Changes in Muscle Thickness and Echo Intensity in Chronic Stroke Survivors: A 2-Year Longitudinal Study

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**Background and Purpose** The objective of this study was to identify 2-year longitudinal changes in the muscle thickness (MT) and echo intensity (EI) of the abdominal, thigh, and lower limb muscles in chronic stroke survivors.

**Methods** This study included 15 chronic stroke survivors aged 74.1 ± 9.9 years. The MT, EI, and subcutaneous fat thickness values of the following muscles on the paretic and nonparetic sides were assessed on transverse ultrasound images: rectus abdominis, external oblique, internal oblique, transversus abdominis, rectus femoris (RF), vastus intermedius, vastus lateralis (VL), vastus medialis, tibialis anterior, gastrocnemius, and soleus. The ultrasound measurements were performed both at baseline and 2 years later.

**Results** After 2 years, the VL on the paretic side showed a significant decrease in MT ($p = 0.031$) and increase in EI ($p = 0.002$), whereas the RF on the nonparetic side showed a significant decrease in EI ($p = 0.046$). Correlation coefficient analyses showed that changes in MT ($r = 0.668, p = 0.012$) and EI ($r = 0.597, p = 0.018$) of the VL on the paretic side were significantly associated with a change in the body mass index.

**Conclusions** The findings of this longitudinal study suggest that the VL on the paretic side is subject to deteriorations in muscle quantity and quality, and conversely that the RF on the nonparetic side shows an improvement in muscle quality after 2 years in chronic stroke survivors.

**Keywords** stroke; muscle mass; intramuscular fat; ultrasound; body mass index.

**INTRODUCTION**

Stroke induces sarcopenia and specific changes in the quantity and quality of muscle. Several previous studies have found that the mass and quantity of muscle were significantly lower in the paretic lower limb than in the nonparetic limb. In addition, changes in muscle quality, such as increased intramuscular fat content and fibrous tissue, were observed. Both the decreased muscle mass and changes in muscle quality were related to reduced muscle strength and loss of gait independence in chronic stroke survivors. Therefore, both muscle quantity and quality play crucial roles in motor function changes in chronic stroke survivors.

Cross-sectional studies have used computed tomography and ultrasonography to determine the changes in muscle quantity and quality in stroke survivors. The paretic side was found to exhibit negative changes in muscle quantity and quality compared with the nonparetic side or healthy controls, with these changes varying between different anatomical sites.

It has also been reported that resistance training can increase muscle mass and decrease intramuscular fat in stroke survivors and healthy individuals. Longitudinal changes in...
Muscle quantity and quality need to be investigated at different anatomical sites in order to ensure effective exercise therapy in chronic stroke survivors. Hence, this study aimed to identify longitudinal changes in muscle quantity and quality of the abdominal, thigh, and lower limb muscles after stroke by using ultrasound.

**METHODS**

This longitudinal study recruited community-dwelling stroke survivors who were undergoing rehabilitation funded by the Japanese long-term-care insurance system in Hyogo Prefecture from June to September 2017. The inclusion criteria were independent ambulation, at least 6 months after stroke, and unilateral stroke. The exclusion criteria were orthopedic or chronic pain conditions, dementia, or other neuromuscular diseases.

Data were collected at baseline from 32 patients with chronic stroke after they were informed about the study procedures and had provided written informed consent. Follow-up data were collected from June to September 2019 at the same place. Of the 32 participants included at baseline, 17 participants were lost to follow-up due to 2 refusing further participation, 7 withdrawing from the rehabilitation program, and 8 becoming ill. Follow-up data were collected from the remaining 9 males and 6 females, who were aged 74.1±9.9 years with a height of 158.2±8.1 cm, weight of 56.1±7.1 kg, and latency of 71.8±54.3 months from the stroke onset at baseline. Paralysis was assessed using Fugl–Meyer (LE-FM) evaluations of the lower extremities. The protocol was approved by the ethics committee of Kobe Gakuin University Graduate School (IRB No. HEB20151202-1).

**Ultrasound measurements**

The ultrasound measurements performed in this study have been described previously. Briefly, the following individual muscles on both the paretic and nonparetic sides were measured using B-mode ultrasound imaging (LOGIQ e, GE Healthcare UK, Chalfont, Buckinghamshire, England) with a multifrequency linear transducer operating at 8–12 MHz: rectus abdominis, external oblique (EO), internal oblique (IO), transversus abdominis, rectus femoris (RF), vastus intermedius, vastus lateralis (VL), vastus medialis, tibialis anterior (TA), gastrocnemius, and soleus. The measurement position and site for each muscle are listed in Table 1. Two consecutive ultrasound images of each muscle were obtained. On the ultrasound images, muscle thickness (MT) was used as the indicator of muscle quantity, and echo intensity (EI) was used as the indicator of muscle quality. EI was evaluated using 256 grayscale analysis with Image-J (version 1.37, U.S. National Institutes of Health, Bethesda, MD, USA), and was expressed as a value between 0 (black) and 255 (white). The regions of interest in which EI was measured included the individual muscles as much as possible while avoiding the surrounding muscle fascia. Unclear sections were excluded from the region of interest for EI measurements. In addition, the subcutaneous fat thickness (SFT) was assessed on each ultrasound image. The mean MT, EI, and SFT values calculated from the two obtained images were used in further analyses. All ultrasound measurements both at baseline and 2 years later were made by the same investigator in order to avoid interobserver variations.

**Statistical analyses**

We calculated the change in each of MT, EI, body mass index (BMI), and LE-FM score as the value after 2 years minus the baseline value. When data appeared to conform to a normal distribution on a histogram, this was confirmed using the Shapiro-Wilk test. The 2-year longitudinal changes in MT, EI, and SFT on both the paretic and nonparetic sides were compared.

**Table 1. Measurement sites and positions for the trunk and lower limb muscles**

| Muscles       | Positions | Measurement sites                                                                 |
|---------------|-----------|-----------------------------------------------------------------------------------|
| Rectus abdominis | Supine    | 3 cm lateral to the umbilicus                                                    |
| External oblique | Supine    | 2.5 cm anterior to the mid-umbilicus line, at the midpoint between the inferior rib and the iliac crest |
| Internal oblique | Supine    | 2.5 cm anterior to the mid-umbilicus line, at the midpoint between the inferior rib and the iliac crest |
| Transversus abdominis | Supine | 2.5 cm anterior to the mid-umbilicus line, at the midpoint between the inferior rib and the iliac crest |
| Rectus femoris   | Supine    | Midway between the anterior superior iliac supine and the proximal end of the patella |
| Vastus intermedius | Supine    | Midway between the anterior superior iliac supine and the proximal end of the patella |
| Vastus lateralis  | Supine    | Midway between the great trochanter and lateral condyle of the tibia             |
| Vastus medialis   | Supine    | 30% proximal between the great trochanter and lateral condyle of the tibia       |
| Tibialis anterior | Supine    | 30% proximal between the lateral malleolus of the fibula and lateral condyle of the tibia |
| Gastrocnemius     | Sitting   | Medial head of gastrocnemius at 30% proximal between the lateral malleolus of the fibula and the lateral condyle of the tibia |
| Soleus           | Sitting   | 30% proximal between the lateral malleolus of the fibula and lateral condyle of the tibia |
using paired t-tests for normally distributed data and the Wilcoxon signed-rank test for nonnormally distributed data. The effect size was calculated as Cohen’s d for normally distributed data and as the correlation coefficient (r) for nonnormally distributed data.

Correlation analysis was performed to investigate the relationship between MT and EI at baseline and at the 2-year follow-up. For MT and EI values that had changed significantly after 2 years, the relationships between the changes in MT and EI values with those in BMI and LE-FM score were investigated. Spearman’s correlation analysis was applied to normally distributed data and Pearson’s product-moment correlation analyses was applied to nonnormally distributed data. The criterion for statistical significance was defined as p<0.05. Statistical analyses were performed using SPSS (version 20.0, SPSS Japan Incorporated, Tokyo, Japan).

**RESULTS**

Table 2 lists the characteristics at baseline and at the 2-year follow-up of all participants. The LE-FM score was significantly higher after 2 years than at baseline (p=0.002), whereas the BMI was unchanged.

Tables 3 and 4 list the changes in MT and EI values on both the paretic and nonparetic sides between baseline and 2 years later. MT had decreased significantly after 2 years only for the paretic VL (p=0.021, Cohen’s d=0.52), while the EI value for the paretic VL increased significantly after 2 years (p=0.010, Cohen’s d=0.65). Conversely, the EI value for the nonparetic RF significantly decreased after 2 years (p=0.046, Cohen’s d=0.50). None of the SFT values changed significantly over the 2-year study period. Fig. 1 presents ultrasound images of the paretic VL at baseline and 2 years later.

Correlation analyses revealed that EO on the nonparetic side at baseline and TA on the nonparetic side at the 2-year follow-up were significantly correlated with MT and EI. The changes in MT and EI for the paretic VL and those in EI for the nonparetic RF were not significantly associated with each other. In addition, the changes in MT and EI for the VL on the paretic side were significantly associated with BMI changes (r=0.668, p=0.012 for MT; r=0.597, p=0.018 for EI) but not with changes in the LE-FM score (Fig. 2). The changes in EI for the nonparetic RF were not significantly associated with changes in BMI or the LE-FM score.

**DISCUSSION**

To the best of our knowledge, this study is the first to investigate longitudinal changes in the individual MT and EI values of chronic stroke survivors. The primary findings of this study were that the VL was the only paretic muscle exhibiting decreases in muscle quantity and quality during the 2-year observation period. In addition, these changes were significantly and positively correlated with BMI changes, suggesting that

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**Table 2.** Characteristics from baseline to the 2-year follow-up

| Characteristic          | Baseline | Follow-up | p   |
|-------------------------|----------|-----------|-----|
| Age (yr)                | 74.1±9.9 | 76.1±9.9  | -   |
| Sex (male/female)       | 21/11    | 9/6       | -   |
| BMI (kg/m²)             | 22.4±2.6 | 22.4±2.7  | 0.967|
| LE-FM score             | 14.7±4.4 | 16.3±4.2  | 0.002|
| Latency (month)         | 75.5±61.4| 99.5±61.4 | -   |

Data are presented as mean±standard deviation. BMI, body mass index; LE-FM, lower extremity Fugl-Meyer.

**Table 3.** Changes in muscle thicknesses at baseline and at the 2-year follow-up (n=15)

| Muscle | Paretic side (cm) | Nonparetic side (cm) |
|--------|-------------------|----------------------|
|        | Baseline | Follow-up | p | Change (%) | Effect size | Baseline | Follow-up | p | Change (%) | Effect size |
| RA     | 0.72±0.15 | 0.72±0.16 | 0.961 | 0.00±0.15 (-2.3) | 0.00* | 0.74±13.8 | 0.72±0.15 | 0.567 | -0.02±0.13 (-4.3) | 0.14* |
| EO     | 0.39±0.13 | 0.44±0.14 | 0.074 | 0.06±0.11 (11.1) | 0.39* | 0.41±0.12 | 0.43±0.12 | 0.536 | 0.02±0.09 (1.4) | 0.17* |
| IO     | 0.64±0.20 | 0.62±0.21 | 0.683 | -0.02±0.11 (-4.7) | 0.12* | 0.60±0.13 | 0.61±0.18 | 0.910 | 0.01±0.10 (-0.9) | 0.22* |
| TrA    | 0.27±0.05 | 0.29±0.08 | 0.629 | 0.03±0.09 (3.5) | 0.13* | 0.27±0.07 | 0.28±0.06 | 0.530 | 0.01±0.05 (3.4) | 0.17* |
| RF     | 1.71±0.32 | 1.56±0.38 | 0.132 | -0.61±0.36 (-15.8) | 0.45* | 1.74±0.33 | 1.78±0.32 | 0.715 | 0.04±0.43 (-0.3) | 0.13* |
| VI     | 1.22±0.31 | 1.20±0.30 | 0.691 | -0.02±0.30 (-5.1) | 0.12* | 1.67±0.47 | 1.47±0.39 | 0.177 | -0.20±0.52 (-19.6) | 0.47* |
| VL     | 1.56±0.32 | 1.40±0.37 | 0.021* | -0.18±0.27 (-18.5) | 0.52* | 1.92±0.36 | 1.74±0.39 | 0.072 | -0.18±0.36 (-14.1) | 0.49* |
| VM     | 1.25±0.33 | 1.17±0.33 | 0.289 | -0.08±0.28 (-13.9) | 0.25* | 1.47±0.22 | 1.41±0.26 | 0.344 | -0.06±0.22 (-5.9) | 0.26* |
| TA     | 1.70±0.21 | 1.73±0.25 | 0.544 | -0.05±0.33 (-5.4) | 0.27* | 1.90±0.20 | 1.85±0.27 | 0.640 | -0.04±0.35 (-4.7) | 0.22* |
| Gas    | 1.32±0.39 | 1.18±0.26 | 0.112 | -0.13±0.30 (-6.0) | 0.43* | 1.41±0.39 | 1.34±0.27 | 0.164 | -0.14±0.33 (-6.9) | 0.37* |
| Sol    | 1.44±0.18 | 1.37±0.27 | 0.401 | -0.07±0.31 (-9.4) | 0.31* | 1.58±0.29 | 1.57±0.30 | 0.947 | -0.01±0.33 (-3.3) | 0.04* |

Data are presented as mean±standard deviation. *Effect size calculated as Cohen’s d; †Effect size calculated as correlation coefficient (r); ‡Statistically significant.

EO, external oblique; Gas, gastrocnemius; IO, internal oblique; RA, rectus abdominis; RF, rectus femoris; Sol, soleus; TA, tibialis anterior; TrA, transversus abdominis; VI, vastus intermedius; VL, vastus lateralis; VM, vastus medialis.
both MT and EI for the VL on the paretic side increase with BMI.

Our previous cross-sectional study found that the decrease in MT and increase in EI were greatest in quadriceps muscles among the paretic muscles compared with the nonparetic side. The results of the current longitudinal study support those of that cross-sectional study, and suggest that the VL could be more susceptible to loss of muscle mass and the accumulation of both fat and other noncontractile tissues within the muscle than are the other quadriceps muscles over the time course of stroke. There are previous reports that adipose differentiation is promoted in association with muscle atrophy. The findings of the present study suggest that in chronic stroke survivors, atrophy of the VL on the paretic side occurs over time and is accompanied by an increase in intramuscular fat.

It is particularly noteworthy that the EI value of the nonparetic RF decreased significantly over 2 years while the MT of the nonparetic RF might increase over >2 years. Because the MT did not change in the present study, the relative proportion of actual contractile tissues within the RF might increase with decreases in fat and

Table 4. Changes in echo intensity at baseline and at the 2-year follow-up (n=15)

| Muscle | Paretic side (a.u.) | Nonparetic side (a.u.) |
|--------|---------------------|-----------------------|
|        | Baseline | Follow-up | Effect size | Change (%) | Baseline | Follow-up | Effect size | Change (%) |
| RA     | 94.6±20.1 | 103.3±29.7 | 0.36* | 8.7±26.2 | (3.2) | 93.5±19.7 | 101.6±22.5 | 0.057 | 8.1±14.6 | (6.2) | 0.40* |
| EO     | 86.2±22.9 | 87.5±14.8 | 0.07* | 1.3±16.8 | (1.7) | 92.6±9.7 | 82.6±13.1 | 0.055 | -9.9±19.7 | (-17.8) | 0.50* |
| IO     | 74.0±17.2 | 76.6±18.5 | 0.15* | 2.5±11.8 | (1.4) | 77.6±17.2 | 74.7±12.6 | 0.200 | -2.8±7.6 | (-3.1) | 0.19* |
| TrA    | 73.8±20.4 | 77.6±20.1 | 0.20* | 3.7±17.7 | (1.7) | 74.4±13.8 | 73.1±12.4 | 0.562 | -1.3±8.3 | (-2.1) | 0.10* |
| RF     | 78.0±12.9 | 74.0±13.8 | 0.32* | -4.0±10.8 | (-6.9) | 73.2±12.6 | 64.8±20.0 | 0.046* | -8.3±14.2 | (-20.4) | 0.50* |
| VI     | 87.5±9.8 | 86.1±12.7 | 0.18* | -1.5±12.4 | (-3.0) | 75.9±10.1 | 74.1±9.1 | 0.515 | -1.8±10.0 | (-3.4) | 0.19* |
| VL     | 69.8±15.2 | 78.1±15.5 | 0.010* | 8.2±8.4 | (10.2) | 64.9±21.6 | 61.6±16.2 | 0.561 | -3.2±17.2 | (-7.5) | 0.16* |
| VM     | 77.6±19.4 | 74.9±18.0 | 0.15* | -2.8±14.5 | (-5.8) | 70.4±16.6 | 63.8±13.8 | 0.107 | -6.6±17.2 | (-14.9) | 0.45* |
| TA     | 66.4±14.5 | 64.2±11.8 | 0.16* | -2.1±12.7 | (-4.9) | 63.5±11.9 | 59.3±11.9 | 0.314 | -4.2±15.1 | (-11.6) | 0.37* |
| Gas    | 71.8±11.0 | 71.5±11.5 | 0.03* | -0.3±11.5 | (-1.9) | 61.5±15.1 | 60.2±13.7 | 0.760 | -1.2±14.9 | (-5.0) | 0.09* |
| Sol    | 73.2±9.2 | 70.0±7.9 | 0.38* | -3.1±10.9 | (-5.4) | 61.7±12.4 | 65.5±10.3 | 0.639 | 3.8±18.0 | (2.9) | 0.35* |

Data are presented as mean±standard deviation. *Effect size calculated as Cohen's d; †Effect size calculated as r; ‡statistically significant.
a.u., arbitrary units; EO, external oblique; Gas, gastrocnemius; IO, internal oblique; RA, rectus abdominis; RF, rectus femoris; Sol, soleus; TA, tibialis anterior; TrA, transversus abdominis; VI, vastus intermedius; VL, vastus lateralis; VM, vastus medialis.

Fig. 1. Changes in muscle thickness and echo intensity. VL, vastus lateralis.
connective tissues. Unfortunately, the reason for the lack of significant changes in the MT and EI of the RF on the paretic side of the VL on the nonparetic side remains unclear. Further studies are needed to clarify the mechanism underlying the muscle-specific differences in changes in MT and EI in the quadriceps based on neurophysiological and kinesiological factors.

The negative change in the paretic quadriceps (decreased MT and increased EI of the VL) and the positive change in the nonparetic quadriceps (decreased EI of the RF) probably represented long-term compensatory strategies in the nonparetic lower limb during daily activities. Physical activity has been reported to be associated with the muscle quantity and quality of the quadriceps femoris in older adults and stroke survivors. Therefore, asymmetry caused by long-term compensation contributes to overactivity of the nonparetic leg muscles and disuse of the paretic leg muscles, thereby affecting the muscle quantity and quality in chronic stroke survivors. The present study did not evaluate the weight distribution between the limbs or the muscle activity during standing and walking, and so further studies are needed to clarify the relevance of these aspects.

Changes in both the MT and EI values of the paretic VL were positively associated with changes in BMI. In a longitudinal study involving 1,678 older adults, Delmonico et al. found that 5-year changes in both the cross-sectional area and muscle fat infiltration of the thigh measured using computed tomography appeared to be positively associated with body weight changes. In another longitudinal study, multiple regression analyses identified age, weight change, and baseline weekly physical activity as predictors of changes in fat mass, while the group with increased body weight had increased fat mass. These findings suggest that there is a positive relationship between weight gain and increased fat mass. Those results might be consistent with the present finding of longitudinal changes in the intramuscular fat being positively associated with those in BMI in chronic stroke survivors.

The mean LE-FM score of the participants in this study improved significantly by 1.6 points over 2 years, whereas they experienced a loss of MT and an increase in EI of the paretic VL. The improvement in the LE-FM score was probably attributable to the rehabilitation services that they received at the daycare center. However, Pandian et al. indicated that a change in LE-FM score of 6 points is the smallest that is clinically significant in chronic stroke survivors, and so the increase in LE-FM score of only 1.6 points in our participants would not have been clinically beneficial.

The present study had some limitations. First, the study included only a small number of participants and had a relatively high dropout rate. Compared with the follow-up cohort, although there were no differences in the LE-FM score, MT, or EI of the quadriceps on both sides, the baseline age tended to be lower (p=0.085) at baseline than in those who did not participate in the follow-up assessment. Second, selection bias might have occurred because the participants in this study were recruited while receiving rehabilitation services funded by the Japanese long-term-care insurance system. Third, we only measured the LE-FM score as an indicator of physical function, and not muscle strength. To clarify the relationship of longitudinal changes in MT and EI with those in physical function in stroke survivors, muscle strength and other function-related parameters should be measured in the future.

In conclusion, 2-year longitudinal negative changes in mus-
cle quantity and quality occurred in the paretic VL, whereas a positive change occurred in the muscle quality of the non-paretic RF. In addition, the 2-year changes in the muscle quantity and quality of the VL on the paretic side were positively associated with that in BMI, while BMI remained unchanged over this 2-year period. These findings suggest that evaluations and exercise should focus on the paretic VL among the abdominal and lower limb muscles. In addition, the VL should be cautiously evaluated because changes in the quantity and quality of that muscle may occur even when BMI remains constant in chronic stroke survivors.

Availability of Data and Material
Data sharing not applicable to this article as no datasets were generated or analyzed during the study.

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Conflicts of Interest
The authors have no potential conflicts of interest to disclose.

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