

## SOURCES OF ERROR WHEN MEASURING ACHILLES TENDON MECHANICS DURING RUNNING ACTIVITIES

Giorgos Krikelis, Matthew T. G. Pain and Laura-Anne M. Furlong

School of Sport, Exercise and Health Sciences, Loughborough University, UK

Accurate measurements of tendon mechanics are necessary for biomechanists when trying to identify injury risk factors, optimise athletic performance and develop musculoskeletal models. Measuring Achilles tendon (AT) mechanics dynamically is now possible by combining motion capture and ultrasound (US). The aim of this study was to quantify sources of error when measuring AT length using motion capture and US, and establish their effect on calculated strain values. Errors in AT insertion tracking and data synchronisation caused differences in AT length and moment arm of  $5.3 \pm 1.1$  mm and  $11.2 \pm 0.9$  mm, respectively; this decreased calculated AT peak strain from  $11.6 \pm 3.5\%$  to  $5.4 \pm 2.5\%$ . These differences could significantly impact a researcher's interpretation of the effects of footwear, technique, and specific kinematics on AT loading.

**KEYWORDS:** Plantarflexors, ultrasound, methods

**INTRODUCTION:** Tendons are viscoelastic force transmitters whose properties are commonly measured by combining dynamometry and ultrasound (US) during isolated isometric contractions. The technological advancement of US has enabled measuring tendon movement during dynamic sporting activities such as running. Technical issues inherent to dynamic methods, such as system synchronisation, marker and probe movement, calibration accuracy, and mis-tracking of key anatomical landmarks, have previously been reported when estimating variables like strain, stiffness and hysteresis (Lichtwark, 2005; Farris, Trewartha and McGuigan, 2012). Finni et al. (2013) emphasised the importance of accurately synchronising force and US data, as failing to do so led to errors of up to 5% and 20% when measuring Achilles tendon (AT) stiffness and hysteresis, respectively. The cumulative effect of such errors when calculating AT properties can lead to false coupling of AT mechanics to specific lower leg kinematics, and identification (or not) of them as potential risk factors of injury. *In vivo* methods that accurately measure dynamic AT mechanics can inform evidence-based plyometric training activities and facilitate identifying potential injurious lower leg kinematics. The aim of this study was to quantify sources of error when measuring AT length with the use of motion capture and US, and establish their effect on calculated strain values. Quantifying these errors would benefit biomechanists interested in measuring AT mechanics during running. Understanding methodological limitations in greater detail would facilitate improved interpretation of results to inform both performance and injury prevention applications.

**METHODS: Data acquisition:** Following institutional ethical committee approval, 15 recreationally active male subjects provided written informed consent to participate in this study (age:  $30.9 \pm 7.1$  years; mass:  $75.61 \pm 11.3$  kg; height:  $1.78 \pm 0.07$  m). All data were acquired from the subject's preferred kicking limb, and subjects had no history of lower limb/shank or foot surgery. Three-dimensional motion (250 Hz, Vicon Vantage, Oxford Metrics Ltd., Oxford, UK) and force (2000 Hz, OR6, AMTI, Watertown, MA, USA) data were acquired during three running trials along a 10 m lane at a comfortable running speed ( $2.9 \pm 0.2$  m/s). Markers, 14 mm diameter, were placed on the toe, 1st metatarsophalangeal (MTP) joint, 5th MTP joint, heel, medial and lateral malleolus, medial and lateral epicondyle of femur of the preferred side, and right and left anterior superior iliac spine, right and left posterior superior iliac spine, xiphoid process, jugular notch, right and left acromion process and C7 vertebrae. A 128-element linear probe (LV7.5/60/128Z-2; LS128 CEXT-1Z; Telemed, Vilnius, Lithuania) secured using self-adhesive bandages was used to acquire B-mode US data (7-8 MHz frequency; 65 mm field of view; 65mm image depth, 80dB dynamic range) of the region surrounding the medial gastrocnemius muscle-tendon junction (MTJ). Three motion analysis markers were attached non-collinearly on a plastic cast firmly secured to the probe, allowing

the US image plane orientation to be calculated in the global coordinate system. The US image plane was calibrated using the protocol described by Lichtwark and Wilson (2005) and Farris, Trewartha and McGuigan (2012). Images were recorded and stored as a video sequence on a computer (through a USB link) at 61 frames per second.

**Data analysis:** MTJ position was manually tracked and digitised on the US video frames by the same investigator, using a custom written MATLAB function (The MathWorks, Inc., Natick, MA, USA). Through the transformations matrices acquired from the US image calibration, the MTJ position was initially expressed in the US probe's local coordinate system, and translated and inversely rotated to the global coordinate system. AT length was defined as the magnitude of the AT vector formed by the digitised MTJ position and its insertion on the calcaneus (represented by the reflective marker on the heel). The same vector was used to describe the AT line of action ( $AT_{LOA}$ ). Following the approach of Lichtwark and Wilson (2006), slack length of the AT was defined as the average length during toe-off. This approach is in line with findings from previous *in vivo* studies, where AT force was directly measured during running and found to be very close to zero at toe-off (Komi, 1990). AT strain was defined as the percent AT length change from its slack length. The ankle centre of rotation was defined as the midpoint between the lateral and medial malleolus markers. Achilles tendon moment arm ( $AT_{MA}$ ) was calculated as the perpendicular distance from the ankle center of rotation to the  $AT_{LOA}$ .

**Source of error:** When calculating AT length, three sources of error were investigated. AT length, strain and  $AT_{MA}$  were calculated for all possible combinations of calcaneal insertion position (CIP) corrections applied and delayed/ synchronised data.

**1. Digitising, US calibration, and marker movement errors:** Coefficients of variation (CV) were calculated for the digitized x and y coordinates, corresponding to the direction that the MTJ is moving along during running and its depth in the US image. The accuracy of projecting these digitised points in the global coordinate system was estimated by scanning a single Perspex® sheet with grooves curved along its length immersed in a container filled with water, whose features could be tracked by both the US and the motion capture system using a thin metallic rod with reflective markers attached (Lichtwark and Wilson, 2005). Finally, the error due to potential marker movement on the probe's rig and soft tissue movement was estimated, as well as the error inherent in the motion capture system when tracking moving markers. The cumulated error in AT length was quantified using equations described by Taylor (1997).

**2. CIP related errors:** To acquire a better estimate of the CIP, the coordinates of the reflective marker on the heel were corrected for the reflective marker's size (14 mm diameter) and skin/fat thickness (measured on MRI scans). Farris, Trewartha and McGuigan (2012) estimated that the error due to the CIP had small effect on AT length and  $AT_{MA}$  at a 90° angle between  $AT_{LOA}$  and the foot. Therefore, it was investigated whether the error becomes substantial at other angles and whether correcting at 90° direction in every ankle angle affects AT length and  $AT_{MA}$ . Hence, the heel marker position was corrected by shifting the calcaneal position by 7 mm + tissue thickness towards the calcaneus at a) 90° relative to the  $AT_{LOA}$ , and b) the direction of the long axis of the foot vector (from heel marker to metatarsal markers midpoint).

**3. Delay between US first frame and TTL pulse:** Motion analysis and US were synchronised via a 5V TTL pulse sent from the beam former, as soon as the US was recording data. The 5V pulse was recorded in Vicon at 2000 Hz. To estimate whether there was a delay between the first frame that was logged on the US computer screen and the first data point of TTL pulse recorded in Vicon a temporal calibration process was applied using a separate experimental setup. The TTL pulse was used to trigger a frequency generator, transmitting a 6 MHz sinewave on a wire coiled around the US probe's cable, causing enough electromagnetic interference to distort the US image. The interference was delayed by a discrete amount every time the US was restarted. As the delay was increased, the distortion started to appear 'later' on the first frame. With the US recording at ~31, ~61 and ~124 fps, the 'effective' delay was determined to the nearest 1 ms, as the one where the distortion could be seen throughout the whole length of the first frame. The effect of the delay on the calculated AT length was investigated only on the kinematic level (AT length change).

**Statistical analysis:** A one-way repeated measure ANOVA (Greenhouse-Geisser correction applied if necessary and a Bonferroni adjustment was used for post-hoc analysis; alpha level  $\alpha \leq 0.05$ ) was used to determine whether statistically significant differences existed in peak strains elicited from method variations of measuring AT length (6 in total). The comparison was conducted between: delayed US data relative to kinematics with no CIP corrections (1), only marker size correction (2), both CIP corrections (3), and synchronised kinematic and US data with no CIP corrections (4), only marker size correction (5), both CIP corrections (6). All CIP corrections were applied with method b as described above in CIP related errors section.

**RESULTS:** The calibration error was estimated as  $0.67 \pm 0.42$  mm at 2 cm depth in the US image. The average error due to marker movement combined with digitising error corresponded to  $1.0 \pm 0.7$  mm (max: 3.5 mm) AT length error. These three sources of error combined resulted in  $1.5 \pm 0.5$  mm (max: 3.7 mm) AT length error. Correcting CIP in a direction parallel to the long axis of the foot (method b) compared to correcting it in a  $90^\circ$  direction relative to the  $AT_{LOA}$  (method a), resulted in  $3.6 \pm 0.8$  mm AT length RMSD (max: 5.8 mm) and  $0.17 \pm 0.05$  mm  $AT_{MA}$  RMSD (max: 0.3 mm). For the current setup used, the effective delay for each framerate was estimated as the corresponding frame duration + 4ms. Differences when applying CIP corrections and synchronising the data can be seen in Table 1.

Method variations caused statistically significant differences in peak AT strain. Sample data from one trial are shown in Figure 1. Post hoc analysis revealed statistically significant differences in calculated peak strain for out of all variations (with one exception) of acquiring AT length, with  $p \leq 0.023$  for all post-hoc comparisons. Only variations 3 (delayed data with both CIP corrections) and 4 (synchronised data without any CIP corrections) did not reach statistical significance.

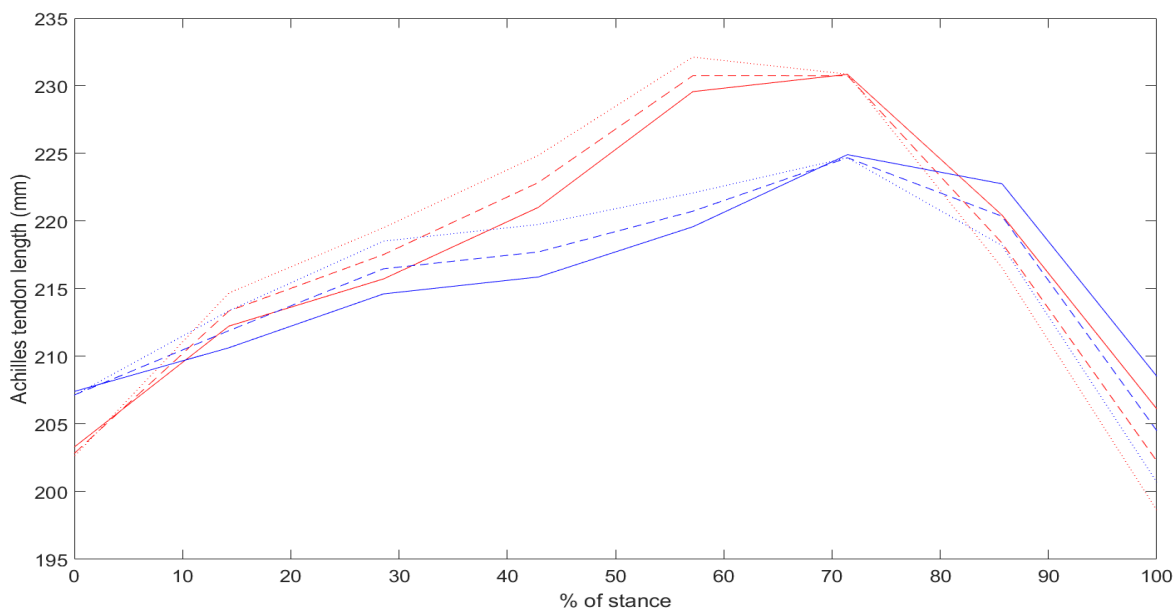
**Table 1. Influence of CIP and synchronisation corrections on AT length,  $AT_{MA}$  and AT peak strain: root mean square differences between corrections applied. AT length and moment arm results are presented as group mean  $\pm$  standard deviation (maximum for the entire group in brackets).**

|                     | No CIP corrections vs.<br>both CIP corrections applied | Delayed vs. synced<br>US-marker data                   |
|---------------------|--|--|
| AT length RMSD (mm) | $3.8 \pm 0.8$ (6.1)                                    | $3.6 \pm 0.8$ (6.3)                                    |
| $AT_{MA}$ RMSD (mm) | $11.1 \pm 0.8$ (12.8)                                  | $1.8 \pm 0.4$ (3.3)                                    |
| AT peak strain (%)  | $8.7 \pm 2.5\%$ to $5.4 \pm 2.5\%$<br>(14.3% to 12.0%) | $8.2 \pm 3.3\%$ to $5.4 \pm 2.5\%$<br>(15.2% to 12.0%) |

**DISCUSSION:** The aim of this study was to quantify various sources of error when measuring AT length with the use of motion capture and US, and establish their effect on calculated strain values. Combined digitising, US calibration, and marker movement errors varied calculated AT lengths by  $1.5 \pm 0.5$  mm. When combined with errors due to US system delay and mis-tracking of the CIP, strain values were significantly decreased for all participants.

Contrary to past reports (Farris et al., 2012), correcting CIP altered both AT length and  $AT_{MA}$  values (average RMSD:  $3.8 \pm 0.8$  mm and  $11.1 \pm 0.8$  mm, respectively). The method of correcting CIP had a small effect on  $AT_{MA}$  but the difference it caused on AT length values could not be dismissed (average RMSD:  $3.6 \pm 0.8$  mm). It was deemed correcting CIP along the direction of the long axis of the foot was more appropriate, as there is no reason to expect that  $AT_{LOA}$  would be at  $90^\circ$  to the foot in all ankle positions. These results show the importance of accurately tracking CIP when trying to understand AT mechanics.

Incorrectly synchronised motion capture and US data caused major differences in the results, as the US data were then matched with incorrect lower leg kinematics. The effect on AT length did not seem greater in magnitude ( $3.6 \pm 0.8$  mm) than CIP corrections, however, it caused large decreases in estimated AT strain from  $8.2 \pm 3.3\%$  to  $5.4 \pm 2.5\%$ , values which are closer to those previously reported during isometric and in vitro studies. Although establishing AT slack length is another methodological challenge (Farris et al., 2012), these results still show how existing delays in US and motion capture data can alter these relative strain values.



**Figure 1. Example graph from one trial showing the effect of CIP corrections and data synchronisation on AT length. Synchronised (blue) and delayed (red) curves presented without CIP corrections (dotted lines), only with marker size corrections (dashed lines) and with both CIP corrections (solid lines).**

In a worst-case scenario, comparing trials where neither CIP nor data delay issues were addressed with those that were, AT length and  $AT_{MA}$  would be different by  $5.3 \pm 1.1$  mm and  $11.2 \pm 0.9$  mm, respectively. Similarly, AT peak strain would be measured as  $11.6 \pm 3.5$  % instead of  $5.4 \pm 2.5$  % when CIP corrections were applied and data properly synchronised. Such large errors would throw AT strain values into unrealistic physiological ranges, considering past *in vitro* studies have reported AT failure strains being  $\sim 10$  % (Wren et al., 2001). This has significant implications when interpreting AT movements and properties and attempting to relate these to lower leg kinematics.

**CONCLUSIONS:** Development of US and motion capture in recent years has provided significant insight into *in vivo* muscle and tendon function. However, the results of this study show errors due to synchronisation delays between US and motion capture data, and in inaccurate tracking of the CIP, significantly influence calculated AT mechanics. These errors may cause erroneous characterisation of lower mechanics in sporting activities, change our perception of the effect of the AT's elastic properties in athletic performance and misinform musculoskeletal models used in a plethora of biomechanical and engineering research fields, such as prosthesis or rehabilitation equipment designs.

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