Original Article

Assessment of ultrasound shear wave elastography within muscles using different region of interest sizes, manufacturers, probes and acquisition angles: an *ex vivo* study

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Background: The application of shear wave elastography (SWE) in assessment of the musculoskeletal system is affected by various factors. This study aimed to explore the influence of machines, probes, region of interest (ROI) sizes, and the acquisition angles on muscle shear wave speed (SWS).

Methods: The SWS of *ex vivo* isolated muscles were acquired using 3 different machines (Aixplorer system, SuperSonic Imagine; Acuson S3000, Siemens Healthcare; Resona 7, Mindray) and 2 linear probes (Aixplorer system, SL 10-2 and SL 15-4). Also, 4 different ROI sizes (diameter 1–10 mm) and 9 different acquisition angles (0–40°) were tested. The SWS acquired under different conditions were compared, and the intra-class correlation coefficients (ICC) were used to evaluate reproducibility.

Results: There was a significant difference in SWS acquired using the 3 different machines (P<0.001) or with 9 different angles (P=0.008). There was no significant difference in SWS acquired using 2 probes (P=0.053) or 4 different ROI sizes (P=0.874, 0.778, and 0.865 for 3 operators, respectively). All machines produced substantial intra-system reproducibility (ICC, 0.61–0.80). Both probes demonstrated an almost perfect degree of intra-system agreement (ICC, >0.80), and nearly all ROI sizes demonstrated an almost perfect degree of intra- and inter-operator agreement (ICC, >0.80). The measurement reliability was higher when the acquisition angles were no more than 20°.

Conclusions: The 3 machines had different SWS values. Attention should be paid when comparing SWS results using different machines. For the Aixplorer system, the ROI size had no effect on the SWS values. Angles larger than 25° will lead to SWS measurements with greater variability compared to smaller angles (≤20°).

Keywords: Shear wave elastography (SWE); shear wave speed (SWS); ultrasound; muscles; influencing factors

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Introduction

Shear wave elastography (SWE) is a promising modality for evaluating the elasticity of the musculoskeletal system. Shear wave speed (SWS) is closely related to tissue stiffness, with faster propagating shear waves representing stiffer tissue (1,2). The feasibility of SWE has been confirmed in the diagnosis of breast (3), liver (4), thyroid (5), and prostate (6) lesions. However, the role of SWE in the

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evaluation of musculoskeletal diseases still requires further research, and more studies are needed to establish a standard protocol. Young's modulus can be estimated from SWS, assuming a linear isotropic and homogeneous tissue model. The relationship between Young's modulus and SWS is $E = 3\rho C^2$. In this equation, E represents Young's modulus, 3 is a constant related to Poisson's ratio for strain, ρ is tissue density (assumed to be 1 g/cm^3), and C is the SWS. Therefore, it is recommended that SWS results are recorded in m/s, considering the anisotropy of muscles (7,8). Pathological changes, such as muscular dystrophy (9), spasticity (10), and inflammation (11), can change the mechanical properties of muscles. The technology of SWE can non-invasively evaluate tissue stiffness qualitatively and quantitatively (12-14) and has the potential to assess muscle properties effectively. However, the application of SWE to the musculoskeletal system must overcome various structural and anatomical problems, such as tissue heterogeneity due to the presence of tendon and aponeurotic structures, muscle contraction and relaxation, location variability, and the complexity of adjacent structures (15,16). All of these features have been shown to influence SWE readings (17-19), further complicating *in vivo* evaluation of muscle elasticity.

Various factors, such as the region of interest (ROI) size, machines from different manufacturers, probes of different frequencies, and probe orientation, all exert influence on SWS muscle measurement. For many machines, there are various ROI sizes for selection. Recently, 2 studies focused on the effect of ROI on muscle SWS and both reported that ROI size (ROI sizes 28–707 mm^2 and 15–200 mm^2, respectively) had little influence on SWS (20,21). Most studies have focused on whether ROI size affects SWS, and almost no studies have focused on the reproducibility of different ROI sizes, which has particular significance for follow-up evaluation of lesions. As far as we know, only a few studies have evaluated the variability and repeatability of SWS measured with different ultrasound elastography machines (22,23). A phantom study demonstrated SWS variability in machines from different manufacturers, indicating a considerable difference in SWS depending on the type of machine used (24). However, it is not appropriate to extrapolate conclusions obtained from phantom studies to the musculoskeletal system. Furthermore, few studies have evaluated the variability of SWS acquired from different machines using different probes within healthy *ex vivo* muscles, which is vital for results comparison in clinical practice and future studies. The human body has pennate muscles, such as the gastrocnemius and vastus lateralis, that require particular attention because the angle between the muscle fiber and the long axis of the muscle is variable. Several previous studies have also indicated variabilities regarding probe orientation. These studies showed that the best reproducibility was associated with the longitudinal plane relative to the muscle fibers in normal skeletal muscles (19,21). Miyamoto *et al.* reported that the reliability of shear modulus measurements remains high if the angle between the probe axis and the orientation of the muscular fibers is below 20° (25). To date, there has been no study revealing the feasible angle range relative to the fascicle direction in the same plane.

Further standardization of the acquisition process is essential for improving measurement accuracy. In this work, 3 professional sonographers acquired the SWS of isolated muscles. The purpose of this study was to evaluate the SWS variations in *ex vivo* muscles with different ROI sizes, machines, probes, and acquisition angles. Furthermore, the intra- and inter-operator reproducibility of the mean SWS under different conditions was investigated. We present the following article in accordance with the Guidelines for Reporting Reliability and Agreement Studies (GRRAS) reporting checklist (available at https://qims.amegroups.com/article/view/10.21037/qims-21-1072/rc).

Methods

Materials

A total of 80 samples (20 for the ROI comparison, 20 for the machine comparison, 20 for the probe comparison, and 20 for the angle comparison) of fresh, *ex vivo* pork tenderloin muscle with clear fiber provided by the same slaughterhouse were obtained. These 80 specimens were selected based on our pre-test experience. They were removed from the pig bodies 6–15 h before use and measured at 20 ℃ temperature, with an average size of [10–15] cm × 7 cm × 7 cm.

Our research was an exploratory study; it did not require a large sample size, so we chose a sample size of 20. The small number of samples not only saved on staffing, financial resources, and time, but also guaranteed the accuracy and reliability of the research results. To minimize the elastic changes of specimens over time, we selected 20 separate specimens for each of the different item comparisons.

Ultrasound machines

We used 3 two-dimensional (2D) SWE machines from different manufacturers: the Aixplorer system (SuperSonic

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Imagine, Aix-en-Provence, France; machine I) equipped with SL 10-2 and SL 15-4 probes (probes A and B), the Acuson S3000 (Siemens Healthcare, Erlangen, Germany; machine II) equipped with a 9L4 probe, and the Resona 7 (Mindray, Shenzhen, China; machine III) equipped with an L14-5WU probe.

The Aixplorer system operates with a spatial resolution of 1 mm × 1 mm in elastography (7). The SWE uses the acoustic radiation force induced by ultrasound beams to stimulate the target tissues. A rectangular translucent color window of 2 cm × 2.5 cm with the ROIs placed at the center of the box at a depth of 2 cm with a couplant of 2 mm was chosen as the acquisition method for all machines. No air bubbles were allowed in the couplant. The Aixplorer system penetration (Pen) condition was chosen. The values were presented in units of m/s (26). During imaging, SWS measurement should avoid "fence" artifacts, areas of signal void, and areas of too high or too low signal to minimize inaccurate measurements. To prevent measurement inaccuracy when evaluating the muscle elasticity, we avoided placing the ROI on the edge of the color window. The muscle samples were placed between the 2 fixators during the examination to maintain the shape, but no pressure was applied. Measurements were repeated 3 times for each condition, and the probe was removed and replaced each time.

Operators

The study was conducted by 3 board-certified sonographers (operators A, B, and C) with more than 3 years' experience in ultrasound elastography. Before the experiment, all operators were trained on the specimens by completing at least 30 successful experimental operations. During the examination, the operator placed the probe along the direction of the muscle fiber. Each operator completed the SWE procedures independently and was blind to the measurement results of the other operators. The measurement order of the 3 operators was random.

ROI

The SWE measurements were acquired using 20 muscle samples with machine I equipped with an SL10-2 linear probe. Circular ROIs of 1, 3, 5, and 10 mm diameter were chosen. Readings were repeated 3 times for each ROI size by each operator, and the probe was removed and replaced each time. The SWE procedures were performed independently by operators A, B, and C.

Different SWE systems

The 3 machines from different manufacturers

We used 3 different machines, including the Aixplorer system equipped with an SL 10-2 probe, the Acuson S3000 equipped with a 9L4 probe, and the Resona 7 equipped with an L14-5WU probe.

A circular ROI with a diameter of 1 mm was selected for machines I and III, while the ROI size was an unchangeable rectangle in machine II (1 mm × 1 mm in size), as shown in *Figure 1*.

The SWS was acquired from 20 muscle samples using the 3 machines consecutively. The measurement order of the 3 machines for each sample was random. Each sample measurement took about 30 min, and the probe was placed at the same position each time. A marker was used to record the specific measurement location of each sample on the surface of the muscle. Measurements were repeated 3 times. The time interval between the 3 measurements was 8–10 min.

The 2 probes of different frequencies

The SWS was acquired from another 20 muscle samples using 2 probes, SL 10-2 and SL 15-4 (with center frequencies of 6 and 8.5 MHz, respectively) with the SuperSonic Aixplorer, as illustrated in *Figure 2*. Similarly, each sample measurement took 30 min to complete.

Operator A conducted these measurements.

Angles

A total of 20 isolated muscle samples were selected to assess the influence of the acquisition angle on muscle SWS using machine I equipped with an SL 10-2 probe and a 5 mm ROI diameter. A depth of 2 cm was chosen as the acquisition method for all angles. The total time for each muscle measurement was no more than 60 min. We used "α" to represent the angle between the probe plane and the muscle surface (*Figure 3*). In the present study, 0° indicated that the probe plane was parallel to the muscle surface. During the examination, the probe was always placed along the direction of the muscle fiber. Measurements were acquired using 9 different angles: 0°, 5°, 10°, 15°, 20°, 25°, 30°, 35°, and 40° at the same position. When the probe was positioned obliquely to the muscle surface, there was a sufficient amount of coupling agent and no bubbles (Figures S1,S2).

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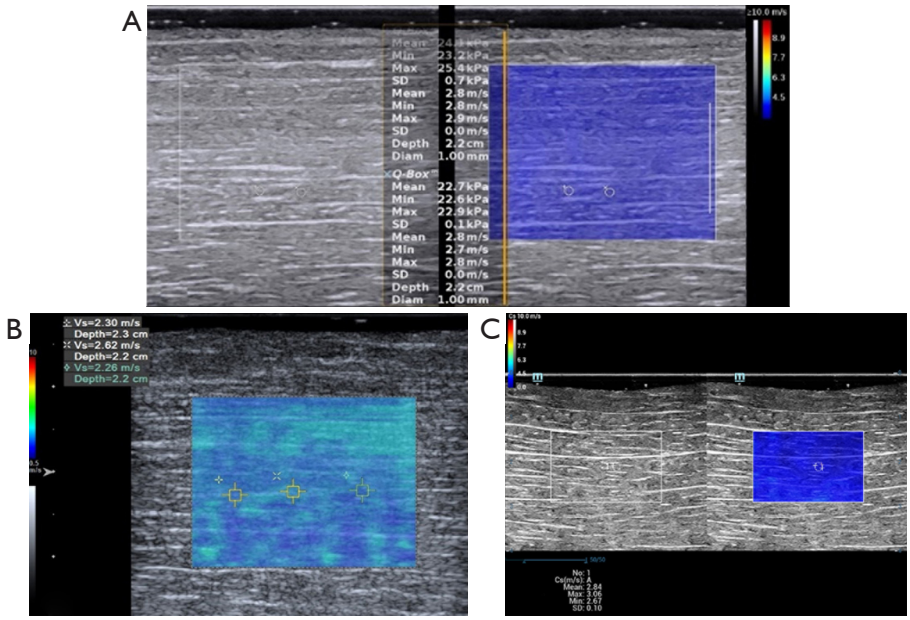

Figure 1 Elastogram obtained from muscles using 3 different machines at a depth of 2 cm with 2 mm couplant: (A) machine I: Aixplorer system equipped with an SL 10-2 probe; (B) machine II: Acuson S3000 equipped with a 9L4 probe; (C) machine III: Resona 7 equipped with an L14-5WU probe. SD, standard deviation.

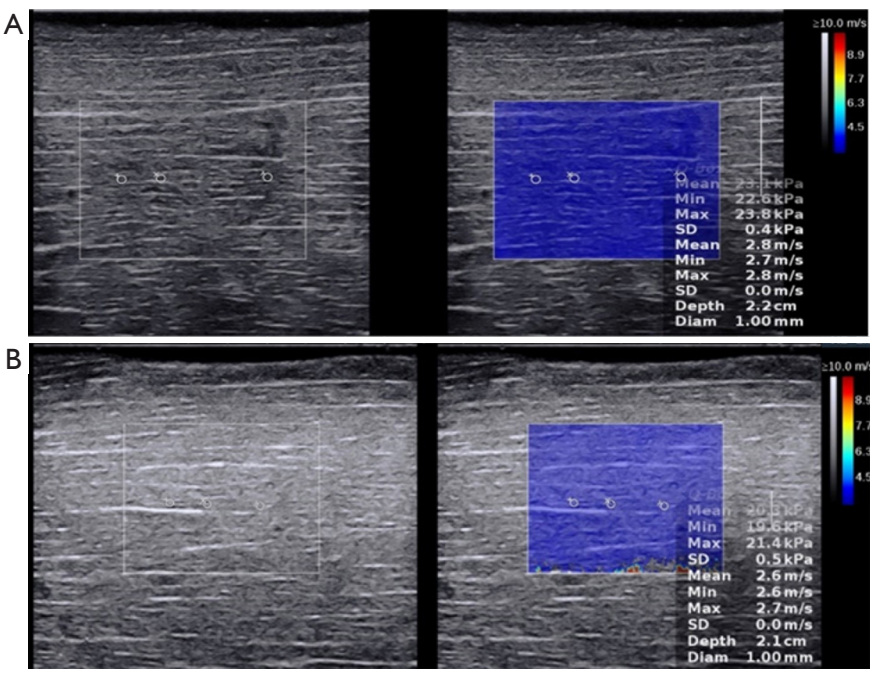

Figure 2 Elastogram obtained from muscles using two different probes at a depth of 2 cm with 2 mm couplant in machine I: (A) SL 10-2 probe (with a center frequency of 6 MHz); (B) SL 15-4 probe (with a center frequency of 8.5 MHz). SD, standard deviation.

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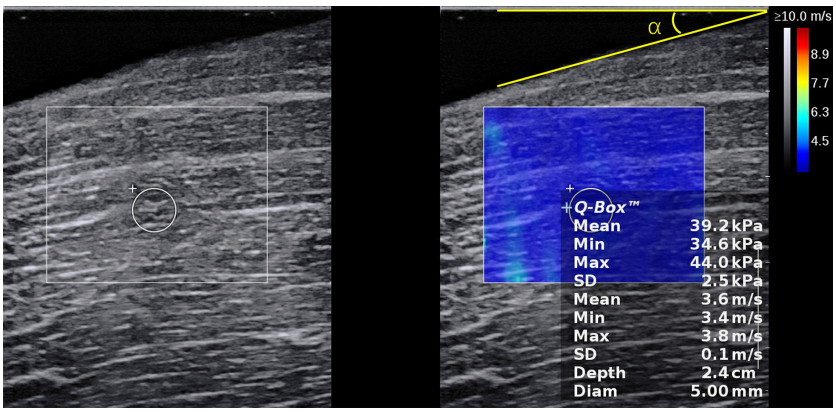

Figure 3 Elastogram acquired from muscles at a depth of 2 cm when the imaging angle (α) was 15°. SD, standard deviation.

No pressure or deformation of the muscle tissues occurred during operation. For each angle, measurements were repeated 3 times. Operator A conducted the above measurements.

Data and statistical analysis

The software SPSS 22.0 (IBM Corp., Armonk, NY, USA) was used for statistical analysis. All data were presented as the mean ± standard deviation (M ± SD), and P value <0.05 was considered statistically significant. For the comparison of variance in the different measurement conditions, the coefficient of variation (CV) was calculated by applying the equation: CV = standard deviation/mean value (27). The larger the CV value, the lower the reliability and repeatability of each measurement. The Kolmogorov-Smirnov test was used to check if the data were normally distributed. The test for homogeneity of variance was applied to check for differences in variance. Analysis of variance (ANOVA) was used to compare the data from the different ROIs and machines. The Kruskal-Wallis test was used to compare the data from the different acquisition angles. A *t*-test was performed to compare SWS between the 2 different probes. For the subgroup analysis, Tamhane's T2 post hoc test was used to compare SWS between the different machines and angles. The repeatability and reproducibility of the measurements were quantified using intra-class correlation coefficients (ICC) (two-way mixed effect model). The ICC values were interpreted as follows: 0.00–0.20 for poor agreement, 0.21–0.40 for fair agreement, 0.41–0.60 for moderate agreement, 0.61–0.80 for substantial agreement, and >0.80 for almost perfect agreement (21). The ICC and 95% confidence interval (CI) were used to evaluate the intra- and inter-operator consistency.

Results

Results obtained with different ROI sizes

The SWS was successfully acquired from 20 muscle samples. The Kolmogorov-Smirnov test showed all P values were >0.05, indicating that the data were normally distributed. The ANOVA results revealed that SWS did not differ substantively by ROI size (*Table 1*). The degrees of intra- and inter-operator agreement under each ROI size were almost perfect (ICC, >0.80) except for the 1 mm ROI diameter in the inter-operator agreement (*Table 2*). *Figure 4* shows the SWS comparisons under the different ROI sizes by the 3 operators.

Comparison of different SWE systems

The SWS was successfully acquired from 20 muscle samples for the comparison of SWS of different machines and probes.

Tables 3,4 present the SWS results obtained using the 3 different machines and the 2 different frequency probes. *Figure 5* shows the SWS distribution using the 3 different machines.

The Kolmogorov-Smirnov test showed all P values =0.20 for the 3 groups of data from the 3 machines. The SWS acquired at a depth of 2 cm was significantly different between the 3 machines (P<0.001). In the subgroup analysis, there were obvious differences between any 2 machines (P<0.001). The CVs were 0.06 in machines I and II, while

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Table 1 The SWS results of 3 operators using different ROI sizes

ROI diameter (mm)	SWS (M ± SD) (m/s) with different ROI diameter			
	A	B	C	P value
1	4.75±0.58	4.35±0.46	4.42±0.39	0.036
3	4.60±0.60	4.39±0.52	4.50±0.36	0.464
5	4.63±0.60	4.47±0.54	4.45±0.40	0.521
10	4.62±0.58	4.51±0.54	4.52±0.38	0.752
P value	0.874	0.778	0.865	

The ANOVA test showed no significant differences among means of any of the analyzed variables; A, B, and C represent the 3 operators. P<0.05 was considered statistically significant. ROI, region of interest; SWS, shear wave speed; M ± SD, mean ± standard deviation; ANOVA, analysis of variance.

Table 2 Reproducibility of different ROI sizes demonstrated as ICC

ROI diameter (mm)	Intra-operator ICC (95% CI)			Inter-operator ICC (95% CI)
	A	B	C	
1	0.96 (0.91–0.98)	0.98 (0.95–0.99)	0.91 (0.81–0.97)	0.74 (0.42–0.89)
3	0.92 (0.83–0.97)	0.98 (0.95–0.99)	0.91 (0.81–0.97)	0.88 (0.73–0.95)
5	0.91 (0.81–0.96)	0.98 (0.95–0.99)	0.95 (0.89–0.98)	0.89 (0.75–0.96)
10	0.92 (0.83–0.97)	0.98 (0.96–0.99)	0.96 (0.91–0.98)	0.85 (0.68–0.94)

A, B, and C represent the 3 operators. ROI, region of interest; ICC, intra-class correlation coefficients; CI, confidence interval.

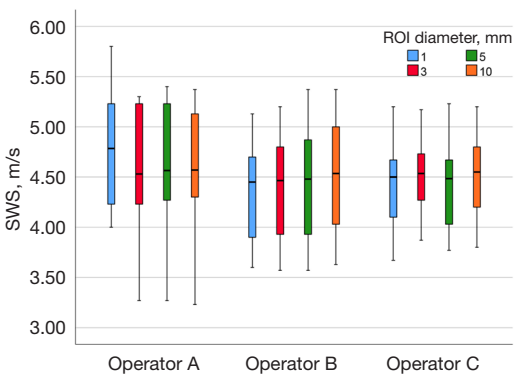

Figure 4 Comparison of muscle SWS under different ROI sizes by 3 operators. SWS, shear wave speed; ROI, region of interest.

the CV was 0.08 in machine III. The highest CV and SWS were associated with machine III, and the lowest CV and SWS were associated with machine II. The SWS acquired from all 3 machines produced a substantial intra-system reproducibility (ICC, 0.61–0.80). The SWS acquired from machine II achieved the best intra-system reliability compared to the results acquired from machines I and III. Comparing the results from the 3 machines, the degree of inter-system agreement reproducibility was poor (ICC, <0.20).

For the probe comparison, the test for homogeneity of variance showed P=0.73, which indicated the variances were equal. No significant difference was found between SWS

Table 3 The SWS results acquired using 3 different machines, showing the intra- and inter-system ICC

Machines	SWS (M ± SD) (m/s)	P value	CV	Intra-system ICC (95% CI)	Inter-system ICC (95% CI)
Machine I	3.24±0.18	<0.001*	0.06	0.64 (0.29–0.84)	0.05 (0–0.57)
Machine II	2.51±0.16		0.06	0.69 (0.42–0.84)	
Machine III	3.51±0.27		0.08	0.61 (0.25–0.82)	

P<0.05 was considered statistically significant. *, denotes a significant difference between groups (P<0.05). Machine I, Aixplorer system, SuperSonic Imagine; machine II, Acuson S3000, Siemens Healthcare; machine III, Resona 7, Mindray. SWS, shear wave speed; ICC, intra-class correlation coefficients; M ± SD, mean ± standard deviation; CV, coefficient of variation; CI, confidence interval.

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Table 4 Results acquired using two different probes and the intra-system ICC of each probe

Probes	SWS (M ± SD) (m/s)	P value	CV	Intra-system ICC (95% CI)
Probe A	2.85±0.22	0.053	0.08	0.84 (0.69–0.93)
Probe B	2.98±0.23		0.08	0.90 (0.81–0.95)

P<0.05 was considered statistically significant. Probe A, Aixplorer system, SL 10-2; Probe B, Aixplorer system, SL. ICC, intra-class correlation coefficients; SWS, shear wave speed; M ± SD, mean ± standard deviation; CV, coefficient of variation; CI, confidence interval.

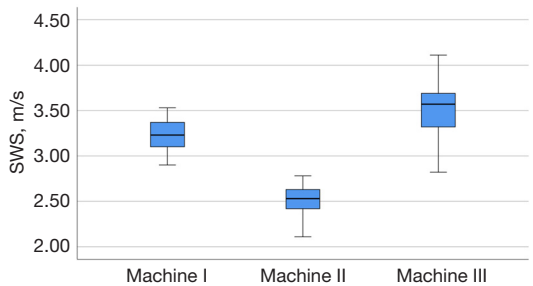

Figure 5 The SWS distribution using three different machines. Machine I: Aixplorer system; machine II: Acuson S3000; machine III: Resona 7. SWS, shear wave speed.

acquired with the 2 linear probes of different frequencies (P>0.05), and both probes had almost perfectly consistent results (ICC, >0.80).

Results obtained under different angles

The SWS was successfully acquired from 15 muscle samples.

The results of SWS with different angles in machine I are shown in *Table 5*. The SWS measured at different angles (0–40°) was significantly different (P<0.05). When the angle was less than 20°, the CVs were small. There was no significant difference between the SWS measured at angles 5–20° and 0°. When the angle increased to 25° or more, the CVs became large, and the SWS difference between the large angles (≥25°) and 0° occurred. The largest CV and the lowest ICC were related to an angle of 35°. The filling rate of the elastogram was extremely low, and only 2 specimens were successfully measured at an angle of 40°. The SWS acquired at 0–25° achieved the best intra-system reliability compared to the other angles. The comparison of SWS measured at different angles is shown in *Figure 6*.

Discussion

The SWE is a valuable and promising tool for disease diagnosis of the musculoskeletal system, and the accurate evaluation of muscle elasticity is of great significance for clinical diagnosis (28). To the best of our knowledge, few studies have explored the same variables using *ex vivo* muscle samples. There is also limited information on the performance of the Mindray SWE system. The present study focused on the effects of different ROI sizes, various probe orientations, probes of different frequencies, and popular machines from 3 different manufacturers on muscle SWS that may be important for the future standardization of muscle evaluation.

The objective of the first part of our study was to evaluate the influence of various ROI sizes on SWS and the reproducibility of SWS under different ROI sizes. Kot *et al.* (29) acquired muscle elasticity with 3 different ROI sizes (8, 10, and 12 mm diameter for the right rectus femoris and 2, 3, and 4 mm diameter for the patella tendon) and reported no significant difference in the mean value of the elasticity modulus (29). Alfuraih *et al.* evaluated the elasticity of the rectus femoris muscle and reported that SWS did not differ substantively by ROI size (15, 75, and 200 mm^2), but reliability indices indicated that the smaller ROI did not perform quite as well as the larger sizes (21). Our study demonstrated that the mean value did not differ with various ROI sizes, and the intra-and inter-operator ICC was almost perfect except for the inter-operator ICC for an ROI of 1 mm. When evaluating the elasticity of muscles with a relatively uniform elastic distribution, the SWS values were the average value of the entire circular ROI, which may explain why no significant difference in the mean value of the SWS was found. Therefore, it is acceptable to report the mean SWS value of the muscle when using different ROI sizes. Our study demonstrated that using an ROI of 1–10 mm produced low variability and high reproducibility. In routine examinations, the ROI size might be tailored to the specific circumstances. A larger ROI may be necessary for diseases affecting a large muscle area, and a smaller ROI may be more suitable for small focal lesions (21).

The second part of this study investigated the variations in SWS of *ex vivo* muscles when using different machines and probes. Shin *et al.* investigated consistency of SWS in ultrasound elastography using different machines and probes (a high-frequency linear probe and a low-frequency convex

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Table 5 Results under different angle conditions in machine I

Parameters	0°	5°	10°	15°	20°	25°	30°	35°	40°
SWS (M ± SD) (m/s)	3.97±0.38	3.96±0.61	3.96±0.57	3.87±0.38	3.91±0.53	4.67±1.09	4.95±1.26	3.69±1.48	5.32±0.92
CV	0.10	0.15	0.14	0.10	0.14	0.23	0.25	0.40	0.17
ICC	0.96 (0.89–0.99)	0.90 (0.77–0.97)	0.92 (0.80–0.97)	0.86 (0.69–0.95)	0.85 (0.67–0.95)	0.89 (0.76–0.96)	0.45 (0.09–0.77)	0.29 (0–0.73)	0.80 (0.08–0.99)
P value					0.008*				

P<0.05 was considered statistically significant. *, denotes a significant difference between groups (P<0.05). Machine I, Aixplorer system, SuperSonic Imagine. SWS, shear wave speed; M ± SD, mean ± standard deviation; CV, coefficient of variation; ICC, intra-class correlation coefficients.

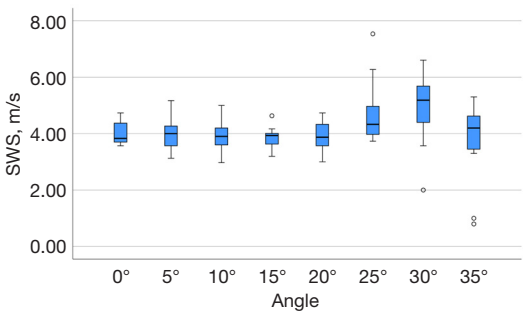

Figure 6 Comparison of muscle SWS under different acquisition angles. SWS, shear wave speed.

probe) and found a considerable difference in SWS (24). However, their research focused on a phantom study, and the results may not apply to muscle tissues. Alfuraih *et al.* compared the reliability between 2 different machines (LOGIQ E9 and Aixplorer) within muscles (21). They reported that the LOGIQ E9 system produced reliable results that were comparable with the Aixplorer system; however, their study was an *in vivo* experiment that was susceptible to muscle state and body posture, and they did not evaluate the influence of different frequency probes. Another study compared SWS using the ACUSON S2000 and ElastPQ machines and revealed significant differences in liver stiffness between the 2 machines (30). However, to our knowledge, only a few studies have compared SWS obtained from 3 different machines of different manufacturers and tested the systems' reliability and consistency on *ex vivo* musculoskeletal imaging. The present study showed a considerable difference in SWS on ultrasound elastography obtained from the machines supplied by different manufacturers. This is likely because machines from different manufacturers are somewhat different in the way they generate and calculate SWS in the tissues. Also, this study briefly compared the degree of agreement between 3 repeated acquisitions using the 3 machines. The results showed that the intra-system consistency of each machine was satisfactory, whereas the inter-system consistency of the 3 machines was unsatisfactory. Therefore, care should be taken in clinical practice or scientific studies when comparing results from different elastography machines for patient follow-up. The CVs of the present study were small, ranging from 0.06 to 0.08, and similar to the results of a previous phantom study, which showed CVs of 0–0.09 at depths of 2–5 cm (24).

Several studies have shown that probes with different frequencies can affect SWS measurement results. Chang *et al.* acquired the SWS of an elasticity phantom and normal liver with convex and linear probes and showed that the SWS with a low-frequency probe was higher at the same depth using the ACUSON S2000 (31). Another study investigated the normal liver stiffness values of healthy children using acoustic radiation force impulse (ARFI) technology with the ACUSON S2000 equipped with convex and linear probes and found that the SWS obtained using a linear probe was lower than that obtained using a convex probe (32). These studies suggested that these differences might be caused by different frequencies and spatial resolutions. Dillman *et al.* used linear probes of 2–10 and 4–15 MHz to evaluate superficial soft phantoms at 1.0, 2.5, and 4.0 cm depths using the Aixplorer system and reported a non-significant effect of the probes on SWS measurements (23). The present study compared 2 linear probes with different frequencies (SL 10-2 and SL 15-4) on *ex vivo* muscles at a 2 cm depth and found no significantly different effects on muscle SWS. This result is probably due to the similar working performance of the 2 probes, and the frequency of the push pulse may have been identical given that the bandwidth of both probes overlaps significantly

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(2–10 and 4–15 MHz). Also, the acquisition depth was not deep enough to make the sound attenuation obvious.

The last part of this study evaluated the effect of the angle between the probe plane and the muscle surface on SWS. Our results suggest that the acceptable detection angle range is between 0–20° when scanning muscles. Alfuraih *et al.* tested the elasticity of the rectus femoris *in vivo* in the longitudinal (along the muscle fiber direction), oblique, and lateral directions, revealing that the longitudinal direction was the best probe orientation (21). Miyamoto *et al.* studied the effect of probe angle on shear wave modulus by placing the probe parallel or 20° obliquely to the fascicle using B-mode images and found that the reproducibility of the shear modulus measurements was high in both the parallel and oblique conditions (25). However, they only assessed 2 angles, and our study applied a larger angle range and explored an acceptable range of angles. Our study found that when the angle was ≤20°, there was no obvious difference in the SWS compared with 0°, along with high reliability and small CVs. However, when the angle was >20°, there was a significant difference in SWS compared with 0°, with larger CVs and suddenly increased or decreased SWS. The heterogeneous high or low tissue elastogram observed in the larger acquisition angles (>20°) may be due to the poor propagation of shear waves in a more anisotropic structure. Larger angles produced a higher level of anisotropy than smaller angles. Therefore, when defining the probe orientation in anisotropic structures, the orientation of the fibers must be considered. An angle of no more than 20° between the muscle fiber and probe will result in better reproducibility and higher reliability, and larger angles (≥25°) will produce inaccurate SWS measurements.

Our study had some limitations. Firstly, isolated porcine muscles were used and successively measured by different operators; therefore, the tissue may have decomposed over time. Secondly, the temperature and perfusion of the isolated muscles were different from the living body temperature, which may have led to elasticity changes in the tissues. Thirdly, for SWS comparison of different angles, the thickness of the couplant applied to the muscle surface was different; larger angles needed more couplant on the muscle surface. This may have caused differences in sound attenuation, further affecting the SWS measurement. Lastly, for better evaluation of the intra-observer agreement, a longer time interval was needed. However, in the present study, 3 measurements were carried out with a time interval between the 3 measurements of only 8–10 min to avoid elasticity changes over time.

Conclusions

Machines from different manufacturers have a significant effect on muscle SWS measurements. Attention should be paid during patient follow-ups when comparing results from different machines. There was no difference in muscle SWS between the 2 linear probes with the Aixplorer system. Under the present study conditions, the size of the ROI had no significant effect on muscle SWS measurements. Therefore, the size of the ROI could be adjusted according to different situations. When evaluating muscle elasticity, the recommended acquisition angle should be no more than 20°, as an angle greater than 25° will lead to inaccurate SWS measurements.

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Footnote

Reporting Checklist: The authors have completed the GRRAS reporting checklist. Available at https://qims.amegroups.com/article/view/10.21037/qims-21-1072/rc

Conflicts of Interest: All authors have completed the ICMJE uniform disclosure form (available at https://qims.amegroups.com/article/view/10.21037/qims-21-1072/coif). The authors have no conflicts of interest to declare.

Ethical Statement: The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

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Supplementary

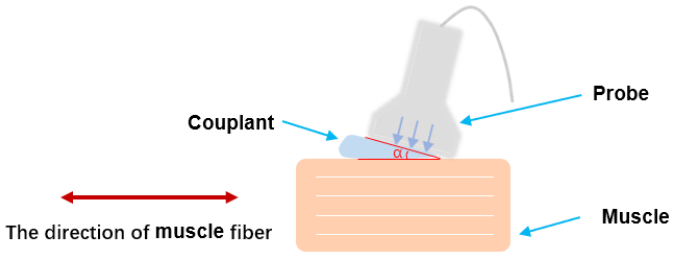

Figure S1 Schematic diagram of acquisition angle (α). The angle α is changed from 0° to 40°. The long axis direction of the probe was always along the direction of the muscle fiber.

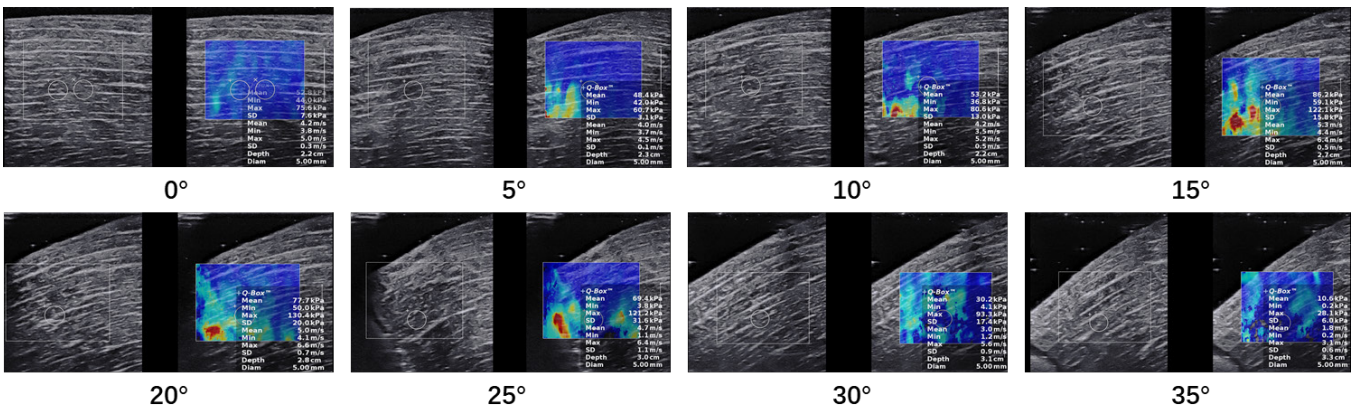

Figure S2 Elastogram acquired from muscles with different imaging angles (0–35°). SD, standard deviation.

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