Title Page

Poor Reproducibility of Compression Elastography in the Achilles Tendon: Same Day and Consecutive Day Measurements

Abstract:

Objective: To determine the reproducibility of compression elastography (CE) when measuring strain data, a measure of stiffness of the human Achilles tendon *in vivo,* over consecutive measures, consecutive days and when using different foot positions.

Materials and methods: 8 participants (4 males, 4 females; mean age 25.5 ± 2.51 years, range 21 – 30 years; height 173.6 ± 11.7 cm, range 156 – 189cm) had five consecutive CE measurements taken on one day and a further five CE measures taken, one per day, at the same time of day, every day for a consecutive five day period. These 80 measurements were used to assess both the repeatability and reproducibility of the technique. Means, standard deviations, coefficient of Variation (CV), Pearson correlation analysis (R) and Intra-class correlation coefficients (ICC) were calculated.

Results: For CE data, all CV's were above 53%, R values indicated no-to-weak correlations between measures at best (range 0.01 - 0.25), and ICC values were all classified in the poor category (range 0.00 - 0.11). CV's for length and diameter measures were acceptably low indicating a high level of reliability.

Conclusions: Given the wide variation obtained in the CE results, it was concluded that CE using this specific system has a low level of reproducibility for measuring the stiffness of the human Achilles tendon *in vivo* over consecutive days, consecutive measures and in different foot positions.

Introduction:

As primary plantar flexor of the ankle, the Achilles tendon experiences high levels of stress, often taking entire body weight at greater than 1g of acceleration with an associated high prevalence of injury (Milgrom et al. 2003; Wren et al. 2001). Despite the frequency and severity of Achilles tendon injuries, primary cause and optimal rehabilitation regimes remain unclear (Murtaugh & Ihm 2013). The inability of traditional B-mode ultrasound imaging to fully assess the mechanical properties of tendon may contribute to this uncertainty. Elastography proposes to assess tissue stiffness (Ophir et al. 1991), and has been suggested as being useful in its ability to depict alterations in tendon matrix before they are seen with traditional B-mode ultrasound imaging (Horton 2013; Sconfienza et al. 2013). Despite elastography software becoming more widespread across ultrasound systems (Sarvazyan et al. 2011) application of the technology to the musculoskeletal imaging field has been slower to progress with many feasibility studies being completed only over the last few years (Itoigawa et al. 2015; Ahn et al. 2014; Porta et al. 2014; Ooi et al. 2013; Yamamoto et al. 2015). The technology is not yet commonly used in the clinical setting with potential reasoning being that the majority of literature into elastography in the musculoskeletal setting have only shown moderate reliability and large gaps remain in the reliability literature such as results obtained over consecutive measures and the impact of previous exercise. Current literature using elastography to assess the Achilles tendon properties, has predominantly studied data obtained from a single measurement (De Zordo et al. 2009; Drakonaki et al. 2009; Klauser et al. 2013; Palle et al. 2011; Tan et al. 2012), with a more recent article assessing intra-rater reliability over 4 consecutive measures and inter-rater reliability from two examiners taking 3 consecutive measures (Yamamoto et al. 2015). No research has yet assessed the reproducibility when taking the same measures over different days, a factor that would be important to assess progression of disease or response to therapy. Previous

research has also not controlled for prior activity of those measured. This may be another important factor as tendon should not be assessed immediately following strenuous activity, as it will cause a physiological response (Boesen et al. 2006) such as that shown in research using ultrasound tissue characterisation (UTC). Exercise caused a loss in normal tendon structure for up to 2 days (Rosengarten et al. 2014) and therefore may significantly influence Achilles tendon properties and interpretation of any measured changes. The lack of knowledge surrounding reproducibility of measures taken over time or controlling for prior activity results in a gap in knowledge regarding the clinical effectiveness of elastography to track tendon changes over time.

The principle of elastography is similar to palpation, involving application of a force (stress) and measurement of the subsequent response (strain) (Sarvazyan et al. 2011). Compression (strain) elastography (CE) is one of the most common commercially available types of elastography that has been used to measure the Achilles tendon (De Zordo et al. 2009; Drakonaki et al. 2009; Klauser et al. 2013; Palle et al. 2011; Tan et al. 2012; Yamamoto et al. 2015). CE provides data relating to the strain experienced within the tissue, with this strain data providing an indication of the tissue stiffness. The effectiveness of CE for measuring the stiffness of the patellar tendon, plantar fascia and muscle has shown some potential, with studies reporting high reliability and reproducibility (Porta et al. 2014; Wu et al. 2011; Chino et al. 2012). However, these studies all tested participants on only one occasion, therefore the reproducibility over consecutive measures and days remains unknown. Good intra-observer reliability was recently shown for Achilles tendon assessment, when used with an acoustic coupler of a known Young's modulus as a reference standard (Yamamoto et al. 2015). Others have questioned its ability to assess tendon properties, as the technique appears to be operator dependant and qualitative, making it unsuitable for objectively measuring tissue stiffness (Arda et al. 2011; Treece et al. 2011).

The aim of this study was to measure the reproducibility of CE technology to measure strain data, which offers a measure of stiffness, to assess the human Achilles tendon *in vivo* over consecutive measures, consecutive days and using different foot positions. A secondary aim was to assess the reproducibility of the conventional ultrasound aspect of the technology to measure Achilles tendon length and diameter.

Materials and Methods:

Participants:

Eight healthy volunteers (4 males, 4 females; mean age 25.5 ± 2.51 years, range 21 – 30 years; height 173.6 ± 11.7 cm, range 156 – 189cm) were recruited from within the University Department where the research took place. All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards. Ethical approval for the study was also obtained from the University of Brighton ethics committee. All participants reported no current or historical injury to their Achilles tendon and mean VISA-A (Victorian Institute of Sport Assessment-Achilles questionnaire) score for the volunteers was 98.9 ± 2.0, indicating no current symptoms of Achilles tendon injury or discomfort (Iversen et al. 2012). Informed consent was obtained from all individual participants included in the study. Participants completed a VISA-A Questionnaire prior to testing to assess tendinopathy symptoms and inclusion criteria was set at a minimum score of 96/100. Exclusion criteria included pregnancy, pain in the Achilles tendon area, previous surgical intervention of the Achilles tendon, pre-diagnosed Achilles tendinopathy, any participants taking fluoroquinolone antibiotics, or anomaly on ultrasound. The B-mode images were independently reviewed by an experienced sports medicine doctor, with more than 15 years' experience in musculoskeletal ultrasound, to exclude any abnormal tendons. No recruited volunteers were excluded from the study.

Materials and Methods:

Data was collected in two testing blocks issued in a randomised order, each time a B-mode scan was taken to identify anatomical locations for points of measurement of CE. In one testing block, participants attended once and during that visit, had five consecutive CE measurements taken over a one hour period (1 hr Measures). In the other testing block, the same participants attended at the same time of day, every day for five consecutive days. On these days a single measure of CE was taken (Daily Measures). Participants were asked to maintain their typical daily walking activity in and around the work place but to refrain from any additional exercise above walking during the testing period.

Scanning techniques:

For all measures, participants lay prone on an examination table with both feet hanging clear of the end of the table. An appropriate amount of ultrasound gel was applied, and all measures were taken on the right Achilles tendon using a Siemens ACUSON S2000™ HELX EVOLUTION Ultrasound System (Siemens Medical Solutions, USA).

Figure 1 here

Conventional Ultrasound Technique

At every session, measures of Achilles tendon length and max AP diameter were made using a conventional ultrasound using a 14L5 probe. A single operator took extended field of view 'SieScape' images of the ‘free’ Achilles tendon. The Achilles tendon (A) is shown between the solid white lines in Fig 1 and length was measured between the insertion of the Achilles tendon at the calcaneus (B), to the lowest fibres of soleus, as shown by the dotted white lines on the B-mode ultrasound image (See Fig 1). Three consecutive SieScape images were taken to measure length, and a mean average of the three measures taken to represent 'free' Achilles tendon length. The ‘mid-point’ of the tendon (D) was calculated as half of free Achilles

tendon length, located and marked on the skin of the participant following calcaneus palpation. This mark was used for all subsequent measures in both longitudinal and transverse planes, to ensure all measures were taken at the relative mid-point of the tendon for each participant.

Figure 2 here

Compression Elastography Technique

All CE Images were obtained with the foot in two different positions, i) a relaxed position, and ii) both feet fixed at a 90° position, measured by goniometry. The participant's feet were fixed using a custom made strap wrapped around the back of both feet and secured to the examination table.

During each measure elastograms were taken using a 9L4 probe at the mid-point of the tendon and strain information calculated at the same Region of Interest's (ROI's) each scan.

Figure 3 here

The ROI's were placed manually using the machine's software in the same order each scan, with ten ROI's placed along the length of the tendon, starting proximally and working distally in the longitudinal plane (See Fig 2), as this was the number of ROI's that fitted within the boundaries of the tendon in each image selected for analysis. Figure 2 shows the B-mode image on the left of the screen with the CE image on the right. In the CE image, the darker the colour, the harder the tissue.

Figure 4 here

Four ROI's were used within transverse measures, placed at the same location and in the same order each time (See Fig 3). Figure 3 illustrates a transverse image with the B-mode

image on the left and the CE image on the right. Again, the darker colours indicate stiffer tissues.

Figure 5 here

The 'strain ratio' seen in Figures 2 & 3, is a ratio that compares two separate areas in the same elastogram (Brandenburg et al. 2014). As strain ratio is derived from the raw strain data, it was deemed inappropriate to use the ratio value for statistical analysis and instead, raw data scores for % displacement were used. As raw strain data was used for analysis, the use of a reference standard was not required. This figure represents the % value of displacement (true strain) of the pixels within the given ROI. Image quality was closely monitored throughout scanning; tissue compression was avoided during examination and quality factors (QF) noted. To ensure the best possible images are used for analysis, CE software provides information on the quality of each image using a QF (Wu et al. 2011) indicating the amount of motion artefact compared to a reference frame (Calvete et al. 2013). A QF above 60 has been used to indicate an image of good quality (Wu et al. 2011), with some suggesting a quality factor between 50-100 represents minimal motion artefact (Calvete et al. 2013). To ensure an excellent standard of image, this study used a minimum QF of 75, observed for at least 5 consecutive frames (Calvete et al. 2013) for image quality standardisation (Note QF of 80 in Fig 2).

Statistical Analysis:

Data are presented as means ± SD. All statistical analysis was performed on SPSS version 20 (SPSS, Chicago, Illinois). Means, standard deviations and coefficient of Variation (CV) scores were analysed for measurements of Achilles tendon length and maximum anteior-posterior (max AP) diameter. Means ± 95% Confidence Intervals are shown for the strain data in table 2. CV's and Pearson correlation analysis (R) were calculated for longitudinal

and transverse CE measures, and for each combination of testing protocol (Daily Measures or 1hr Measures), foot position (fixed or relaxed) and averaged over time (Measure 1, 2, 3, 4, & 5). Intra-class correlation coefficient (ICC) was calculated to determine intra-rater reliability for CE, calculated for each combination of measures and averaged over time.

Results:

The standard deviation scores for measurements of Achilles tendon length and max AP diameter were all low and CV scores for measures of length and max AP diameter were also all acceptably low as shown in table 1 implying a very good level of reliability in these measures.

Table 1 here

For each participant, CE data was collected from 10 ROI's in the longitudinal plane and 4 ROI's in the transverse plane.

The data from each participant was averaged to provide one value from each measurement session.

Scores for Mean ± 95% confidence Interval, CV, R and ICC for the various CE testing procedures are shown in table 2.

Table 2 here

The results demonstrate the CV's for all measures of length and max-AP diameter were all very low indicating a high level of reliability. The CV's for the CE variables however are all above 53%, indicating a low level of reliability among the measures. The correlation values indicate no correlation to weak correlations between the measures at best (P values = 0.38 - 0.60), and the ICC values are all classified in the 'poor' category, again indicating a low level

of reliability between the measures. Some negative ICC values were observed, however as it has been said that negative ICC's have no theoretical legitimacy and should not be quoted (Giraudeau 1996), these values were treated as zero.

Discussion:

The main findings of this paper are that the CV's for the CE variables were all over 53% and the correlation and ICC values were all in the weak or poor categories indicating low levels of reliability. In contrast, the CV's for measures of tendon length and max-AP diameter calculated for conventional ultrasound all indicated a high level of reliability.

A threshold for CV measures of ≤12% was adopted as an acceptable level of reproducibility (Chino et al. 2012). The calculated CV's for measures of length and diameter were all very low, indicating a high level of reliability for these measures, in agreement with previous literature (Ying et al. 2003). The calculated CV's for the CE data however, all exceeded 53%, therefore not classed as acceptable. The correlation values ranged from no correlation to weak correlations, and the ICC values were all classified as poor. These results demonstrate high levels of reliability when CE is used to assess length and diameter of the Achilles tendon, but low levels of repeatability and reproducibility when CE is used to assess strain data (and hence measures of stiffness) of the human Achilles tendon *in vivo*.
CE scanning took place using two different foot positions, with feet relaxed and with feet fixed at 90°. Within CE assessment, scanning using a relaxed foot position is common, as it avoids tendon stress (De Zordo et al. 2009; Chen et al. 2013), however previous research attributed low levels of repeatability with the technique to movement of both the tendon and participant during scanning (Peltz et al. 2013). This study compared both approaches, and the results appear more reliable when the foot was relaxed as opposed to fixed, although both remained poor (see table 1).

The differences between transverse and longitudinal measures were also assessed. The differences in R and ICC scores between longitudinal and transverse measures were very marginal, therefore looking at differences in CV values, whilst remaining poor, transverse measures appeared slightly more reliable than longitudinal measures. This finding was unexpected, given that in previous studies, the reproducibility of CE appears to be higher in longitudinal planes (Drakonaki et al. 2009). Waves propagate along fibres more easily than they do across them (Eby et al. 2013), therefore longitudinal scans should provide more reliable results, especially as transverse scans are more prone to inhomogeneous compression towards the edges of the probe (Klauser et al. 2014). It is important to note that the differences in reproducibility between longitudinal and transverse scans were very marginal. A potential reason for transverse scans being more reproducible than longitudinal scans is that they take in a larger portion of the tendon which could give rise to a more heterogeneous sample of tendon. In contrast, the transverse scans take in a smaller area of the tendon resulting in a more homogenous sample and therefore a lower CV score.

Other research into CE report high inter- and intra-rater reliability, excellent interpretive reliability and a confirmed ability to measure absolute muscle hardness (Wu et al. 2011; Chino et al. 2012). However these studies were assessing the use of CE with either tissue mimicking materials, plantar fascia or muscle and not tendon. The authors of both studies noted major limitations and neither looked at the results obtained across consecutive days. An early study looking at the use of CE with the Achilles tendon (Sconfienza et al. 2010) noted no difference between normal and pathological Achilles tendons at the enthesis, retrocalcaneal bursa or myotendinous junction, however symptomatic tendons did show significantly lower elasticity (by 32%) in the middle portions. This study also demonstrated excellent correlation with grey scale ultrasound and sonoelastography and excellent intraobserver reliability was shown for tendon thickness, echotexture, fragmentation and

interruption. Interobserver reliability ranged from poor to excellent for the same measures, however the reliability of CE itself was only measured by the agreement of the determination of the prevailing colour of the tendon elastogram. Measurements in this study were only taken during one session and were evaluated at the time of measurement and again 6 months afterwards, therefore the actual reproducibility of the technique over consecutive measures was again not assessed.

Despite the previously noted positive findings, other research has questioned the ability of CE to fully assess the properties of tendon, finding the technique to have low reproducibility and be heavily operator dependant (Arda et al. 2011). In this study, a QF was utilised to reduce operator variance as the QF chosen related to an image of very high quality. Furthermore, it cannot be avoided that CE is noted as a purely qualitative technique, which some argue renders it unsuitable for measuring tissue stiffness (Treece et al. 2011), particularly with respect to repeated measures over time. The results of this study imply that CE has a low level of reproducibility for assessing the Achilles tendon and is not suitable for producing longitudinal measures over time. Three separate reliability measures were conducted on the data, with all three indicating very low levels of reliability. The CV values were very high, the strength of correlations were very low and ICC values were also very low. Therefore, the degree of variation in the data was too high to infer statistically, or clinically significant results to the wider population.

This study carries limitations, including a small, homogenous sample size, limiting the extrapolation of the results to the wider population. Due to the extremely high level of variability in the results obtained from only eight subjects, further study was deemed not ethically justifiable. However, a number of separate measures were made on each participant in this study, providing a total of 80 measures for analysis, equating to more or a similar amount of measures analysed in similar research in the Achilles tendon (De Zordo et al.

2009; Drakonaki et al. 2009; Klauser et al. 2013; Tan et al. 2012). Another limitation is the use of a single operator in the study (Obuchowski & Lieber 2008).

There are many different types of elastography available (Hoskins 2012), with other systems recently emerging including ultrasound tissue characterisation (UTC) and shear wave elastography (SWE). As CE only offers a qualitative or semi-quantitative value (strain ratio), there are no absolute values to be obtained with this modality. Some have used retrocalcaneal fat as a reference for strain ratio calculations (Drakonaki et al. 2009), however due to the potential presence of oedema and fibrosis in the fat pad associated with tendinopathy, this will not be a known or fixed variable. An acoustic coupler has been suggested as a more valid reference point (Yamamoto et al. 2015), with recent publications suggesting the technique to be more reproducible when using an acoustic coupler as a reference standard (Yamamoto et al. 2015), but does not state the Young's modulus of this acoustic coupler or the manufacturer. This further limits the clinical utility of the technique, as results can only then be used when access to the specific acoustic coupler was guaranteed. Another issue in this field is that replication of studies using machines from different manufacturers is also difficult due to the lack of standardisation of colour coding of tissue stiffness between difference elastography software.

Alternative methods of assessing tendon are emerging, yet the value of these technologies in the clinical setting remains unknown. These emerging technologies include shear wave elastography which measures tissue elasticity by quantifying the velocity of shear waves as they pass through a tissue (Drakonaki et al. 2012; Hoskins 2012; Ooi et al. 2013; Sarvazyan et al. 2011), and Ultrasound Tissue Characterisation (UTC), designed to assess the structural integrity of tendon (van Schie et al. 2003). More research is needed to assess reproducibility over consecutive measures and consecutive days as well as research that assesses the impact of previous bouts of exercise on the measures obtained and how this relates to tendon

management. It would be beneficial for future research to elaborate on how these technologies can fit into the clinical setting and how this information relates to health, function, strength and injury. A comparison between the different forms of elastography would also be useful.

In conclusion, this study is the first to control for previous activity and examine the repeatability of five consecutive measures and the reproducibility over five consecutive days when using CE to measure the length, diameter and strain data (and hence stiffness) of the Achilles tendon. The results demonstrate that the specific CE system utilised in this study provides good B-mode images and has a high level of reproducibility for assessing Achilles tendon length and diameter. This particular system however was shown to have a low level of reproducibility for measuring strain data (and hence stiffness) of the human Achilles tendon *in vivo.* Before CE is suggested for use in the clinical setting, researchers and clinicians need to be cautious in interpreting the results from previously reported studies that have yet to consider the reproducibility of the technique across repeated measures over days and the influence of prior activity to the results obtained.

Acknowledgements: N/A

1. Conflict of Interest: The authors declare that they have no conflict of interest

2. Ethical Approval: All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

3. Informed Consent: Informed consent was obtained from all individual participants included in the study.

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Supplementary Material: N/A

Figure Legends:

Figure 1 Title: **Figure 1: Photograph demonstrating the scanning position of participants, in the prone position with feet hanging clear of the examination table. Photograph also demonstrates probe positioning during examination of the right Achilles tendon in the relaxed foot position.**

Figure 2 Title: **Figure 2: B-Mode Ultrasound Extended field of view Image of the Right Achilles Tendon of a 26 year old female participant. The Achilles tendon (A) is shown between the solid white lines. Achilles tendon length (shown by the dotted white line) was measured as the length between the insertion of the Achilles tendon at the calcaneus (B) and the lowest fibres of the soleus muscle (C). The mid-point of the tendon (D) is also shown.**

Figure 3 Title: **Figure 3: Photograph demonstrating the use of the 9L4 probe in the longitudinal plane for obtaining elastograms. The transducer position is shown parallel to the tendon and between the markings for calcaneus and musculo-tendinous junction.**

Figure 4 Title: **Figure 4: Longitudinal elastogram of the Right Achilles tendon taken from a 30 year old male participant. The 10 Regions of Interest (ROI's) used to collect CE data when imaging in the longitudinal plane are shown by the yellow dotted circles. The corresponding values for strain are shown in the box at the right hand side of the image. Note the Quality Factor (QF) of 80 circled in white at the bottom of the image.**

Figure 5 Title: **Figure 5: Transverse elastogram of the Right Achilles tendon taken from a 30 year old male participant. The 4 Regions of Interest (ROI's) used to collect CE data when imaging in the transverse plane are shown by the yellow dotted circles. The corresponding values for strain are shown in the box at the bottom of the image.**

Table 1 Title: **Table 1: Analysis of Achilles tendon length and diameter measurements (mm) including raw data, averages, standard deviations and coefficient of variation taken from each participant over each differing testing protocol.**

Table 2 Title: **Table 2: Analysis of CE data from each testing protocol including coefficient of variation (CV), Pearson's correlation coefficient (R) and Intra-class Correlation Coefficient (ICC). Classification scales are also shown.**

Tables:

Table 1:

AT Length (mm)																
	1 hr								Daily							
Participant	1	2	3	4	5	Ave	SD	CV	1	2	3	4	5	Ave	SD	CV
1	37.6	33.4	34.3	34.5	35.0	35.0	1.6	4.5%	34.5	32.3	33.3	34.1	33.7	33.6	0.8	2.5%
2	59.5	57.0	57.8	59.2	58.3	58.4	1.0	1.7%	59.0	59.4	58.2	57.8	58.3	58.5	0.6	1.1%
3	38.6	41.5	38.7	39.2	40.2	39.6	1.2	3.1%	39.5	39.7	41.5	38.7	38.6	39.6	1.2	2.9%
4	45.2	45.9	44.3	46.2	45.5	45.4	0.7	1.6%	45.7	45.2	44.7	46.0	45.0	45.3	0.5	1.2%
5	55.8	56.2	55.7	54.9	55.2	55.6	0.5	0.9%	56.4	55.5	54.9	55.7	55.2	55.5	0.6	1.0%
6	69.9	68.7	67.9	68.2	69.3	68.8	0.8	1.2%	69.4	68.8	67.7	68.2	69.0	68.6	0.7	1.0%
7	29.6	30.5	31.2	29.7	30.9	30.4	0.7	2.3%	29.9	31.1	30.5	30.9	31.4	30.8	0.6	1.9%
8	53.7	54.3	53.5	53.9	54.7	54.0	0.5	0.9%	54.0	55.7	55.1	53.9	54.2	54.6	0.8	1.4%
Max A-P Diameter (mm)																
	1 hr								Daily							
Participant	1	2	3	4	5	Ave	SD	CV	1	2	3	4	5	Ave	SD	CV
1	5.1	4.9	5.0	4.9	5.0	5.0	0.1	1.7%	4.9	4.9	5.0	4.9	5.0	4.9	0.1	1.1%
2	4.4	4.4	4.5	4.5	4.4	4.4	0.1	1.2%	4.4	4.5	4.5	4.4	4.4	4.4	0.1	1.2%
3	4.7	4.8	4.9	4.8	4.7	4.8	0.1	1.8%	4.8	4.8	4.8	4.7	4.7	4.8	0.1	1.2%
4	4.6	4.7	4.7	4.6	4.6	4.6	0.1	1.3%	4.6	4.6	4.7	4.6	4.6	4.6	0.0	1.0%
5	5.7	5.7	5.8	5.7	5.7	5.7	0.1	1.0%	5.8	5.7	5.7	5.7	5.7	5.7	0.0	0.8%
6	5.3	5.4	5.3	5.5	5.3	5.4	0.1	1.9%	5.3	5.4	5.4	5.3	5.4	5.4	0.1	1.0%
7	4.0	4.2	4.1	4.1	4.0	4.1	0.1	2.4%	4.0	4.1	4.1	4.1	4.0	4.1	0.1	1.3%
8	4.6	4.5	4.6	4.6	4.6	4.6	0.0	1.0%	4.6	4.6	4.6	4.5	4.6	4.6	0.0	1.0%

Table 2:

	Mean	CV	R	ICC
Longitudinal Fixed 1hr	0.010 ± 0.007	111.5%	0.25 (p=0.49)	0.11
Longitudinal Fixed Daily	0.014 ± 0.009	92.8%	0.04 (p=0.60)	0.01
Longitudinal Relaxed 1 hr	0.007 ± 0.005	105.7%	0.13 (p=0.53)	0.06
Longitudinal Relaxed Daily	0.009 ± 0.003	80.8%	0.07 (p=0.38)	0.10
Transverse Fixed 1hr	0.010 ±0.007	112.4%	-0.06 (p=0.49)	0.00
Transverse Fixed Daily	0.010 ±0.003	80.4%	0.03 (p=0.38)	0.02
Transverse Relaxed 1 hr	0.007 ±0.001	53.6%	-0.04 (p=0.50)	0.00
Transverse Relaxed Daily	0.008 ±0.002	60.9%	0.01 (p=0.42)	0.11

CV = Coefficient of Variation, R = Pearson's correlation coefficient, ICC = Intra-class Correlation Coefficient

Scale for correlations: 0 = no correlation, 0.1-0.3 = weak, 0.4-0.6 = moderate, >0.7 = strong and 1 = perfect (Dancey, C, P., & Reidy 2004)

Scale for ICC results: 0.00 - 0.20 = Poor, 0.20-0.40 = Fair, 0.40-075 = Good, >0.75 = Excellent (Drakonaki et al. 2009; Chino et al. 2012).

Figures:

Figure 1:

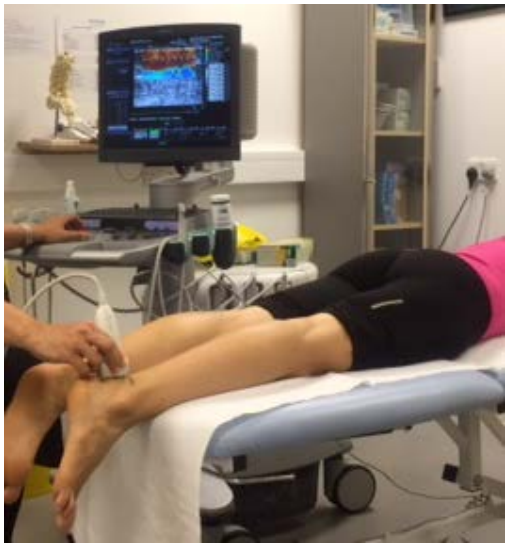

Figure 2:

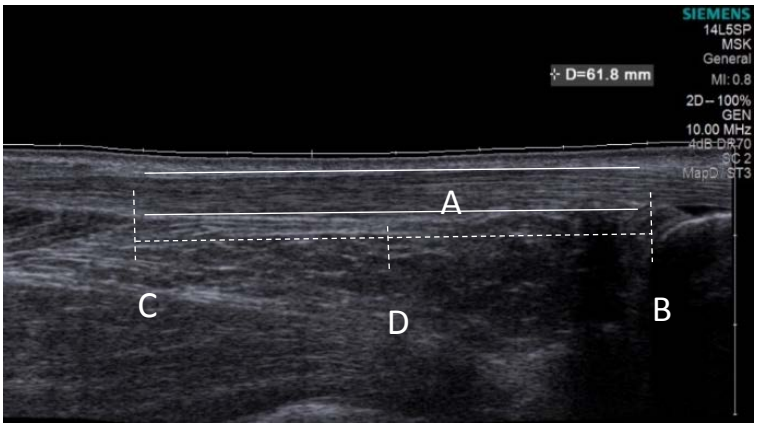

Figure 3:

Figure 4:

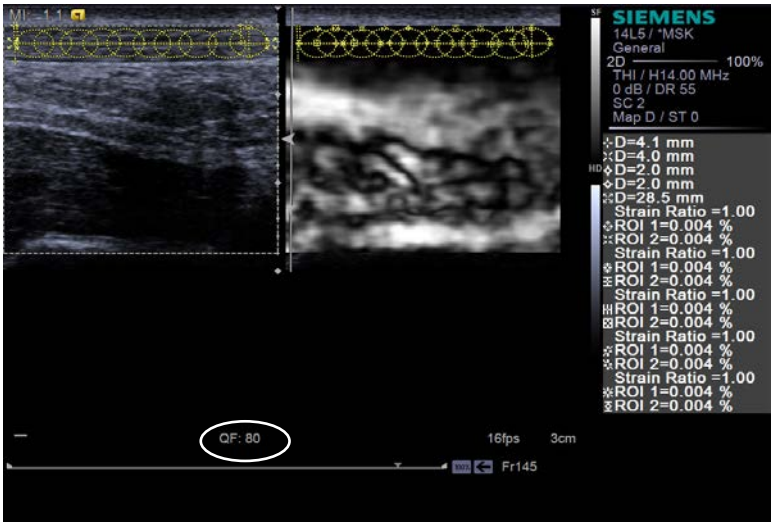

Figure 5: