Quantitative Sonographic Assessment of the Quadriceps Femoris Muscle in Healthy Japanese Adults

Tsuneo Watanabe, PhD, Hiroki Murakami, MS, Daisuke Fukuoka, PhD, Nobuo Terabayashi, MD, PhD, Sohee Shin, PhD, Tamotsu Yabumoto, PhD, Hiroyasu Ito, MD, PhD, Hiroshi Fujita, PhD, Toshio Matsuoka, PhD, Mitsuru Seishima, MD, PhD

Objectives—The aim of this study was to evaluate the relationships among aging, muscle strength, and image feature analysis of the quadriceps femoris muscle and to evaluate the relationship between aging, muscle strength, and sonographic findings.

Methods—One hundred forty-five healthy volunteers participated in the study. The participants were classified into 6 groups on the basis of sex and age. To assess muscle quality, texture analysis was defined with the following parameters: mean, skewness, kurtosis, inverse difference moment, sum of entropy, and angular second moment. The knee extension force in the sitting position and thickness of the quadriceps femoris muscle were also measured.

Results—The quadriceps femoris thickness, skewness, kurtosis, inverse difference moment, angular second moment, and muscle strength were significantly decreased in elderly participants versus those in the younger and middle-aged groups ($P < .05$). In contrast, the mean and sum of entropy were significantly decreased in the younger group compared with the middle-aged and elderly groups.

Conclusions—Sonography has the capacity to quantitatively assess muscular morphologic changes due to aging and could be a valuable tool for early detection of musculoskeletal disorders.

Key Words—knee extension strength; musculoskeletal disorders; musculoskeletal ultrasound; sonography; texture analysis

Aging populations have been a major point of concern in developed nations in recent years. In Japan, at least 20% of the population is 65 years or older, which makes it one of the oldest populations in the world. Early detection of musculoskeletal disorders is expected to help promote well-being and self-reliance among elderly people and to reduce the need for nursing care. Sarcopenia has been defined as the loss of skeletal muscle mass and strength that occurs with advanced age.[1] The European Working Group on Sarcopenia in Older People provided a working definition of sarcopenia as "a syndrome characterized by progressive and generalized loss of skeletal muscle mass and strength with a risk of adverse outcomes such as physical disability, poor quality of life, and death."[2] Previous studies have assessed the quality of skeletal muscle using computed tomography (CT)[3] and magnetic resonance imaging.[4] Over the past few years, there have been many improvements in high-resolution diagnostic ultrasound equipment. Echogenicity, the characteristic of tissue or other material to reflect ultrasonic waves, is

Received July 21, 2016, from the Division of Clinical Laboratory, Gifu University Hospital, Gifu, Japan (T.W, H.I., M.S.); Departments of Intelligent Image Information (H.M., H.F.), Orthopedic Surgery (N.T.) and Sports Medicine and Sports Science (S.S., T.Y., T.M.), Gifu University Graduate School of Medicine, Gifu, Japan; and Faculty of Education, Gifu University, Gifu, Japan (D.F.). Manuscript accepted for publication September 23, 2016.

This study was supported by a research grant to T.W. from the Series of Single-Year Grants (JSPS KAKENHI 15H00698) and a research grant to H.F. from the Grants-in-Aid for Scientific Research on Innovative Areas (Multidisciplinary Computational Anatomy, JSPS KAKENHI 26108005).

Address correspondence to Tsuneo Watanabe, PhD, Division of Clinical Laboratory, Gifu University Hospital, 1-1 Yanagido, Gifu 501-1194, Japan.

E-mail: tsuneo_w@gifu-u.ac.jp

Abbreviations
ANOVA, analysis of variance; CT, computed tomography; DVT, deep venous thrombosis; GLCM, gray-level co-occurrence matrix

doi:10.7863/ultra.16.07054

typically used as a sonographic estimate of muscle quality. Measures of muscle echogenicity have been shown to be significantly associated with intramuscular adipose tissue, rather than fibrotic tissue, based on the biochemical analysis of biopsied tissue samples from 82 people with muscle disorders.[5] Recently, several investigators have used computer-aided echo intensity analysis (histogram analysis) to evaluate the quality of skeletal muscle.[6–10] We have also reported a positive correlation between sonography and magnetic resonance imaging in the assessment of supraspinatus muscle fatty infiltration.[11] Watanabe et al[12] reported that muscle thickness and muscle quality assessed from echo intensity measured by sonography independently contributed to isometric knee extension strength in elderly men (65–91 years). However, it is well known that the value of echo intensity on histogram analysis is affected by the thickness of tissue. Thus, it seems the accuracy of using echo intensity analysis to estimate muscle quality is limited.

Meanwhile, advancements in image analysis, aided by computer software improvements, provide important information about the structural arrangement of surfaces and their relationship with the surrounding environment. Several investigators demonstrated that gray-level co-occurrence matrix (GLCM)[13] statistical measures, when computed, provided useful information for discrimination purposes in clinical studies. Gomez et al[14] used texture features to classify sonograms in the breast. The texture descriptors that contributed notably to distinguish breast lesions were contrast and correlation computed from GLCMs with an orientation of 90° and a distance of greater than 5 pixels. Regarding musculoskeletal disorders, Shen et al[15] reported significant differences in GLCM parameters derived from high-resolution magnetic resonance images between postmenopausal women with osteoporosis and osteoarthritis. Molinari et al[16] used texture features to discriminate between sexes and muscle types from the quantitative assessment of skeletal muscle sonograms. Their results concluded that multitexture analysis may be useful to investigate muscle damage and myopathic disorders. However, these studies examined quantitative assessment using texture analysis and did not examine aging. Thus, to our knowledge, there are no published studies in English examining the effect of aging on the quadriceps femoris muscle using sonographic texture analysis. We hypothesized that there would be no differences in the sonographic characteristics of the quadriceps femoris muscle among various age and sex groups. The primary objective of this study was to evaluate the relationships among aging, muscle strength, and image feature analysis of the quadriceps femoris muscle. The secondary objective was to estimate muscle strength in healthy adults via sonographic findings.

Materials and Methods

Study Design and Setting

This cross-sectional observational study was conducted to evaluate the relationships among aging, muscle strength, and image feature analysis of the quadriceps femoris muscle at Gifu University. Image acquisition was completed in 2 local community centers or the laboratory of a sports medicine department. A post–image acquisition analysis was undertaken within a laboratory at the university.

Participants

One hundred forty-five healthy volunteers who lived in Gifu, Japan, were examined. Participants had no injuries or disabilities in their lower limbs, and every participant was able to walk unaided. Exclusion criteria were as follows: (1) the ability to walk with an assistive device; (2) history of lower limb trauma or surgery; (3) neuromuscular disorders; (4) acute or chronic disease that impaired strength and power; and (5) severe dementia (which might have influenced informed consent). The participants were classified into 6 groups on the basis of sex and age as follows: younger group (<44 years; mean age ± SD, 23.2 ± 5.0 years; n = 68), middle-aged group (45–64 years; mean age, 55.7 ± 6.3 years; n = 31), and elderly group (≥65 years; mean age, 73.2 ± 5.3 years; n = 46). We studied a total of 290 lower limbs in the 145 participants, who underwent both knee extension strength and sonographic examinations. This study was conducted in accordance with the 1964 Declaration of Helsinki and was approved by an Institutional Ethics Committee; written informed consent was obtained from all participants.

Measurement of Muscle Thickness

Muscle thickness and quality were evaluated sonographically with a 6.0–14.0-MHz linear transducer and portable ultrasound apparatus (LOGIQ *e*; GE Healthcare, Pittsburgh, PA). Sonographic examinations were performed by a board-certified sonographer, who was blinded to the group assignment. Muscle thickness was evaluated according to the procedure of Fukumoto et al[8] with modifications. The quadriceps femoris muscle thickness was defined as the sum of the thickness of the rectus femoris and vastus intermedius muscles. The quadriceps femoris thickness was measured in the axial view at the midpoint between the lateral epicondyle of the femur and the anterior superior iliac spine. The transducer was positioned perpendicular to the longitudinal axis of the quadriceps femoris.

Measurement of Muscle Quality

To assess muscle quality, first- and second-order statistical analyses were performed. All images were analyzed by a single observer (H.M.), who was blinded to personal information such as age and sex. The background of this observer included medical image analysis and computer engineering. In statistical analyses, texture features were computed from the statistical distributions of observed combinations of intensities at specified positions relative to each other in the image. According to the number of intensity points (pixels) in each combination, statistics were classified as first-order and second-order statistics.[17] The histogram contained first-order statistics. Based on the first-order statistics, the following 7 features were extracted from the image of a region of the interest: coarseness, mean, entropy, variance, skewness, kurtosis, and energy. The mean, skewness, and kurtosis were used to analyze these parameters. Skewness describes a measure of asymmetry of the histogram, and kurtosis shows a measure of peakedness and tailedness. Meanwhile, the GLCM is a mathematical method used for statistical texture analysis; GLCM texture measurement was proposed by Haralick and Shanmugam[13] with 14 different textural features, and GLCM computation can be performed in 4 directions: 0°, 45°, 90°, and 135°. In the proposed work, an averaged 4-direction value is used for feature extraction to avoid dependence on the direction.[17] These features were mathematically defined with the following parameters: angular second moment, contrast, correlation, sum of squares, inverse difference moment, sum of average, sum variance, sum of entropy, difference variance, difference entropy, information measures of correlation, and maximal correlation coefficient[18]; this study was implemented by using 3 parameters (inverse difference moment, sum of entropy, and angular second moment). These parameters summarize important information about the structural arrangement of surfaces by discerning likelihoods that pixels have the same or different gray-level values as their neighbors and distances between pixel pairs of equal intensity. The Haralick textural features are as follows:

The GLCM is a square matrix with a dimension equal to the number of gray levels in the image. Let C be the matrix containing the GLCM. The element $C(i,j)$ measures the number of times in which a pixel of a given gray level i is found adjacent to a pixel of gray level j. G is the number of gray levels used; μ is the mean value of C; μ_x, μ_y, σ_x, and σ_y are the means and standard deviations of C_x and C_y.

The inverse difference moment describes the homogeneity of an image, with a higher value reflecting more similarity between pixels:

$$f1=\sum_{i=0}^{G-1}\sum_{j=0}^{G-1}\frac{1}{1+(i-j)^2}C(i,j). \tag{1}$$

The sum of entropy measures the disorder or complexity within an image:

$$f2=-\sum_{i=0}^{2G-2}C_{x+y}(i)\log\{C_{x+y}(i)\}; \tag{2}$$

Table 1. Intraobserver and Interobserver Reproducibility

Parameter	Intra-CC (Inter-CC)	95% CI
Mean		
Observer A	0.973	0.935–0.992
Observer B	0.966	0.920–0.990
Observers A and B	(0.970)	0.885–0.993
Skewness		
Observer A	0.936	0.854–0.981
Observer B	0.958	0.903–0.988
Observers A and B	(0.975)	0.903–0.993
Kurtosis		
Observer A	0.926	0.833–0.978
Observer B	0.939	0.860–0.982
Observers A and B	(0.973)	0.895–0.993
Inverse difference moment		
Observer A	0.992	0.982–0.997
Observer B	0.992	0.982–0.998
Observers A and B	(0.992)	0.970–0.998
Angular second moment		
Observer A	0.982	0.956–0.994
Observer B	0.983	0.961–0.995
Observers A and B	(0.977)	0.901–0.995
Sum of entropy		
Observer A	0.987	0.968–0.996
Observer B	0.988	0.972–0.996
Observers A and B	(0.982)	0.928–0.995

CI indicates confidence interval; Inter-CC, interclass correlation coefficient; and Intra-CC, intraclass correlation coefficient.

$$C_{x+y}(k)=\sum_{i=0}^{G-1}\sum_{j=0}^{G-1}C(i,j)i+j=k \text{ for } k=0,1,...2(G-1). \quad (3)$$

The angular second moment is low when all elements in the GLCM are close to either 0 or 1 and high when the GLCM has equal values or pixels are similar, which is calculated by the following:

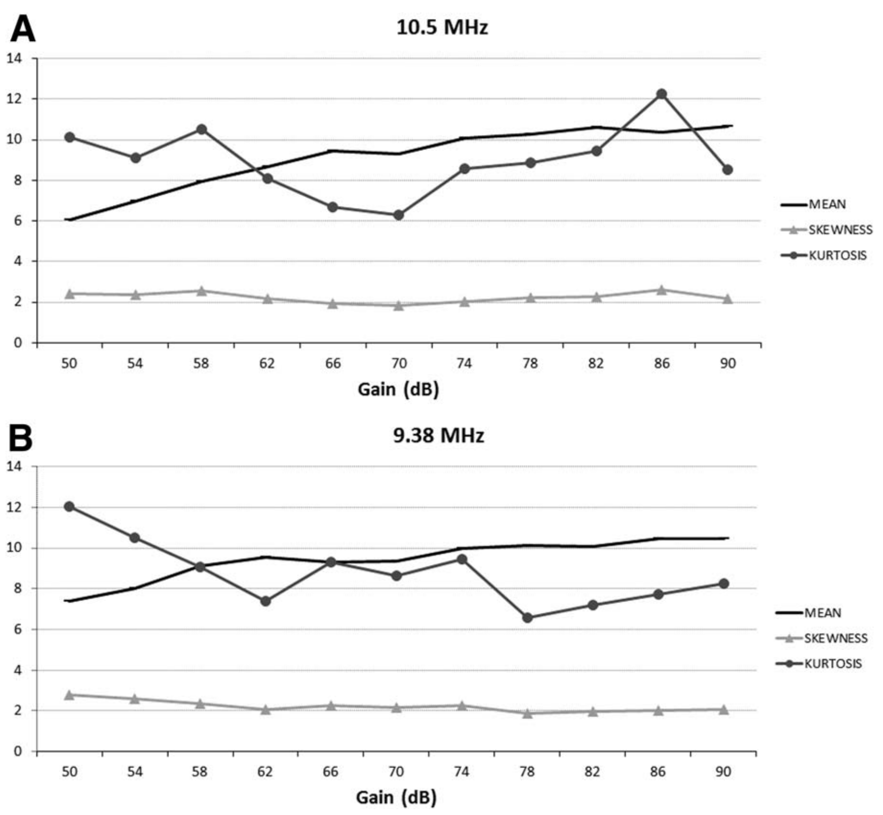
$$f3=\sum_{i=0}^{G-1}\sum_{j=0}^{G-1}\{C(i,j)\}^2. \quad (4)$$

Intraobserver and interobserver reliabilities were assessed for 10 images before beginning this study. Texture analysis of the quadriceps femoris was repeated 5 times for each image by 2 observers. Intraobserver reliability was estimated by using calculations of intraclass correlation coefficients (1-way analysis of variance [ANOVA]). Interobserver reliability was estimated by using calculations of interclass correlation coefficients (2-way ANOVA). The intraobserver and interobserver reproducibilities of the texture analyses were high (Table 1). A correlation coefficient value of greater than 0.9 was considered very good. Furthermore, we conducted a preliminary examination to confirm the change by gain or frequency. The results of the effects of gain and frequency shifts on the analysis are shown in Figures 1 and 2. The Haralick features (GLCM) had little effect on the change by gain or frequency, in comparison with the case of using a conventional first-order statistical feature (histogram analysis).

Measurement of Muscle Strength

Muscle strength was evaluated according to the procedure of Watanabe et al.[12] The maximum isometric torque of knee extension at an angle of 90° was measured in a sitting position on a custom-made dynamometer chair. The ankle was attached to a strain-gauge system

Figure 1. Effects of gain and frequency shifts on the first-order statistical features. **A**, Frequency was set at 10.5 MHz. **B**, Frequency was set at 9.38 MHz. Lines connecting rectangles show the change in the mean; lines connecting triangles show the change in the skewness; and lines connecting circles show the change in the kurtosis according to the gain shift.



(TKK5715; Takei Scientific Instruments Co, Ltd, Niigata, Japan). After participants were familiarized with the test procedure, 2 trials at maximum effort were made with a 1-minute recovery period. The greater value obtained was used for analysis. The length between the lateral epicondyle of the femur and the ankle attachment was measured.

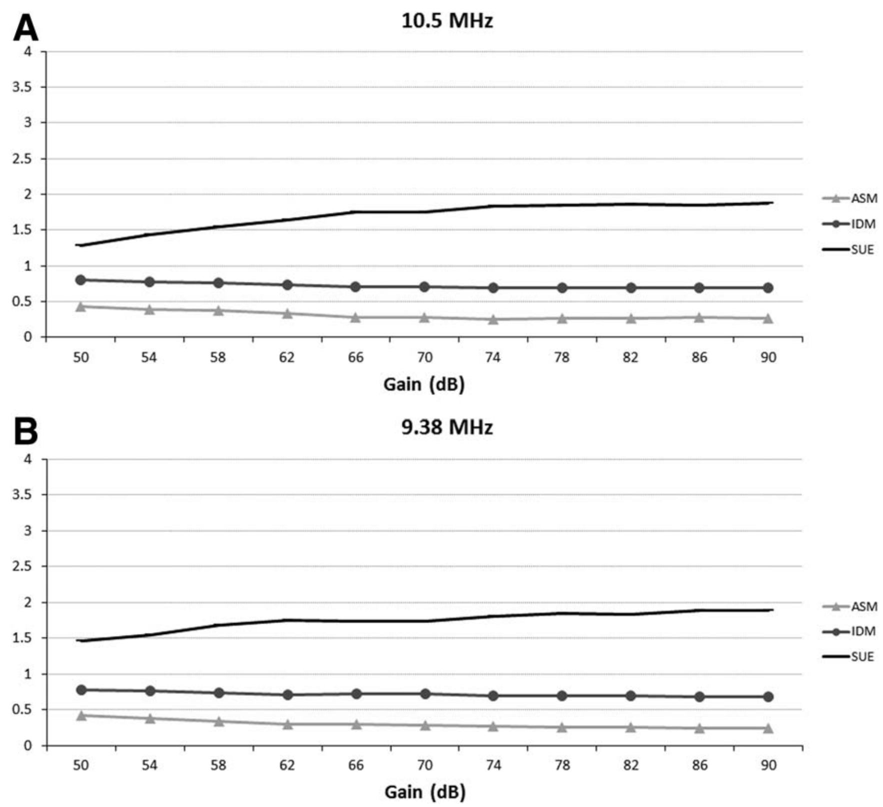
Statistical Analyses

Results are presented as mean ± standard deviation where applicable, and statistical significance was indicated by $P < .05$. A 2-way factorial ANOVA test was used to determine differences between quadriceps femoris thickness, mean, skewness, kurtosis, inverse difference moment, angular second moment, sum of entropy, and muscle strength (3 × 3: male/female × younger/middle-aged/elderly group). The Tukey honestly significant difference test was applied to assess differences among multiple comparisons when ANOVA indicated a significant difference for a particular factor. Pearson correlation coefficients were used to investigate the correlation of age and muscle strength with sonographic parameters. Although the age effects for lower limb strength were quantified by a quadratic method, the sonographic or physical characteristics and participants' ages were investigated by a stepwise multivariate analysis. A multiple logistic model was applied with adjustment for age, sex, height, quadriceps femoris thickness, mean, skewness, kurtosis, inverse difference moment, angular second moment, and sum of entropy. SPSS Statistics for Windows version 19.0 (IBM Corporation, Armonk, NY) was used for all calculations.

Results

The demographic and physiologic characteristics of the participants are shown in Table 2. Measurements for several parameters according to group and sex are shown in Table 3. Significant interactions were observed in the quadriceps femoris thickness $[F_{(2,284)} = 6.5; P = .002]$,

Figure 2. Effects of gain and frequency shifts on the Haralick features (GLCM). **A**, Frequency was set at 10.5 MHz. **B**, Frequency was set at 9.38 MHz. Lines connecting rectangles show the change in the sum of entropy (SUE); lines connecting triangles show the change in the angular second moment (ASM); and lines connecting circles show the change in the inverse difference moment (IDM) according to the gain shift.



mean [$F_{(2,284)} = 3.8$; $P = .024$], skewness [$F_{(2,284)} = 13.7$; $P < .001$], kurtosis [$F_{(2,284)} = 13.7$; $P < .001$], angular second moment [$F_{(2,284)} = 4.1$; $P = .018$], and muscle strength [$F_{(2,284)} = 6.7$; $P = .002$]. For the quadriceps femoris thickness, skewness, kurtosis, inverse difference moment, angular second moment, sum of entropy, and muscle strength, significant main effects for the various groups and sex were observed ($P < .001$). Despite the absence of any significant main effects for sex, a significant main effect according to group was observed in the mean ($P < .001$). Subsequently, the quadriceps femoris thickness and muscle strength were significantly higher in men than in women. In contrast, the mean and sum of entropy of men in the middle-aged group were significantly lower than those of women in middle-aged group. The relationship between aging and sonographic features is illustrated in Figure 3. Figure 4 shows changes in the quadriceps femoris thickness value for each group. For men, the quadriceps femoris thickness significantly decreased in the elderly group compared with the younger and middle-aged

Table 2. Anthropometric Characteristics in 145 Participants

Characteristic	Mean	SD	Range
Age, y	46.0	23.0	19–86
Height, cm	162.4	9.4	143.5–183.0
Weight, kg	58.4	10.5	35.6–99.0
Body mass index, kg/m^2	22.1	3.0	15.9–33.0

Table 3. Analysis of Variance of Several Parameters in the Participants

			F (2-way ANOVA)			
Parameter	**Male**	**Female**	**Group**	**Sex**	**Interaction**	**Post Hoc Test (Tukey HSD)**
Quadriceps femoris thickness, mm						
Younger	32.4 ± 6.4	28.9 ± 5.2	44.4	74.7	6.5	Male: younger, middle > elderly
Middle	34.3 ± 5.7	24.4 ± 6.6	(*P* < .001)	(*P* < .001)	(*P* = .002)	Female: younger > middle > elderly
Elderly	25.9 ± 5.7	20.7 ± 4.8				Younger, middle, elderly: male > female
Mean						
Younger	11.4 ± 1.5	11.7 ± 1.6	34.6	5.0	3.8	Male: younger, middle < elderly
Middle	11.9 ± 1.2	13.7 ± 2.1	(*P* < .001)	(*P* = .261)	(*P* = .024)	Female: younger < middle, elderly
Elderly	13.9 ± 2.4	13.7 ± 2.1				Middle: male < female
Skewness						
Younger	2.3 ± 0.4	2.3 ± 0.3	44.8	21.1	13.7	Male: younger, middle > elderly
Middle	2.4 ± 0.3	1.8 ± 0.2	(*P* < .001)	(*P* < .001)	(*P* < .001)	Female: younger > middle, elderly
Elderly	1.9 ± 0.2	1.8 ± 0.3				Middle: male > female
Kurtosis						
Younger	9.2 ± 2.4	8.9 ± 2.0	36.2	19.2	13.7	Male: younger, middle > elderly
Middle	9.9 ± 1.9	6.4 ± 1.2	(*P* < .001)	(*P* < .001)	(*P* < .001)	Female: younger > middle, elderly
Elderly	6.4 ± 1.7	6.6 ± 1.6				Middle: male > female
Inverse difference moment						
Younger	0.73 ± 0.04	0.73 ± 0.03	35.4	14.7	1.6	Male: younger > middle, elderly
Middle	0.71 ± 0.03	0.68 ± 0.03	(*P* < .001)	(*P* < .001)	(*P* = .211)	Female: younger > middle, elderly
Elderly	0.70 ± 0.04	0.68 ± 0.03				Middle, elderly: male > female
Angular second moment						
Younger	0.33 ± 0.05	0.32 ± 0.04	38.6	24.6	4.1	Male: younger > elderly
Middle	0.31 ± 0.05	0.26 ± 0.03	(*P* < .001)	(*P* < .001)	(*P* = .018)	Female: younger > middle, elderly
Elderly	0.28 ± 0.06	0.24 ± 0.04				Middle, elderly: male > female
Sum of entropy						
Younger	1.79 ± 0.14	1.82 ± 0.12	43.4	16.1	2.3	Male: younger < elderly
Middle	1.87 ± 0.12	2.00 ± 0.12	(*P* < .001)	(*P* < .001)	(*P* = .102)	Female: younger < middle, elderly
Elderly	1.96 ± 0.15	2.02 ± 0.15				Middle: male < female
Muscle strength, kg						
Younger	51.8 ± 12.2	33.9 ± 8.3	56.8	120.8	6.7	Male: younger > middle > elderly
Middle	40.5 ± 7.3	26.6 ± 7.7	(*P* < .001)	(*P* < .001)	(*P* = .002)	Female: younger, middle > elderly
Elderly	33.3 ± 11.9	25.1 ± 6.8				Younger, middle, elderly: male > female

Values are mean ± SD where applicable. HSD indicates honestly significant difference.

groups ($P < .001$ for both). For women, the quadriceps femoris thickness significantly decreased in the elderly group compared with the younger and middle-aged groups ($P < .001$; $P < .05$, respectively) and in the middle-aged compared with the younger group ($P < .01$). Concerning the texture analysis, the mean for women and the sum of entropy significantly decreased in the younger group compared with the middle-aged and elderly groups ($P < .001$). For men, the mean significantly increased in the elderly group compared with the younger and middle-aged groups ($P < .001$; $P < .05$). The skewness and kurtosis for women, as well as the inverse difference moment and angular second moment, significantly increased in the younger group compared with the middle-aged and elderly groups ($P < .001$). For men, the skewness and kurtosis significantly decreased in the elderly group compared with the younger and middle-aged groups ($P < .001$). Figure 5 shows changes in the muscle strength value for each group. For women, the muscle strength significantly increased in the younger group compared with the middle-aged and elderly groups ($P < .001$ for both).

Figure 3. A, Longitudinal sonogram of the quadriceps femoris in the younger female group. **B**, Longitudinal sonogram of the quadriceps femoris in the middle-aged female group. **C**, Longitudinal sonogram of the quadriceps femoris in the elderly female group. RF indicates rectus femoris; and VI, vastus intermedius.

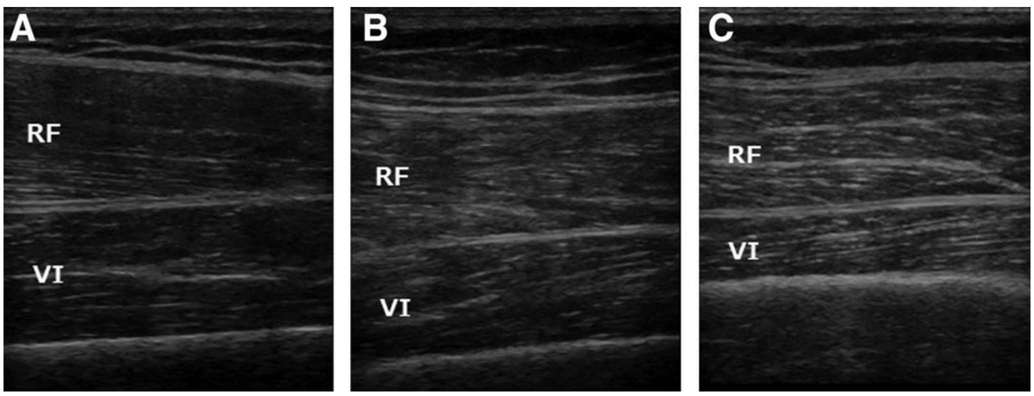
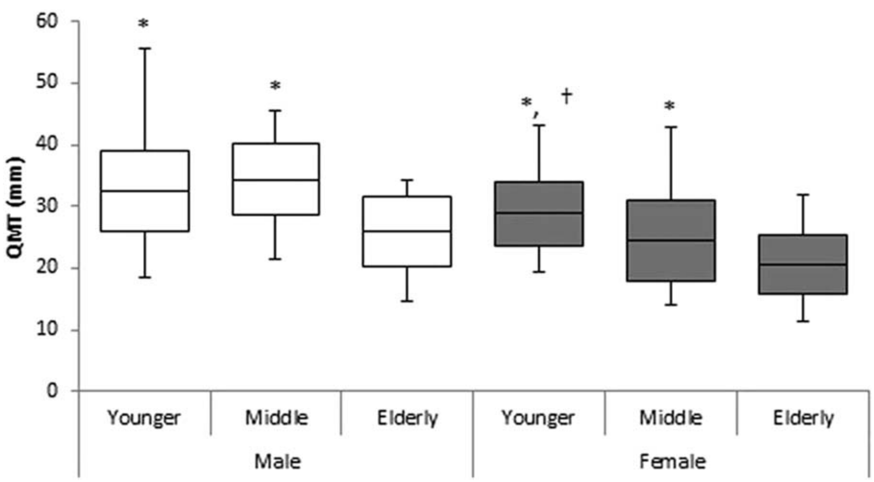


Figure 4. Changes in the quadriceps femoris muscle thickness (QMT) by sex and age groups. *$P < .05$ versus the elderly group; †$P < .05$ versus the middle-aged group.



For men, the muscle strength significantly decreased in the elderly group compared with the younger and middle-aged groups ($P < .001$; $P < .01$) and in the middle-aged compared with the younger group ($P < .001$).

Table 4 shows the correlation results among age, muscle strength, and various parameters for all participants. Age showed a significant correlation with several characteristics, with the exception of weight. Meanwhile, muscle strength showed a significant correlation with all parameters. In addition, the relative contributions of each parameter (age, sex, height, quadriceps femoris thickness, mean, skewness, kurtosis, inverse difference moment, angular second moment, and sum of entropy) to the muscle strength level were examined in a stepwise multivariate linear regression analysis (Table 5). The variables independently associated with muscle strength level were age, sex, quadriceps femoris thickness, and sum of entropy. Among these variables, the sex difference was the strongest independent variable that contributed to muscle strength ($\beta = -0.354$; $P < .001$). The following equation was developed to determine pre-

Figure 5. Changes in the muscle strength by sex and age groups. *$P < .05$ versus the elderly group; †$P < .05$ versus the middle-aged group.

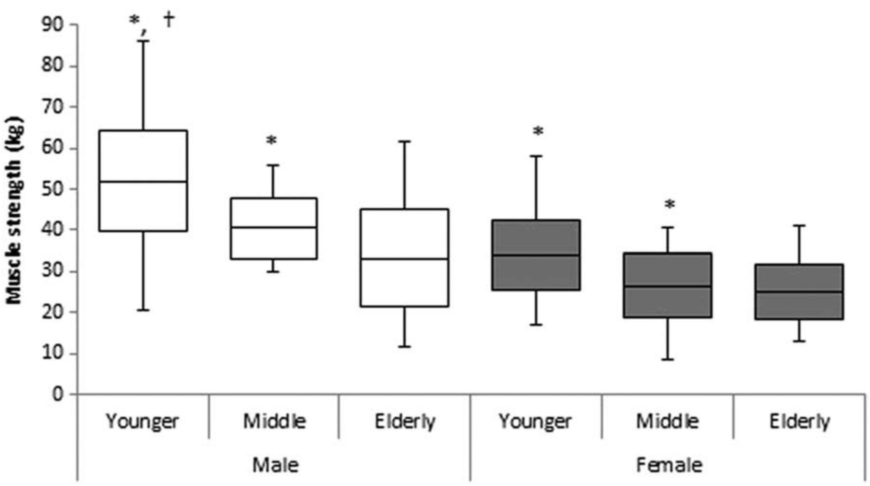


Table 4. Correlation Among Age, Muscle Strength, and Various Parameters

	Age		Muscle Strength	
Parameter	R^2	P	R^2	P
Age	NA	NA	0.29	<.001
Height	0.25	<.001	0.28	<.001
Weight	0.03	.004	0.54	<.001
Body mass index	0.04	<.001	0.03	.003
Quadriceps femoris thickness	0.24	<.001	0.36	<.001
Mean	0.24	<.001	0.25	<.001
Skewness	0.29	<.001	0.26	<.001
Kurtosis	0.24	<.001	0.25	<.001
Inverse difference moment	0.29	<.001	0.27	<.001
Angular second moment	0.31	<.001	0.27	<.001
Sum of entropy	0.32	<.001	0.32	<.001
Muscle strength	0.29	<.001	NA	NA

NA indicates not applicable.

Table 5. Multivariate Regression Analysis for Muscle Strength With Various Parameters as Independent Variables

Parameter	β	P
Sex	–0.354	<.001
Age	–0.225	<.001
Height	NE	NA
Quadriceps femoris thickness	0.216	<.001
Angular second moment	NE	NA
Sum of entropy	–0.234	<.001
Mean	NE	NA
Skewness	NE	NA
Inverse difference moment	NE	NA
Kurtosis	NE	NA

Sex: male = 1; female = 2. NA indicates not applicable; and NE, not entered.

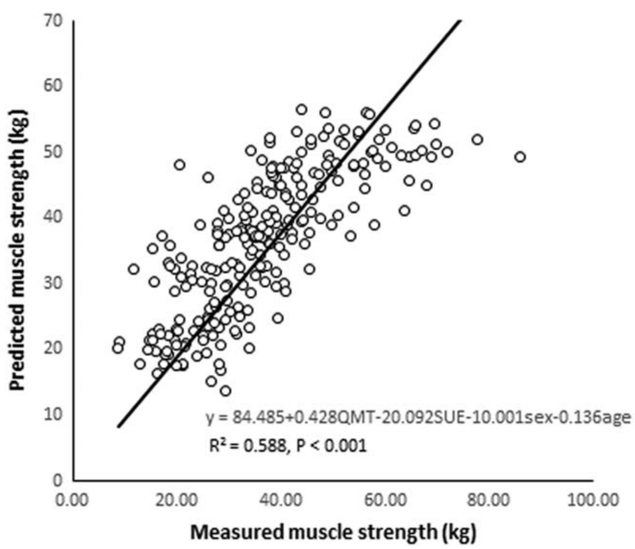
dicted muscle strength: $y = 84.485 + 0.428 \times$ quadriceps femoris thickness – $20.092 \times$ sum of entropy – $10.001 \times$ sex – $0.136 \times$ age (Figure 6).

Discussion

A common and often devastating problem among elderly people is falling, which causes a tremendous amount of morbidity, mortality, and use of health care services, including premature nursing home admissions.[19] Rubenstein[19] reported that the most important fall risk factors are muscle weakness and problems with gait and balance. Muscle weakness, in particular, approximately quintuples the risk of falling. Thus, accurate assessment of the muscle condition of the lower limbs is crucial for early detection of musculoskeletal disorders. This study investigated the influence of aging on lower limb muscle strength and quadriceps femoris thickness in healthy adults. Furthermore, we have attempted to develop a method for prediction of muscle strength using sonography. To our knowledge, this work is the first study to propose a muscular index using sonography. The study demonstrated the influence of aging on the lower limbs, such as decreased muscle strength, thickness, skewness, kurtosis, inverse difference moment, and angular second moment, and a significant increase in texture analysis, such as mean and sum of entropy.

Several researchers have already investigated the effects of aging on skeletal muscle. For example, Ogawa et al[20] evaluated muscle thickness using CT and concluded that loss of muscle thickness is associated with aging. A magnetic resonance spectroscopic study showed that elderly individuals have significantly greater intramuscular lipids than young individuals.[21] In another study, there was an association between fat mass and muscle strength and aging, suggesting that a high degree of fat mass is associated with lower muscle quality.[22] Our results showed a significant decrease in muscle strength related to a decrease in texture and aging, which is in agreement with previous studies showing the effects of aging on muscle strength and muscle quality. Recently, some research groups have used computer-aided grayscale analysis to evaluate the quality of skeletal muscle.[6–8,11] Watanabe et al[12] reported echo intensities that showed a significant negative correlation with muscle strength in 184 elderly men (65–91 years). Fukumoto et al[8] also reported that muscle thickness and muscle quality as assessed by echo intensity independently were associated with muscle strength in middle-aged and elderly women (51–87 years). Despite these results, the echo intensity method has a major shortcoming that limits its clinical use because it is dependent on ultrasound settings. Furthermore, it is well known that echo intensity is able to capture the overall change in

Figure 6. Correlation between the predicted and measured muscle strength. A scatter diagram and a regression line are shown. Regression analysis revealed that the values were positively correlated. QMT indicates quadriceps femoris muscle thickness; and SUE, sum of entropy.



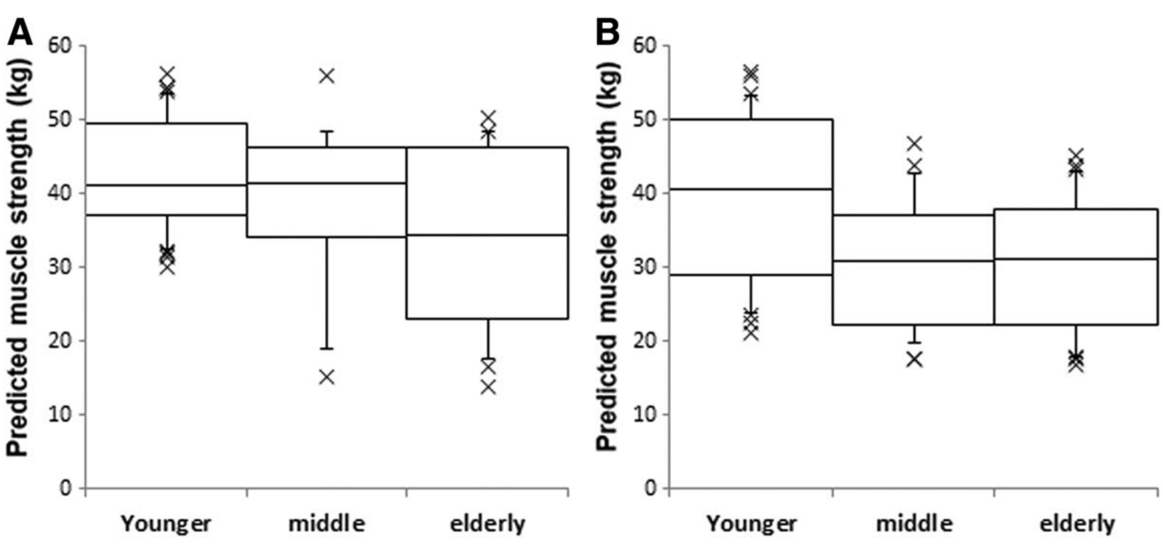
brightness of a muscle, but this numeric parameter is highly dependent on the ultrasound scanner settings.[23] In contrast, few studies have attempted to develop quantification methods that can overcome these limitations. Molinari et al[16] proposed texture features as superior to echo intensity, which they used to measure 5 muscles (biceps brachii, rectus femoris, vastus lateralis, tibialis anterior, and medial gastrocnemius) of 20 healthy participants to assess how the performance of higher-order texture descriptors differ between sexes and among muscles. Their results showed that Haralick features (energy, entropy, and correlation measured along different angles), local binary pattern features, and entropy were highly linked to sex, whereas Haralick entropy and symmetry, Galloway texture descriptors, and local binary pattern entropy helped distinguish muscle types. Our results showed a significant decrease in muscle strength related to increased age and texture features such as the mean and sum of entropy and decreased texture features such as the skewness, kurtosis, inverse difference moment, and angular second moment.

In general, sarcopenia, including muscle atrophy, is frequently observed with aging. The latest reports have shown it to be associated with chronic disease such as liver disease, and sarcopenia is now in the spotlight for several aspects.[24] Recently, various working groups on sarcopenia throughout the world have proposed that a definition should be formed by using a combined approach based on muscle mass and muscle quality. Psoas muscle atrophy shown by CT was reported to have predictive value for mortality in cirrhotic patients, independent of Model for End-Stage Liver Disease and Model for End-Stage Liver Disease–Na scores.[25] Baumgartner et al[26] used dual-energy x-ray absorptiometry to define sarcopenia, whereas Tanimoto et al[27] reported a bioelectrical impedance analysis cutoff for muscle atro-

Table 6. Distribution of the Predicted Muscle Strength According to the Age Category

	Male		Female	
Age, y	**Mean (± 2 SD)**	**Median (5th%–95th%)**	**Mean (± 2 SD)**	**Median (5th%–95th%)**
<45	43.1 (36.0–50.1)	41.2 (32.5–53.6)	39.3 (27.8–50.8)	40.5 (23.7–53.3)
45–64	38.2 (26.2–50.1)	41.4 (19.0–48.4)	30.1 (21.4–38.8)	30.8 (19.8–42.8)
≥65	33.5 (21.4–45.6)	34.3 (17.6–48.8)	30.4 (21.8–38.9)	31.1 (17.9–43.0)

Figure 7. A, Predicted muscle strength values in men by age group. **B**, Predicted muscle strength values in women by age group. Boxes represent 50% of the data; bars represent the remainder; horizontal lines represent medians; and × symbols represent outliers.



phy. Although CT can evaluate muscle volume, it is not widely available, involves the use of ionizing radiation, and is not cost efficient. Moreover, it is difficult to perform dual-energy x-ray absorptiometry and a bioelectrical impedance analysis because most institutions do not have the appropriate equipment. Although grip strength and respiratory function tests are less-expensive alternatives for evaluation of muscular weakness, those tests cannot evaluate morphologic changes. On the other hand, although there is some diversity of opinion regarding the bioeffects of ultrasound regarding the relationship between acoustic output and acoustic bioeffects such as the mechanical index and thermal index,[28] sonography remains highly useful because of the low cost, lack of radiation exposure, and ability to directly show morphologic changes in muscle. Therefore, we propose a new method for quantitative assessment of age-related changes in the lower limbs using sonography. Concerning the diagnostic cutoff value for muscle atrophy, Baumgartner et al[26] used dual-energy X-ray absorptiometry and defined sarcopenia as appendicular skeletal muscle mass (kilograms)/height (meters)^2 of less than 2 SDs below the mean of a young reference group. Normal and reference values for predicted muscle strength are presented in Table 6 and Figure 7. The mean values for men in the younger, middle-aged, and elderly groups were 43.1 $\pm$ 7.1, 38.2 $\pm$ 12.0, and 33.5 $\pm$ 12.1, respectively, and the values for women were 39.3 $\pm$ 11.5, 30.1 $\pm$ 8.7, and 30.4 $\pm$ 8.6. Therefore, the optimal cutoff values derived at less than 2 SDs for men and women in the middle-aged groups were 26.2 and 21.4. The knee extension strength cutoff points for maintaining mobility were recorded by using an isokinetic dynamometer. The results showed that the incidence of severe mobility limitation corresponded to less than 1.13 and greater than 1.71 N-m/kg in men and less than 1.01 and greater than 1.34 N-m/kg in women.[29] There are, however, some concerns about measurement of knee extension strength using an isokinetic dynamometer, given that data are unlikely to be easily interpreted. Regarding investigation of elderly Japanese patients, Inagaki et al[30] measured the knee extension strength of 831 participants older than 60 years using the same device used in this study (TKK5715). In that study, the average values for knee extension strength were approximately 30 kg in men and 20 kg in women. Thus, we established the cutoff values in our study in alignment with their findings. Furthermore, it is well known that disuse syndrome is developed by a sedentary lifestyle, which is shown by several symptoms, such as muscular deconditioning and atrophy resulting from inactivity or immobilization. In patients with severe motor and intellectual disabilities who are confined to bed and with decreased mobility of the lower extremities, there is a higher risk of the complication of deep venous thrombosis (DVT).[31] This condition can have an asymptomatic clinical course, but some cases of DVT develop pulmonary thromboembolism, possibly causing sudden death.[32] The data in this study demonstrate that the proposed index correlated significantly with muscle strength. The proposed index may be a useful marker in the assessment of muscular strength for early detection of DVT because muscle weakness may be involved in DVT formation. Therefore, we think that predicting muscle strength by using sonography is an effective method for rehabilitation program creation in elderly individuals with dementia or who have disorders of consciousness and in patients with low activities of daily living.

There were several limitations to this study. First, it did not include patients with low activities of daily living and did not evaluate the potential enhancement of onset of DVT in sarcopenia. We collected no data to assess the prognostic value of the increase in the predictive formula for patients with an impaired musculoskeletal system. Second, we only used a single sonographer and a single piece of equipment; therefore, the actual capability of using texture analysis, including the influence of a plurality of devices or the influence of detection on a sonogram during imaging, remains unknown. Third, the study did not include multiple ethnic groups. It examined only Japanese individuals; therefore, there was the potential of an ethnicity bias. Finally, our study did not investigate the exercise habits of the participants. Marcus et al[33] reported that resistance exercise can decrease intermuscular adipose tissue in older individuals. Further studies with other devices and prospective follow-up studies will be needed to validate the utility of the predictive formula.

In conclusion, this work showed the influence of aging on the lower limbs, such as decreased muscle strength and thickness, by using sonography. The proposed index of muscle strength outlined in this article can quantitatively assess muscular morphologic changes due to aging and may be a valuable tool for early detection of musculoskeletal disorders.

References

1. Cruz-Jentoft AJ, Landi F, Schneider SM, et al. Prevalence of and interventions for sarcopenia in ageing adults: a systematic review. Report of the International Sarcopenia Initiative (EWGSOP and IWGS). *Age Ageing* 2014; 43:748–759.
2. Cruz-Jentoft AJ, Baeyens JP, Bauer JM, et al. Sarcopenia: European consensus on definition and diagnosis: Report of the European Working Group on Sarcopenia in Older People. *Age Ageing* 2010; 39:412–423.
3. Goodpaster BH, Carlson CL, Visser M, et al. Attenuation of skeletal muscle and strength in the elderly: the Health ABC Study. *J Appl Physiol* 2001; 90:2157–2165.
4. Kent-Braun JA, Ng AV, Young K. Skeletal muscle contractile and noncontractile components in young and older women and men. *J Appl Physiol* 2000; 88:662–668.
5. Reimers K, Reimers CD, Wagner S, Paetzke I, Pongratz DE. Skeletal muscle sonography: a correlative study of echogenicity and morphology. *J Ultrasound Med* 1993; 12:73–77.
6. Arts IM, Pillen S, Schelhaas HJ, Overeem S, Zwarts MJ. Normal values for quantitative muscle ultrasonography in adults. *Muscle Nerve* 2010; 41:32–41.
7. Cadore EL, Izquierdo M, Conceicao M, et al. Echo intensity is associated with skeletal muscle power and cardiovascular performance in elderly men. *Exp Gerontol* 2012; 47:473–478.
8. Fukumoto Y, Ikezoe T, Yamada Y, et al. Skeletal muscle quality assessed from echo intensity is associated with muscle strength of middle-aged and elderly persons. *Eur J Appl Physiol* 2012; 112:1519–1525.
9. Harris-Love MO, Seamon BA, Teixeira C, Ismail C. Ultrasound estimates of muscle quality in older adults: reliability and comparison of Photoshop and ImageJ for the grayscale analysis of muscle echogenicity. *PeerJ* 2016; 4:e1721.
10. Ismail C, Zabal J, Hernandez HJ, et al. Diagnostic ultrasound estimates of muscle mass and muscle quality discriminate between women with and without sarcopenia. *Front Physiol* 2015; 6:302.
11. Watanabe T, Terabayashi N, Fukuoka D, et al. A pilot study to assess fatty infiltration of the supraspinatus in patients with rotator cuff tears: comparison with magnetic resonance imaging. *Ultrasound Med Biol* 2015; 41:1779–1783.
12. Watanabe Y, Yamada Y, Fukumoto Y, et al. Echo intensity obtained from ultrasonography images reflecting muscle strength in elderly men. *Clin Interv Aging* 2013; 8:993–998.
13. Haralick RM, Shanmugam K. *Land Use Classification Using Texture Information in ERTS-1 MSS Imagery*. Springfield, VA: National Technical Information Service; 1973. NTIS E73–10461.
14. Gomez W, Pereira WC, Infantosi AF. Analysis of co-occurrence texture statistics as a function of gray-level quantization for classifying breast ultrasound. *IEEE Trans Med Imaging* 2012; 31: 1889–1899.
15. Shen Y, Zhang YH, Shen L. Postmenopausal women with osteoporosis and osteoarthritis show different microstructural characteristics of trabecular bone in proximal tibia using high-resolution magnetic resonance imaging at 3 Tesla. *BMC Musculoskelet Disord* 2013; 14:136.
16. Molinari F, Caresio C, Acharya UR, Mookiah MR, Minetto MA. Advances in quantitative muscle ultrasonography using texture analysis of ultrasound images. *Ultrasound Med Biol* 2015; 41:2520–2532.
17. Albregtsen F. *Statistical Texture Measures Computed From Gray Level Co-occurrence Matrices*. Oslo, Norway: Department of Informatics, University of Oslo; 2008; 5:1–14.
18. Shanthi SR. Ultrasound image classification for Down syndrome during first trimester using Haralick features. *Int J Eng Technol* 2014; 6: 781–788.
19. Rubenstein LZ. Falls in older people: epidemiology, risk factors and strategies for prevention. *Age Ageing* 2006; 35(suppl 2):ii37–ii41.
20. Ogawa M, Yasuda T, Abe T. Component characteristics of thigh muscle volume in young and older healthy men. *Clin Physiol Function Imaging* 2012; 32:89–93.
21. Cree MG, Newcomer BR, Katsanos CS, et al. Intramuscular and liver triglycerides are increased in the elderly. *J Clin Endocrinol Metab* 2004; 89:3864–3871.
22. Koster A, Ding J, Stenholm S, et al. Does the amount of fat mass predict age-related loss of lean mass, muscle strength, and muscle quality in older adults? *J Gerontol A Biol Sci Med Sci* 2011; 66:888–895.
23. Pillen S, van Dijk JP, Weijers G, Raijmann W, de Korte CL, Zwarts MJ. Quantitative gray-scale analysis in skeletal muscle ultrasound: a comparison study of two ultrasound devices. *Muscle Nerve* 2009; 39: 781–786.
24. Hiraoka A, Aibiki T, Okudaira T, et al. Muscle atrophy as pre-sarcopenia in Japanese patients with chronic liver disease: computed tomography is useful for evaluation. *J Gastroenterol* 2015; 50:1206–1213.
25. Durand F, Buyse S, Francoz C, et al. Prognostic value of muscle atrophy in cirrhosis using psoas muscle thickness on computed tomography. *J Hepatol* 2014; 60:1151–1157.
26. Baumgartner RN, Koehler KM, Gallagher D, et al. Epidemiology of sarcopenia among the elderly in New Mexico. *Am J Epidemiol* 1998; 147:755–763.
27. Tanimoto Y, Watanabe M, Sun W, et al. Association between sarcopenia and higher-level functional capacity in daily living in community-dwelling elderly subjects in Japan. *Arch Gerontol Geriatr* 2012; 55:e9–e13.
28. Nelson TR, Fowlkes JB, Abramowicz JS, Church CC. Ultrasound biosafety considerations for the practicing sonographer and sonologist. *J Ultrasound Med* 2009; 28:139–150.
29. Manini TM, Visser M, Won-Park S, et al. Knee extension strength cutpoints for maintaining mobility. *J Am Geriatr Soc* 2007; 55:451–457.
30. Inagaki A, Yatsushiro R, Hirai M, et al. Physical fitness test at periodical health examination based on the "Law of Health and Welfare for

the Elderly": necessity and test battery [in Japanese]. *Jpn J Nurs Health Sci* 2005; 6:2–15.

31. Ohmori H, Kanaoka Y, Murata Y, et al. Deep vein thrombosis in patients with severe motor and intellectual disabilities, especially diagnosis and prevention of recurrence for chronic thrombosis: serial changes of sonography and D-dimer. *Ann Vasc Dis* 2015; 8:290–296.
32. Yamada N, Nakamura M, Ishikura K, et al. Epidemiological characteristics of acute pulmonary thromboembolism in Japan. *Int Angiol* 2003; 22:50–54.
33. Marcus RL, Addison O, Kidde JP, Dibble LE, Lastayo PC. Skeletal muscle fat infiltration: impact of age, inactivity, and exercise. *J Nutr Health Aging* 2010; 14:362–366.