles by ultrasonography as a non-invasive method.

Findings: By ultrasonography, we found that old people had a smaller geniohyoid muscle area and greater geniohyoid muscle brightness than young people. Our analyses indicated that age and whole-body skeletal muscle mass were associated with mass and quality of the geniohyoid muscle.

Message: Ultrasonography effectively identified the association between atrophy of the swallowing muscles and aging and loss of whole-body skeletal muscle mass.

Abstract

Purpose: Sarcopenia of swallowing muscles is a potential cause of dysphagia. We investigated age-related changes in mass and quality of swallowing muscles by ultrasonography as a non-invasive and convenient examination in subjects without dysphagia.

Methods: A total of 104 subjects (34 males, 70 females) participated in this study. Age, physical status, and mass and strength of skeletal and swallowing muscles were investigated. Ultrasonography was performed to measure cross-sectional area and brightness of the geniohyoid muscle as a swallowing muscle. Calf circumference was measured to evaluate skeletal muscle mass. Hand grip strength was measured to evaluate skeletal muscle strength. Subjects were divided into two groups: young (< 65 years old) and old (≥ 65 years old). We performed univariate and multivariate analyses to analyze the differences between the groups.

Results: The number of subjects in the young group was 35, and 69 in the old group. The mean ± SD of measurements in each group was as follows (young / old): age, 35.4 ± 13.9 / 74.5 ± 5.5 years old; calf circumference, 37.4 ± 4.1 / 33.9 ± 2.7 cm ($p < 0.001$); hand grip strength, 35.6 ± 10.2 / 25.8 ± 7.6 kg ($p < 0.001$); cross-sectional area of geniohyoid muscle, 229.5 ± 52.2 / 174.1 ± 40.7 mm^2 ($p < 0.001$); and brightness of geniohyoid muscle, 46.6 ± 11.1 / 59.6 ± 10.8 ($p < 0.001$). The old group had a significantly smaller geniohyoid muscle area and significantly greater geniohyoid muscle brightness than the young group ($p < 0.01$). Age and calf circumference were independent explanatory factors for geniohyoid muscle area ($p < 0.01$). Age and sex were independent explanatory factors for geniohyoid muscle brightness ($p < 0.01$).

Conclusions: Ultrasonography revealed a smaller area and greater brightness, which suggested smaller mass and greater infiltration of fat, in the geniohyoid muscle in old people than in young people.

Key words

Swallowing, Muscle, Ultrasonography, Sarcopenia

Main text

Background and Purpose

Sarcopenia increases physical dysfunction [1, 2]. Previous research reported that sarcopenia might cause dysphagia [3-7]. Sarcopenia and dysphagia are important problems among old people; particularly, the prevalence of sarcopenic dysphagia was reported to be 32% in rehabilitation patients in Japan, a rapidly aging society [8]. Sarcopenic dysphagia is defined as a swallowing disorder due to sarcopenia of whole-body skeletal and swallowing muscles [3, 9].

As sarcopenia can be treated [1, 2], confirming the status of muscles is important to prevent muscle dysfunction. Feng et al. and Molfenter et al. reported that old people had lower geniohyoid and pharyngeal muscle mass than young people when evaluated by CT or MRI, respectively [10, 11]. Compared with CT and MRI, ultrasonography is characterized by being radiation-free, portable, easy to use, and having a low cost, and short examination time, thus, making it useful for screening many people in order to confirm their muscle status. Several reports evaluated swallowing muscle mass and brightness, later as a measure of muscle quality, by ultrasonography [12-19]. Muscle brightness suggests infiltration of fat [20]. However, age related changes of these parameters measured by ultrasonography are unclear.

In this study, we measured age-related changes in swallowing muscle mass and quality by ultrasonography. We investigated the cross-sectional area and brightness of the geniohyoid muscle as a representative swallowing muscle and compared between young (< 65 years old) and old (≥ 65 years old) people.

Methods

Participants and parameters

The study design was cross-sectional. Subjects were divided into two groups: young (< 65 years old) and old (≥ 65 years old). Old subjects were recruited from three preventative care services and one regional preventative care event for older citizens, and young subjects were recruited from a medical facility. Inclusion criterions were independently living subjects without severe illness that affected their physical function. Subjects with dysphagia were excluded using Food intake LEVEL scale [21]. The study period was between Dec. 2017 and Feb. 2019. The ethical committees of Southern Tohoku General Hospital and Tohoku University approved this study. All patients were given documents and provided written informed consent prior to enrollment.

We assessed parameters concerning both skeletal and swallowing muscles. Age, sex, height, body weight, body mass index (BMI), calf circumference, hand grip strength, and Mini Nutritional Assessment® short form (MNA-SF)

[22] score for nutritional status were assessed. The thickest part of the lower leg was assessed using a measure as a calf circumference. Calf circumference was regarded as an indicator of whole-body skeletal muscle mass [23]. The hand grip strength was measured using a Smedley hand dynamometer in the standing position. Hand grip strength was used as an indicator of whole-body muscle strength [1, 2]. The maximum strength was taken as a result. MNA-SF was used because nutritional status and dysphagia had a clear association [24]; it is a screening questionnaire to assess nutritional status consisting of six questions. A total score of 12-14 points indicates a healthy condition, 8-11 means risk, and 0-7 malnutrition.

For swallowing muscles, Food Intake LEVEL Scale (FILS) was measured to evaluate swallowing function. Maximum tongue pressure was measured to evaluate swallowing muscle strength. And, ultrasonography was performed. FILS is a validated ranking scale for evaluating swallowing function [20]. A score of 10 indicates normal function, 7-9 indicates mild dysphagia, 4-6 indicates moderate dysphagia and 1-3 indicates severe dysphagia. In this study, we considered subjects with a FILS score of 10 to have normal swallowing function, and those with FILS score 9 and under, to have dysphagia. Maximum tongue pressure was assumed to demonstrate swallowing muscle strength [25]. A tongue pressure measurement instrument (JMS, Hiroshima, Japan) was used to measure tongue pressure [26]. Subjects were instructed to press a balloon to their palate folds with their tongues. Tongue pressure measurements were performed three times for each subject, and the maximum score was taken as a result. The cross-sectional area of the geniohyoid muscle was used as an indicator of swallowing muscle mass. Brightness of the geniohyoid muscle was used as an indicator of swallowing muscle quality because high intensity in the muscle indicates the presence of fat [20].

Ultrasonography

We used a LOGIQ e V2 (GE Health Care Japan, Tokyo, Japan) and convex-array probe (Model No. is 4C-RS) for ultrasonography. We created a parameter setting button on the screen of the ultrasound machine for this study, labeled as “swallow” mode. In the swallow mode, the parameters were set as follows: gain, 60; dynamic range, 78; frequency, 4.0 MHz; depth, 7 cm. Before every measurement, the tester set the ultrasound machine to “swallow” mode. The parameters were recorded with each ultrasound image simultaneously, and we checked the status of parameters in the image analyses. Subjects were instructed to sit on a chair, face forward, keep a neutral position, and close the mouth. We instructed the subjects not to move or swallow while the US pictures were being taken. A probe was set under the jaw in the central sagittal plane. Ultrasonography measurements were performed three times, and the mean score was taken as a result. The area and brightness of the geniohyoid muscles in the sagittal plane were calculated using image J (Fig. 1, All images belong to author T.M.). An area of interest was set in the manual polygon mode. Brightness was assessed by the mean echo level of the area of interest, and the range was 0-255. Ultrasonography and analysis were performed by one speech therapist (T.M.) to reduce any potential bias caused by different testers.

Statistical analysis

The means of age, height, weight, BMI, calf circumference, hand grip strength, MNA-SF, maximum tongue pressure, and the cross-sectional area and brightness of the geniohyoid muscle were calculated. The median FILS scores were also calculated. We evaluated the relationships among the parameters using Pearson's product moment correlation coefficient. The subjects were divided into two groups: young (< 65 years old) and old (≥ 65 years old). Univariate analyses were performed to investigate differences between the two groups using the Student's t-test. Conditions for sample size calculation were set as follows: α error, 0.05; power, 0.8. The required sample size to detect differences in area and brightness of the geniohyoid muscle between the young and old groups was 11 in the young group and 22 in the old group. In multiple regression analysis, age, sex, height and calf circumference were chosen as explanatory variables for the area and brightness of the geniohyoid muscle because these parameters can affect skeletal muscle mass. All analyses were performed using EZR software [27, 28], p - values < 0.05 were considered significant.

Results

A total of 104 subjects participated in this study, 34 males and 70 females. There were 35 subjects (18 males and 17 females) in the young group and 69 subjects (16 males and 53 females) in the old group. Table 1 shows significant differences between young and old groups in age, height, weight, calf circumference, hand grip strength, maximum tongue pressure, and geniohyoid muscle area and brightness. Table 2 shows differences between young and old groups in each gender. There were significant differences in area and brightness of the geniohyoid muscles between young and old subgroups of both the male and female groups. The old group had smaller area and greater brightness of the geniohyoid muscle (Figs. 2 and 3). The area of the geniohyoid muscle was correlated with height, calf circumference, hand grip strength, and maximum tongue pressure, and was negatively correlated with age and brightness of the geniohyoid muscle (Table 3). The brightness of the geniohyoid muscle was correlated with age, and negatively correlated with height, calf circumference, hand grip strength and area of geniohyoid muscle (Table 3).

Tables 4 and 5 show the results of the multivariate regression models for the area and brightness of the geniohyoid muscle. Age and calf circumference were independent explanatory factors for a small geniohyoid muscle area. Age and sex were independent explanatory factors for great brightness of the geniohyoid muscle.

Discussions

In this study, the area of the geniohyoid muscle was associated with calf circumference, grip strength, and maximum tongue pressure. This muscle moves the hyoid bone and elevates the larynx in the swallowing motion [29]. Loss of geniohyoid muscle mass suggests atrophy of swallowing muscles. Calf circumference is an indicator of whole-body skeletal muscle mass [22] and grip strength is an indicator of whole-body skeletal muscle strength [1, 2]. Maximum tongue pressure is an indicator of swallowing muscle strength [24]. In the univariate analysis, the area of the geniohyoid muscle was correlated with calf circumference (r = 0.58, $p < 0.01$), hand grip strength (r = 0.68, $p < 0.01$), and maximum tongue pressure (r = 0.39, $p < 0.01$). These results indicated that the area of the geniohyoid muscle, as a representative of swallowing muscle mass, was associated with whole-body skeletal muscle mass, and whole-body and swallowing muscle strength. These results are consistent with those of previous research. Yoshimi et al. reported that geniohyoid muscle area in the coronal plane correlated with trunk muscle mass index and tongue pressure [30]. Tamura et al. reported that tongue thickness in the coronal plane correlated with arm muscle area [14]. Baba et al. reported that geniohyoid muscle area in the coronal plane correlated with maximum tongue pressure [17].

Our data suggested that the area of the swallowing muscle decreased with aging. Old people had a smaller geniohyoid muscle area and greater brightness than young people. The sample size to detect a significant difference between the two groups in this study was sufficient (Calculated size was 33, and the actual size was 104). These results indicated that swallowing muscle mass and quality decreased with age and whole-body muscle mass. In the multivariate analyses, age ($p < 0.01$) and calf circumference ($p < 0.01$) were independent expl1anatory factors for loss of geniohyoid muscle mass. These results indicated that swallowing muscle mass was affected by age and whole-body skeletal muscle mass. Age (p < 0.01) and sex (p < 0.01) were independent risk factors for the low quality of the geniohyoid muscles. These results are consistent with those of previous research. Feng et al. used CT and reported that old people had a smaller geniohyoid muscle area in the sagittal plane than young people [10]. Molfenter et al. used MRI and reported that old people had a smaller pharyngeal muscle area in the horizonal plane than young people [11]. Baba et al. used ultrasonography and reported that geniohyoid muscle area in the coronal plane correlated with age [17]. As far as we know, there has been no previous research showing age-related changes in brightness of the geniohyoid muscle by ultrasonography, suggesting infiltration of fat into the muscle.

Our study showed that ultrasonography can measure the differences in the geniohyoid muscle area in the sagittal plane between young and old people and is the first to demonstrate the differences in geniohyoid muscle brightness between young and old people.

Sarcopenia can be treated by resistance training and nutrition. [1, 2]. Feng et al. and Baba et al. reported a correlation between swallowing function and swallowing muscle mass [10, 17]. If sarcopenia of the swallowing muscles is detected at an early stage, swallowing muscle training should be started to prevent sarcopenic dysphagia.

There are two limitations in this study. First, muscle area measurement by ultrasonography was reported to be correlated with MRI measurement [31, 32]. However, for geniohyoid muscle area, an association between ultrasonography and MRI is not clear. In this study, we did not examine the association between ultrasonography and MRI for the measurement of geniohyoid muscle area. Second, effects of thoracic kyphosis or cervical lordosis on US measurements were not evaluated. To avoid stretching the geniohyoid muscle, neck position of the subjects was adjusted to central position. However, the presence of thoracic kyphosis or cervical lordosis was not assessed.

In conclusion, we suggest that atrophy and fatty change of the swallowing muscles are associated with aging and loss of whole-body skeletal muscles, and efficacy of the ultrasonography in their evaluation.

Compliance with ethical standards

Conflict of interest: Takashi Mori, Shinichi Izumi, Yoshimi Suzumkamo, Tatsuma Okazaki, and Susumu Iketani declare no conflicts of interest.

Ethical approval: Ethical committee of Southern Tohoku General Hospital and Tohoku University was approved this study.

Informed consent: All patients were given documents and provided written informed consent prior to enrollment.

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Tables

Table. 1

	Total (*N* = 104)	Young (*N* = 35)	Old (*N* = 69)	*p*
Sex, male, n (%)	70 (67.3)	18 (51.4)	16 (23.2)	0.007
Age, years, mean (SD)	61.3 (20.5)	35.4 (13.9)	74.5 (5.5)	< 0.001
Height (cm), mean (SD)	157.4 (10.0)	164.7 (7.6)	153.7 (10.1)	< 0.001
Weight (kg), mean (SD)	56.6 (10.8)	61.5 (12.9)	54.2 (8.8)	0.004
BMI, mean (SD)	22.7 (3.1)	22.5 (3.6)	22.9 (2.9)	0.647
Calf circumference (cm), mean (SD)	35.1 (3.5)	37.4 (4.1)	33.9 (2.7)	< 0.001
Hand grip strength, (kg), mean (SD)	29.1 (9.7)	35.6 (10.2)	25.8 (7.6)	< 0.001
MNA-SF, score, median (IQR)	13 (12-14)	13 (12-14)	13 (12-14)	0.26*
FILS, score, median (IQR)	10 (10-10)	10 (10-10)	10 (10-10)	n, s**
Maximum tongue pressure (kPa), mean (SD)	37.1 (7.9)	41.2 (7.7)	35.1 (7.3)	< 0.001
Geniohyoid muscle area (mm^2), mean (SD)	192.7 (51.4)	229.5 (52.2)	174.1 (40.7)	< 0.001
Geniohyoid muscle brightness, mean (SD)	55.2 (12.3)	46.6 (11.1)	59.6 (10.8)	< 0.001

Table 1: Characteristics of the subjects
p indicates the *p* value of the Student's t-test except for MNA-SF and FILS, * indicates the *p* value of the Mann-Whitney U test, ** FILS scores for all the subjects were 10 points.

Table.2

(a) Male

	Young (n = 18)	Old (n = 16)	*p* value
Age, years, mean (SD)	33.9 (12.1)	76.6 (5.3)	<0.001
Height, cm, mean (SD)	172.5 (4.3)	163.2 (7.5)	<0.001
Weight, kg, mean (SD)	70.8 (8.0)	63.1 (8.3)	0.009
BMI, mean (SD)	23.9 (3.2)	23.6 (2.1)	0.796
Calf Circumference, cm, mean (SD)	40.0 (3.4)	35.2 (1.8)	<0.001
Hand grip strength, kg, mean (SD)	44.1 (5.8)	34.6 (8.7)	0.001
MNA-SF, score, median (IQR)	14 (13-14)	12.5 (12-14)	0.016
FILS, score, median (IQR)	10 (10-10)	10 (10-10)	n, s**
Maximum Tongue Pressure, kPa, mean (SD)	43.4 (9.2)	33.7 (6.3)	0.001
Geniohyoid muscle area, mm^2, mean (SD)	262.8 (42.1)	190.7 (49.1)	<0.001
Geniohyoid muscle brightness, mean (SD)	40.6 (8.7)	54.2 (8.6)	<0.001

(b) Female

	Young (n = 17)	Old (n = 53)	*p* value
Age, years, mean (SD)	36.9 (15.8)	73.8 (5.5)	<0.001
Height, cm, mean (SD)	156.4 (7.6)	150.(4.8)	0.001
Weight, kg, mean (SD)	51.7 (9.7)	51.5 (7.1)	0.938
BMI, mean (SD)	21.1 (3.7)	22.7 (3.1)	0.1
Calf Circumference, cm, mean (SD)	34.5 (2.7)	33.5 (2.8)	0.189
Hand grip strength, kg, mean (SD)	26.7 (4.4)	23.1 (4.8)	0.009
MNA-SF, score, median (IQR)	38.9 (5.0)	35.5 (7.5)	0.091
FILS, score, median (IQR)	10 (10-10)	10 (10-10)	n, s**
Maximum Tongue Pressure, kPa, mean (SD)	12 (12-13)	13 (12-14)	0.259
Geniohyoid muscle area, mm^2, mean (SD)	194.2 (36.8)	169.0 (36.9)	0.017
Geniohyoid muscle brightness, mean (SD)	53.0 (10.0)	61.2 (10.9)	0.007

Table. 2: Comparison between young and old group in each gender

p indicates the *p* value of the Student's t-test except for MNA-SF and FILS, * indicates the *p* value of the Mann-Whitney U test, ** FILS scores for all the subjects were 10 points.

Table 3

	Age	Height	Weight	BMI	CC	HG	MTP	GH area	GH br.
Age	1	-0.54*	-0.06	0.02	-0.49*	-0.49*	-0.39*	-0.58*	0.52*
Height		1	-0.03	-0.18	0.56*	0.76*	0.24*	0.59*	-0.50*
Weight			1	0.987*	0.15	0.05	0.12	0.04	0.03
BMI				1	0.04	-0.07	0.07	-0.06	0.10
CC					1	0.58*	0.43*	0.58*	-0.30*
HG						1	0.33*	0.68*	-0.51*
MTP							1	0.39*	-0.14
GH area								1	-0.47*
GH br.									1

Table 3: Univariate correlations

Results show Pearson's correlation coefficients, * indicates the p value < 0.05, CC, calf circumference; HG, hand grip strength; MTP, maximum tongue pressure; GH area, cross-sectional area of the geniohyoid muscle in the sagittal plane; GH br., brightness of the geniohyoid muscle in the sagittal plane

Table 4

	Estimated regression coefficient	Lower 95% confidence interval	Upper 95% confidence interval	Standard error	*p* value
(Intercept)	26.58	-228.71	281.86	128.66	0.84
age	-0.87	-1.33	-0.40	0.23	< 0.01
sex	-19.22	-44.72	6.27	12.84	0.14
height	0.69	-0.65	2.03	0.68	0.31
CC	-29	-46.586	-10.492	9.097	< 0.01

Adjusted $R^2 = 0.48$, p value < 0.001

Table 4: Multivariate regression analysis for area of the geniohyoid muscle
CC, calf circumference.

Table 5

	Estimated regression coefficient	Lower 95% confidence interval	Upper 95% confidence interval	Standard error	*p* value
(Intercept)	7.74	-59.52	75.00	33.90	0.82
age	0.30	0.18	0.42	0.06	< 0.01
sex	11.40	4.68	18.12	3.39	< 0.01
height	0.00	-0.35	0.36	0.18	0.99
CC	0.59	-0.10	1.28	0.35	0.09

Adjusted $R^2 = 0.39$, p value < 0.001

Table 5: Multivariate regression analysis for brightness of the geniohyoid muscle
CC, calf circumference.

Figure Legends

Fig. 1: Ultrasonography method
(a) posture during ultrasonography, (b) ultrasonography image of the geniohyoid muscle in the sagittal plane, (c) detection of an area of interest using image J. All images belong to author T.M.

Fig. 2: Cross-sectional area of the geniohyoid muscle in the young and old groups.
p value by Student's t-test

Fig. 3: Brightness of the geniohyoid muscle in the young and old groups.
p value by Student's t-test