
REPORT

Accuracy of tissue thickness of the rectus femoris region as measured by ultrasound

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Abstract In order to verify the accuracy of the thickness of the rectus femoris muscle and overlying subcutaneous fat as assessed by ultrasound, experiments were conducted on 10 healthy subjects aged over 30 years to assess the following : 1) concordance of measurements made by two different observers ; 2) concordance of repeated measurements made by the same observer ; and 3) concordance of repeated measurements made by the same observer with and without an imaging aid (ultrasound gel block).

Between two observers with varying experience in ultrasound, no significant difference in measured muscle thickness was observed ; however, there was a significant difference in subcutaneous fat thickness. When measurements were made by the same observer on two separate days, a significant positive correlation was evident for muscle and subcutaneous fat thickness between the first and second tests. While use of the imaging aid was associated with higher concordance of measurements between the first and second tests, the measurements themselves were affected.

Although ultrasonic measurement of tissue thickness requires further investigation to minimize intra-observer error and examine correlations with other imaging techniques, the accuracy of the measurements in the present study suggests that clinical application may be possible.

Key words : leg muscle thickness , leg subcutaneous fat thickness, ultrasonography

Introduction

In order to objectively assess the body in clinical settings, various machines are used to quantify body composition. At present, nurses often use noninvasive machines for clinical and research purposes.

Imaging techniques that quantify and assess body composition in a noninvasive manner include CT¹⁻⁴⁾, DXA^{5,6)},

bioimpedance analysis⁷⁾ and ultrasonography^{8,9)}. While CT is useful in quantitatively assessing muscle tissue, repeated measurements expose subjects to high levels of radiation, and not every region of the body can be scanned. Similarly, DXA cannot be performed in every region. With body impedance, it is difficult to accurately estimate percent body fat and lean body mass if body weight cannot be measured.

On the other hand, ultrasound is noninvasive measurement can be repeatedly measured to the same patient. Also, the greatest advantage is being able to carry it anywhere. Studies using human cadavers have shown

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that tissue thickness as assessed by ultrasound and as measured by anatomic dissection differ by $< 5\%$ ^{10,11}.

Ultrasonography has been performed to assess not only body composition^{8, 9, 12}, but also muscle tissue in fields such as rehabilitation^{13,14} and sports medicine^{14,15}. Hence, health care workers in addition to physicians use ultrasonography. While there have few studies on the use of ultrasonography in the field of nursing, we have reported the use of ultrasonography by nurses in the assessment of leg muscles¹⁶.

The use of ultrasonography by nurses as an objective assessment method facilitates the evidence-based verification of nursing practices and evaluation of nursing care. When using any machine, it is necessary to confirm validity, to investigate measurement methods that are suitable for target patients, and to evaluate reliability. Because leg muscles are one of the most important muscle groups for mobility in daily living, investigating methods to measure leg tissue thickness are very relevant for assessment of nursing practice.

In order to facilitate the clinical use of ultrasonography by nurses, the present study investigated the reliability of ultrasonographic measurement of leg tissue thickness in subjects in the recumbent position.

Methods

In this study, the accuracy of tissue thickness of the rectus femoris muscle as measured by ultrasound was assessed in terms of : 1) concordance of measurements made by two different observers ; 2) concordance of repeated measurements made by the same observer ; and 3) concordance of repeated measurements made by the same observer with and without an imaging aid.

1. Observers

In Experiment 1, two observers took measurements (the author was one of these two observers), and in Experiments 2 and 3, the author alone took measurements.

2. Subjects

In Experiment 1, subjects were five healthy women aged over 30 years (mean : 44.6 ± 11.6 years), and in Experiments 2 and 3, subjects were ten healthy indi-

viduals (3 men and 7 women) aged over 30 years (mean : 45.1 ± 11.6 years).

3. Ethical considerations

Prior to the study, informed consent was obtained after orally explaining the study objectives and the following : 1) participation in the study was voluntary ; 2) consent could be withdrawn at any time ; 3) the testing equipment was not harmful to the human body ; 4) the skin would be marked using a permanent marker ; and 5) measurements would be used in a scientific study.

4. Study period and location

The study was conducted from April 2001 to March 2003 at a room used for practical training at the medical technology college of a university.

5. Measurement methods

1) Determination of test site and tissue differentiation

Ultrasound was performed using a diagnostic ultrasound machine (Rtfino[®]: GE Yoga Medical System) and probe (LP probe). The oscillating frequency was set at 7.5 MHz so that the boundary between subcutaneous fat and muscle could be clearly differentiated⁹.

Muscle and subcutaneous fat tissues were differentiated according to the method of Fukunaga et al.^{11,17} : the thickness of subcutaneous fat was defined as the distance from the contact surface between the skin and probe to the boundary with the muscle (fascia), while the thickness of muscle tissue was defined as the distance from the fascia to the bone. The thickness of subcutaneous fat and muscle was measured as shown in Figure 1.

The ultrasound probe was placed over the rectus femoris of the dominant leg. This muscle was selected because, being one of the femoral extensor muscles, it is most likely to be affected by recumbency ; a study showed that thickness of this muscle decreased the most following laparotomy¹⁸. The probe was placed at the midpoint between the greater trochanter and the lateral condyle of the tibia, where cross-sectional area of the thigh was greatest¹¹. In addition, because concordance of measurements was evaluated, it was necessary to measure thickness at the same site. Therefore, with approval of each subject, a mark was placed using a permanent marker.

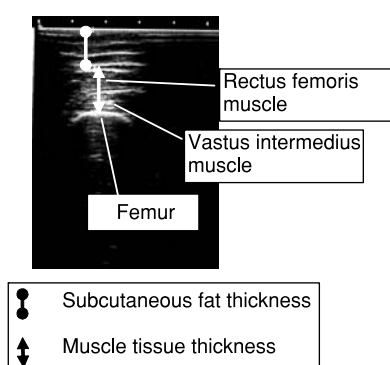


Figure 1 . An ultrasound image of the rectus femoris region

2) Body and limb positions

In previous basic methodologic studies on ultrasound measurement^{10,19)}, subjects stood upright so that measurements could be taken at various points on the body. However, many patients requiring nursing care cannot stand. Consequently, in this study, measurements were taken in the supine position with the legs extended (Figure 2).



Figure 2 . Leg position

3) Measurement procedures and count

In order to avoid the effects of walking and to rest muscles, each subject was asked to lie down in bed for 10 minutes before all experiments. In Experiment 1, alternative measurements were made on the same day by an expert ultrasound technician (observer A) and the author, having one year of ultrasound experience (observer B). In Experiment 2, three measurements were made by observer B on two separate days. The second test was conducted about seven days later. The two tests were conducted at the same time of the day, and because observer B only had one year of experience, three measurements were made in this experiment, instead of two, as was the case with Experiment 1.

In Experiment 3, in order to disperse the pressure on the tissue, SonarGel[®] (Toshiba; Width : 10.0 cm, Length : 20.0 cm, Height : 1.0 cm, Weight : 220.0g) was used as an imaging aid. SonarGel[®] (SonarGel) is made of a material called macromolecular gel that improves the imaging accuracy of surface areas (Figure 3). On both sides of SonarGel, ultrasound gel was applied, and the probe was placed on top to make measurements. Observer B took measurements on two separate days, and the second test was conducted about seven days later. As was the case with Experiment 2, three measurements were made with and without SonarGel.

Measurement conditions were standardized by paying attention to the following : 1) the gel-covered probe was always positioned perpendicular to the skin surface ; 2) the probe was lightly applied to the skin ; and

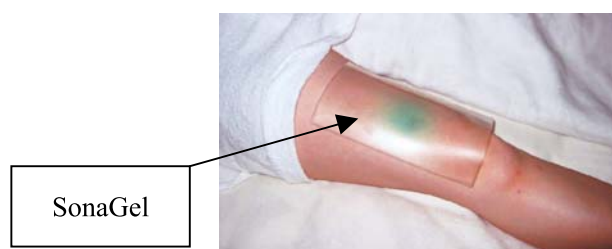


Figure 3 . Thickness measurement with SonaGel

3) subjects were asked to relax and to avoid contraction of the leg muscles.

Analysis methods

In Experiment 1, two measurements were made by each observer, and mean subcutaneous fat and muscle thicknesses and difference between measurements by the two observers were calculated. Next, for each observer, the median value was calculated, and a Wilcoxon signed-rank test was used to analyze inter-observer difference.

In Experiment 2, three measurements were made on each of two separate days, and mean muscle and subcutaneous fat thicknesses and difference between the first and second measurements were calculated. Next, after ranking the mean value for the first and second measurements, a Spearman rank correlation test was used to

analyze concordance.

In Experiment 3, three measurements were made with SonarGel, and three were made without. Mean muscle and subcutaneous fat thicknesses and difference between the first and second measurements were calculated. Next, after ranking the mean value for the first and second tests with or without SonarGel, a Spearman's rank correlation test was used to analyze concordance.

Statistical analyses were carried out using SPSS (Version 9.0) with the level of significance set at $p < 0.05$.

Results

1. Concordance of measurements made by two observers

Mean muscle thickness measured by observer A was 3.03 ± 0.64 cm, while that measured by observer B was 2.94 ± 0.61 cm. The difference between measurements taken by the two observers was 0.09 cm (not statistically significant, N.S., Table 1).

Mean subcutaneous fat thickness measured by observer A was 1.14 ± 0.19 , while that measured by observer B was 1.25 ± 0.18 cm. The difference between measurements taken by the two observers was 0.11 cm ($p = 0.042$, Table 1).

2. Concordance of measurements made by the same observer on two separate days

Mean muscle thickness was measured at 2.54 ± 0.45

cm in the first test vs. 2.56 ± 0.38 cm in the second test; a difference of 0.02 cm. Mean subcutaneous fat thickness was measured at 1.00 ± 0.31 cm in the first test vs. 1.04 ± 0.32 cm in the second test; a difference of 0.04 cm. For the ten subjects, the largest difference between the two tests was 0.35 cm for muscle thickness and 0.30 cm for subcutaneous fat thickness.

For both muscle and subcutaneous fat, a significant positive correlation was evident between the two tests (Table 2).

3. Concordance of measurements made with and without SonarGel

Without SonarGel, mean muscle thickness was measured at 2.63 ± 0.47 cm in the first test vs. 2.60 ± 0.47 cm in the second test; a difference of 0.03 cm. Mean subcutaneous fat thickness was measured at 1.15 ± 0.28 cm in the first test vs. 1.11 ± 0.28 cm in the second test; a difference of 0.04 cm.

With SonarGel, mean muscle thickness was measured at 2.49 ± 0.42 cm in the first test vs. 2.52 ± 0.42 cm in the second test; a difference of 0.03 cm. For mean subcutaneous fat thickness, no difference was apparent between the two tests; 1.21 ± 0.29 cm in both.

Of the ten subjects, the largest difference in muscle thickness between the two tests was 0.39 cm without SonarGel and 0.21 cm with SonarGel, while the largest difference in subcutaneous fat thickness was 0.12 cm without SonarGel and 0.10 cm with SonarGel. For both

Table 1. Comparison of measurements made by two different observers

| Subjects | Muscle thickness | | | Subcutaneous fat thickness | | |
|---------------------------|-----------------------------|-----------------------------|-----------------|-----------------------------|-----------------------------|-----------------|
| | Observer A Measurement (cm) | Observer B Measurement (cm) | Difference (cm) | Observer A Measurement (cm) | Observer B Measurement (cm) | Difference (cm) |
| a | 4.00 | 3.90 | 0.10 | 1.30 | 1.45 | 0.15 |
| b | 2.55 | 2.60 | 0.05 | 0.90 | 1.10 | 0.20 |
| c | 2.50 | 2.30 | 0.20 | 1.00 | 1.05 | 0.05 |
| d | 3.35 | 3.10 | 0.25 | 1.15 | 1.20 | 0.05 |
| e | 2.75 | 2.78 | 0.03 | 1.35 | 1.43 | 0.08 |
| Mean | 3.03 | 2.94 | 0.09 | 1.14 | 1.25 | 0.11 |
| SD | 0.64 | 0.61 | | 0.19 | 0.18 | |
| Wilcoxon signed-rank test | $Z = -1.214$ $p = 0.225$ | | | $Z = -2.032$ $p = 0.042$ | | |

Difference: Absolute difference between measurements made by observers A and B.

muscle and subcutaneous fat, SonarGel was associated with a smaller maximum difference.

For both muscle and subcutaneous fat, there was a significant positive correlation between the two measurements (Table 3), but the correlation coefficient was above 0.9 with SonarGel (muscle thickness : $r_s=0.948$;

subcutaneous fat thickness : $r_s=0.952$). Nonetheless, in almost all subjects, muscle thickness measured with SonarGel was lower than that measured without SonarGel, while the converse was true for subcutaneous fat thickness.

Table 2 . Comparison of measurements made on two different days by the same observer

| Subjects | Muscle thickness | | | Subcutaneous fat thickness | | |
|---|-----------------------------|------------------------------|-----------------|-----------------------------|------------------------------|-----------------|
| | First test Measurement (cm) | Second test Measurement (cm) | Difference (cm) | First test Measurement (cm) | Second test Measurement (cm) | Difference (cm) |
| a | 2.90 | 2.55 | 0.35* | 0.50 | 0.50 | 0.00 |
| b | 2.35 | 2.48 | 0.13 | 1.10 | 1.13 | 0.02 |
| c | 3.15 | 3.00 | 0.15 | 1.00 | 1.30 | 0.30* |
| d | 2.55 | 2.50 | 0.05 | 0.80 | 0.80 | 0.00 |
| e | 1.90 | 2.10 | 0.20 | 0.70 | 0.70 | 0.00 |
| f | 3.10 | 3.25 | 0.15 | 1.25 | 1.25 | 0.00 |
| g | 2.60 | 2.60 | 0.00 | 1.48 | 1.57 | 0.08 |
| h | 1.82 | 1.93 | 0.11 | 1.37 | 1.25 | 0.12 |
| i | 2.70 | 2.53 | 0.17 | 0.77 | 0.85 | 0.08 |
| J | 2.34 | 2.69 | 0.35 | 1.04 | 1.03 | 0.01 |
| Mean | 2.54 | 2.56 | 0.02 | 1.00 | 1.04 | 0.04 |
| SD | 0.45 | 0.38 | | 0.31 | 0.32 | |
| Spearman's rank correlation coefficient | $r_s=0.855$ $p<0.01$ | | | $r_s=0.863$ $p<0.01$ | | |

Table 3 . Comparison of measurements made by the same observer on two separate days with and without SonaGel

| Subjects | Muscle thickness | | | | | | Subcutaneous fat thickness | | | | | |
|--|-----------------------------|------------------------------|-----------------|-----------------------------|------------------------------|-----------------|-----------------------------|------------------------------|-----------------|-----------------------------|------------------------------|-----------------|
| | Without SonaGel | | | With SonaGel | | | Without SonaGel | | | With SonaGel | | |
| | First test Measurement (cm) | Second test Measurement (cm) | Difference (cm) | First test Measurement (cm) | Second test Measurement (cm) | Difference (cm) | First test Measurement (cm) | Second test Measurement (cm) | Difference (cm) | First test Measurement (cm) | Second test Measurement (cm) | Difference (cm) |
| a | 2.78 | 2.85 | 0.07 | 2.79 | 2.93 | 0.14 | 0.90 | 0.83 | 0.07 | 1.01 | 0.92 | 0.10* |
| b | 2.97 | 2.58 | 0.39* | 2.60 | 2.55 | 0.05 | 1.15 | 1.20 | 0.05 | 1.22 | 1.25 | 0.03 |
| c | 2.48 | 2.33 | 0.15 | 2.38 | 2.32 | 0.06 | 1.13 | 1.10 | 0.03 | 1.24 | 1.23 | 0.00 |
| d | 3.15 | 3.15 | 0.00 | 2.85 | 3.00 | 0.15 | 1.10 | 1.07 | 0.03 | 1.21 | 1.17 | 0.05 |
| e | 1.63 | 1.60 | 0.03 | 1.60 | 1.60 | 0.00 | 1.10 | 1.07 | 0.03 | 1.25 | 1.18 | 0.07 |
| f | 2.22 | 2.38 | 0.16 | 2.05 | 2.26 | 0.21* | 1.03 | 1.07 | 0.03 | 1.08 | 1.16 | 0.09 |
| g | 2.65 | 2.73 | 0.08 | 2.58 | 2.62 | 0.04 | 1.68 | 1.59 | 0.10 | 1.75 | 1.75 | 0.00 |
| h | 2.80 | 2.71 | 0.09 | 2.65 | 2.57 | 0.08 | 0.77 | 0.65 | 0.12* | 0.67 | 0.75 | 0.08 |
| i | 3.18 | 3.28 | 0.10 | 3.03 | 2.98 | 0.05 | 1.58 | 1.50 | 0.08 | 1.67 | 1.59 | 0.08 |
| j | 2.47 | 2.43 | 0.04 | 2.33 | 2.32 | 0.01 | 1.00 | 0.97 | 0.03 | 1.04 | 1.10 | 0.06 |
| Mean | 2.63 | 2.60 | 0.03 | 2.49 | 2.52 | 0.03 | 1.15 | 1.11 | 0.04 | 1.21 | 1.21 | 0.00 |
| SD | 0.47 | 0.47 | | 0.42 | 0.42 | | 0.28 | 0.28 | | 0.31 | 0.29 | |
| Spearman's rank correlation coefficients | $r_s=0.855$ $p<0.01$ | | | $r_s=0.948$ $p<0.001$ | | | $r_s=0.991$ $p<0.001$ | | | $r_s=0.952$ $p<0.001$ | | |

Difference : The absolute difference between the first and second tests.

* : The maximum difference between two tests

Discussion

1. Concordance of measurements made by two observers

Because the same individual cannot take all measurements in clinical settings, it is important to ascertain the concordance of measurements made by different observers. Unfortunately, few studies have investigated inter-observer concordance for ultrasonic measurements²⁰. In one such study, however, Fukunaga, et al.²⁰ documented high correlation coefficients for muscle and subcutaneous fat thicknesses ($r=0.990$ and 0.995 , respectively). In this study, although no significant difference in measured muscle thickness was evident between the two observers, a significant inter-observer difference in subcutaneous fat thickness was found. Observer B had one year of experience in leg measurement, and observer A had 20 years of experience in ultrasound, but no experience in leg measurement. Therefore, the difference between the two observers could have been attributable to this difference in experience with regard to leg measurement. One study compared measurements made using calipers among three observers²¹, and the results confirmed significant inter-observer differences. While the degree of inter-observer difference appears to be small for muscle thickness, it will be necessary to standardize measurement methods for subcutaneous fat thickness in order to minimize inter-observer differences attributable to experience.

2. Concordance of measurements made by the same observer on two separate days

The reliability of repeated ultrasound measurements made by the same observer has not been widely investigated^{10,20}. Fukunaga, et al.¹⁰ reported that when repeated measurements were made by the same individuals, reproducibility (r) was high: 0.992 for muscle thickness and 0.953 for subcutaneous fat thickness. Subcutaneous fat thickness is generally measured using calipers, but measurements can vary depending on the site of measurement and how subcutaneous fat tissue is pinched. Matsuo, et al.²¹ reported that when subcutaneous fat thickness was measured using calipers, the correlation coefficient of two measurements made at different sites

was low ($r=0.5$). The reproducibility (r_s) of measurements made on two separate days in this study was 0.855 for muscle thickness and 0.863 for subcutaneous fat thickness, and while these figures were slightly lower than those reported by Fukunaga, et al.¹⁰, they were higher than those obtained using calipers. Therefore, the reproducibility of repeated ultrasonic measurements made by the same individual appears relatively high.

3. Concordance of measurements made with or without SonarGel

One of the ways of minimizing measurement errors related to probe usage is to use SonarGel. Based on the high correlation coefficient and maximum difference between the two tests, measurements appear to be more stable with SonarGel than without. However, in most subjects, SonarGel resulted in smaller measurements for muscle thickness and larger measurements for subcutaneous fat thickness. When measuring the thickness of extremities, the probe is likely to push away muscle tissue more than subcutaneous fat tissue²². Limb thickness was measured in this study, and SonarGel, which weighs 220g, may have compressed muscle tissue. In addition, because the SonarGel layer was 1 cm thick, the ultrasound signals may have been attenuated as they traveled through the body. While SonarGel can minimize measurement errors in repeated measurements, it is not necessarily useful in assessing muscle thickness. Hence, it is necessary to further improve this technique and compare measurements with SonarGel to true values.

4. Suggestions to nurses

While measurement error cannot be ignored, ultrasonography is a noninvasive and convenient imaging technique that nurses can use. When performing ultrasonography in clinical settings, the results of investigations on measurement concordance among different testers have shown that ultrasonography should be performed by the same individuals whenever possible. Furthermore, in order to improve measurement accuracy and minimize measurement errors, adequate training is necessary.

5. Future directions of research

In this study, ultrasonography was performed on

adults in the recumbent position, but in clinical settings, many patients are elderly. As a result, it will be necessary to investigate measurement accuracy in a range of cases, including the elderly.

We also believe that it will be necessary to determine the relationship between tissue thickness of the rectus femoris as assessed by ultrasonography and total muscle mass of the leg, and to investigate a means to improve measurement accuracy by comparing ultrasonography with other quantitative imaging techniques such as CT and DXA.

Conclusions

In order to verify the accuracy of thicknesses of rectus femoris muscle and overlying subcutaneous fat in healthy adults as measured by ultrasound, three experiments were conducted to assess : 1) concordance of measurements made by two different observers ; 2) concordance of repeated measurements made by the same observer ; and 3) comparison of repeated measurements made with and without SonarGel. The results were as follows :

1. Between two observers with different levels of experience in ultrasound, a significant difference was evident for subcutaneous fat thickness, but not for muscle thickness.
2. When measurements were made by the same observer on two separate days, a significant positive correlation was noted, suggesting favorable reproducibility.
3. When measurements were made by the same observer on two separate days with and without SonarGel, the correlation coefficient between the two tests was high. However, in many subjects, SonarGel resulted in smaller measurements for muscle thickness and larger measurements for subcutaneous fat thickness.

In order to measure tissue thickness of the rectus femoris by ultrasound, it will be necessary to minimize intra-observer differences and to determine the correlation between ultrasound findings and those of other imaging techniques. However, based on the accuracy of measurements obtained in the present study, ultrasound appears to be appropriate for clinical settings.

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