*Manuscript

A method to characterize *in vivo* tendon force-strain relationship by combining ultrasonography, motion capture and loading rates

[1]Pauline Gerus, [1] Guillaume Rao, and [1] Eric Berton

[1]*Institute of Movement Sciences E-J Marey, Aix-Marseille Université, Marseille, France*

Address correspondence to:

Pauline GERUS

Institute of Movement Sciences EJ. Marey, UMR CNRS 6233

Aix-Marseille Université

Parc Scientifique et Technologique de Luminy,

163, Avenue de Luminy,

13288 Marseille cedex 09, France

Phone : + 33 491 17 04 78

Fax : + 33 491 17 22 52

Email : pauline.gerus@univmed.fr

Keyword: Ultrasound, Gastrocnemius, tendon length, mechanical characterization, loading rate

Abstract

The ultrasonography contributes to investigate *in vivo* tendon force-strain relationship during isometric contraction. In previous studies, different methods are available to estimate the tendon strain, using different loading rates and models to fit the tendon force-strain relationship. This study was aimed to propose a standard method to characterize the *in vivo* tendon force-strain relationship. We investigated the influence on the force-strain relationship for *medialis gastrocnemius* (MG) of 1) one method which takes into account probe and joint movements to estimate the instantaneous tendon length, 2) models used to fit the force-strain relationship for uniaxial test (polynomial vs. Ogden), and 3) the loading rate on tendon strain. Subjects performed ramp-up contraction during isometric contractions at two different target speeds: 1.5 sec and minimal time with ultrasound probe fixed over the muscle-tendon junction of the MG muscle. The used method requires three markers on ultrasound probe and a marker on calcaneum to take into account all movements, and was compared to the strain estimated using ultrasound images only. The method using ultrasound image only overestimated the tendon strain from 40% of maximal force. The polynomial model showed similar fitting results than the Ogden model (R^2 = 0.98). A loading rate effect was found on tendon strain, showing a higher strain when loading rate decreases. The characterization of tendon force-strain relationship needs to be standardized by taking into account all movements to estimate tendon strain and controlling the loading rate. The polynomial model appears to be appropriate to represent the tendon force-strain relationship.

Introduction

Ultrasonography (US) associated with force measurements contributes to estimate the tendon force-strain relationship during muscle contraction (Maganaris, 2002) and allows to compare populations differing on age (Kubo et al., 2000) or sex (Burgess et al., 2008). Consequently, a method that accurately estimates the tendon force-strain relationship is essential.

During isometric contractions meaning that the muscle-tendon complex is held at constant length, firmly fixing the foot on plate does not prevent from movement of the body. Different methods were used to estimate *in vivo* muscle-tendon junction (MTJ) displacement while minimizing the measurement errors due to joint and probe movements. One method consists in using a skin marker and fixing the ultrasound probe (Burgess et al., 2008) on the leg but it leads to overestimate the MTJ elongation (Muramatsu et al., 2001). Another method called passive correction (Maganaris, 2005) using additional measurements during passive task leaded to inaccurate elongation value (Maganaris, 2005). An alternative method (Maganaris et al., 2005) requiring the use of two ultrasound probes over the MTJ and the calcaneum remains restrictive because of the second probe. These previous methods lead to either erroneous MTJ elongation or need additional tasks or US devices. A previous method combining motion capture and ultrasonography and providing an easy way to estimate the instantaneous tendon length has been used for dynamics tasks (Lichtwark and Wilson, 2005). However, this promising method hasn't been used during isometric contractions whereas this particular task is often used to characterize the tendon mechanical properties.

The tendon presents a non-linear behavior that could be represented in Finite Elements Modeling (FEM) by a hyperelastic mechanical model, such as Ogden formulation (Cheng and Gan, 2008, Shibata et al., 2006). *In vivo* force-strain relationships were also fitted using a second-order polynomial function (Burgess et al., 2008, Pearson et al., 2007) based on good fitting with experimental data. It is not known whether the use of a second-order polynomial fit is able to catch all the features of the force-strain relationship in comparison with an Ogden model.

The tendon force-strain relationship has been previously estimated during different loading rates meaning that different ramp-up contraction durations were used. The presence of loading rate effect on tendon could have important implications to test and compare the mechanical properties and needs to be quantified for further studies on tendon mechanical characterization.

This study was aimed to propose a standard method to characterize the *in vivo* tendon force-strain relationship. Thus we investigated the influence on the force-strain relationship for *medialis gastrocnemius* (MG) of 1) one method which takes into account probe and joint movements to estimate the instantaneous tendon length, 2) models used to fit the force-strain relationship for uniaxial test (polynomial vs. Ogden), and 3) the loading rate on tendon strain. We hypothesize that these factors will affect the overall characterization of the tendon and need to be included when estimating the *in vivo* tendon force-strain relationship during isometric contraction.

Method

Experimental protocol

Eight healthy males (26.0 ± 1.5 years, 73.9 ± 9.6 kg, 1.77 ± 0.05 m) gave their consent to participate. Subjects were seated on the bench of a custom ergometer with the knee fully extended and the sole of the foot perpendicular to the shank. The right foot was set in a rigid shoe fixed to a plate with both joint and static torquemeter (CS3, Meiri) axis aligned. The net ankle joint moment was sampled at 2000 Hz.

Subjects performed three 3sec maximal voluntary isometric contraction (MVIC) plantarflexion trials with verbal encouragement and on-screen joint moment feedback. Then they performed ramp-up contractions (from rest to 100% MVIC) in 1.5 sec ($C_{1.5}$), and in minimal time (C_{max}) with on-screen feedback on the expected moment. Three trials were recorded for each condition with a resting time of 2 min.

A 50mm linear array ultrasound probe (BK-Med-Pro-Focus) at 10 MHz was positioned over the MTJ of the MG muscle. The probe was carefully attached to the leg using foam fixation and secured with elastic bandage. Images were recorded at 30 Hz with an image acquisition device (NI, PCI 1410).

Three markers were attached to the probe in order to create a probe reference frame (PRF) parallel to the image plane (Figure 1). Additional markers were attached to the Achilles' Tendon insertion over the calcaneum, the first metatarsal head of the foot, the lateral and medial malleoli, and the fibula head. Markers positions were recorded at 125 Hz (Vicon System) and synchronized with US data.

Estimation of *in vivo* tendon strain

US images were used to track the MTJ defined as the intersection between the MG muscle fascicles and the MG tendon. Raw images were processed by adjusting the contrast and then binarized using a graythreshold value. The MTJ displacements were tracked using the method presented by Korstanje (2010) with manual adjustment when necessary. All possible pixel displacements within the search region were evaluated with normalized cross-correlation, while the higher value is used to determine the MTJ position on the next frame.

On each frame, two different methods were used to estimate the tendon elongation (Δl). The Method 1-UIRF consisted in computed Δl in the US Image Reference Frame as the difference between the MTJ position during the contraction and the MTJ position at rest (Burgess et al., 2008; Duclay et al., 2009). In the method 2-PRF, the tendon length (l_t) was computed as the distance between the markers placed on calcaneum and the MTJ position in the global coordinate system using the PRF. Δl was defined as the difference between l_t and the initial tendon length (l_0).

The same value of l_0 was used to compute the tendon strain for the two methods.

The tendon strain was defined as: $$\varepsilon^T = \frac{\Delta l}{l_0} \qquad (1)$$

Estimation of *in vivo* normalized force-strain relationship

The relative moment contribution of the MG to the ankle net joint moment and the moment arm of MG were assumed constant (Kubo et al., 2004). Consequently, The normalized tendon force ($\overline{F_T}(t)$) was computed as the measured joint moment normalized by its maximum value during the task.

Two different models were used to represent the tendon force-strain relationship:

(1) a second-order polynomial model:

$$\overline{F}_T(t) = a\,\varepsilon^T(t)^2 + b\,\varepsilon^T(t) \qquad (3)$$

With a and b parameters adjusted using Matlab.

(2) a second-order hyperelastic Ogden model:

$$\overline{F}_T(t) = \frac{2\mu_1}{\alpha_1}((\varepsilon^T(t)+1)^{\alpha 1-1} - (\varepsilon^T(t)+1)^{(-\alpha 1/2)-1}) + \frac{2\mu_2}{\alpha_2}((\varepsilon^T(t)+1)^{\alpha 2-1} - (\varepsilon^T(t)+1)^{(-\alpha 2/2)-1}) \quad (4)$$

With μ_1, μ_2, α_1 and α_2 parameters adjusted using ABAQUS (Dassault-Systems, v. 6.82).

Data analysis and statistics

The fits between experimental data and each model were compared using R^2 (squared correlation coefficient) and Root Mean Square (RMS_{error}) values.

The tendon strain was estimated using the two methods (1-UIRF & 2-PRF) every 10% of maximal force for each condition (C_{max} and $C_{1.5}$) and each model (Ogden and polynomial).

Two-factor ANOVA (*conditions* vs. *force levels*) was conducted on tendon strain to compare $C_{1.5}$ and C_{max}. Two-factor ANOVA (*methods* vs. *force levels*) and (*models* vs. *force levels*) were conducted on tendon strain. Two-factor ANOVA (*models* vs. *methods*) were conducted on RMS_{error} and R^2 values. A significance level of 0.05 was used for all comparisons and Newman-Keuls Post-Hoc testing was used whenever necessary.

Results

The estimation of *in vivo* tendon strain was significantly higher from 40% to 100% of maximal force for the method 1-UIRF (maximal strain: 9.4 ±3.5 %) compared to the method 2-PRF (maximal strain: 7.8 ±2.2 %).

No difference on RMS_{error} (mean value: 3.5 ± 1.7), on R^2 (mean value: 0.98 ± 0.02), and on tendon strain was found between polynomial and Ogden models.

Whatever the model (polynomial or Ogden), the tendon strain was significantly higher for $C_{1.5}$ compared to C_{max} above 20% of maximal force (Table 2 and Figure 2-3).

Discussion

The main findings of this study were that (1) using the Method 1-UIRF overestimated the tendon strain from 40% of maximal force, (2) the polynomial model showed similar results than an Ogden hyperelastic model, and (3) the loading rate influences the MG tendon strain.

Relative to the method 2-PRF, using the Method 1-UIRF overestimated the tendon strain from 40% of normalized force despite the ultrasound probe being firmly fixed on the leg and the small ankle joint rotation (Table 1). This result agrees with previous studies (Arampatzis et al., 2008; Muramatsu et al., 2001) and confirms the necessity of another methodology to measure tendon strain. In this study, the movements of the Achilles' tendon insertion were taken into account in the tendon length estimation using marker on the calcaneum. This solution gives similar results than using an ultrasound probe over the tendon insertion (Maganaris, 2005), but without needing a supplementary probe. Thus, the method requiring three markers on ultrasound probe and one marker on Achilles' tendon insertion could be easily implemented to directly estimate tendon strain during isometric and dynamic tasks (Lichtwark et al., 2005).

The second-order polynomial model has shown similar good fitting results without difference in estimated tendon strain relative to the Ogden model. Thus, if the mechanical characterization of the tendon is not dedicated to a further use in FEM, a second-order polynomial model could be an easy to implement and suitable physiological representation of the non-linear behavior of tendon.

The maximal tendon strain obtained with the method 2-PRF was in the range of other study (Pearson et al., 2007). In addition, as reported by Pearson et al. (2007) on the patellar tendon, a loading rate dependency was found above 20% of maximal force on the MG tendon strain when comparing two different ramp-up time durations. Thus, the standardization of ramp-up time duration is of high importance when characterizing the tendon mechanical properties. This tendon property is even more important and needs to be taken into account for studies of highly dynamic tasks such as hopping, jumping or running because the tendon become stiffer.

To conclude, the tendon force-strain relationship have to be estimated using a method that takes into account (1) joint and probe movements by combining for example ultrasound and motion capture, and (2) a controlled ramp-up time and allows a standardized characterization of the tendon properties. The second-order polynomial model appears to be appropriated to model the tendon force-strain relationship during uniaxial test.

Conflict of interest statement

None declared.

References

Arampatzis, A., Monte, G. D., Karamanidis, K., 2008. Effect of joint rotation correction when measuring elongation of the gastrocnemius medialis tendon and aponeurosis. Journal of Electromyography Kinesiology, 18(3), 503–508.

Cheng, H.-Y. K., Lin, C.-L., Wang, H.-W., Chou, S.-W., 2008. Finite element analysis of plantar fascia under stretch-the relative contribution of windlass mechanism and achilles tendon force. Journal of Biomechanics, 41(9),1937–1944.

Burgess, K. E., Graham-Smith, P. and Pearson, S. J., 2008. Effect of acute tensile loading on gender-specific tendon structural and mechanical properties. Journal of Orthopaedics Research. 27, 510-6

Duclay, J., Martin, A., Duclay, A., Cometti, G., Pousson, M., 2009. Behavior of fascicles and the myotendinous junction of human medial gastrocnemius following eccentric strength training. Muscle Nerve, 39(6), 819–827.

Korstanje, J.-W. H., Selles, R. W., Stam, H. J., Hovius, S. E. R., Boshc, J. G., 2010. Development and validation of ultrasound speckle tracking to quantify tendon displacement. Journal of Biomechanics, 43(7), 1373–1379.

Kubo, K., Akima, H., Ushiyama, J., Tabata, I., Fukuoka, H., Kanehisa, H. and Fukunaga, T. 2004. Effects of 20 days of bed rest on the viscoelastic properties of tendon structures in lower limb muscles. Br J Sports Med. 38, 324–330.

Kubo, K., Kanehisa, H., Kawakami, Y. and Fukunaga, T. 2000. Elasticity of tendon structures of the lower limbs in sprinters. Acta Physiol Scand. 168, 327–335.

Lichtwark, G. A., Wilson, A. M., 2005. In vivo mechanical properties of the human achilles tendon during one-legged hopping. Journal of Experimental Biology, 208(24), 4715–4725.

Maganaris, C. N. 2002. Tensile properties of in vivo human tendinous tissue. Journal of Biomechanics, 35(8), 1019–1027.

Maganaris, C. N. 2005. Validity of procedures involved in ultrasound-based measurement of human plantarflexor tendon elongation on contraction. Journal of Biomechanics, 38(1), 9–13.

Muramatsu, T., Muraoka, T., Takeshita, D., Kawakami, Y., Hirano, Y. and Fukunaga, T. 2001. Mechanical properties of tendon and aponeurosis of human gas-trocnemius muscle in vivo. J Appl Physiol. 90, 1671–1678.

Pearson, S. J., Burgess, K. and Onambele, G. N. L. 2007. Creep and the in vivo assessment of human patellar tendon mechanical properties. Clin Biomech. 22, 712–717.

Shibata, T., Botsis, J., Bergomi, M., Mellal, A. et Komatsu, K. 2006. Mechanical behavior of bovine periodontal ligament under tension-compression cyclic displacements. European Journal of Oral Science, 114(1), 74–82.

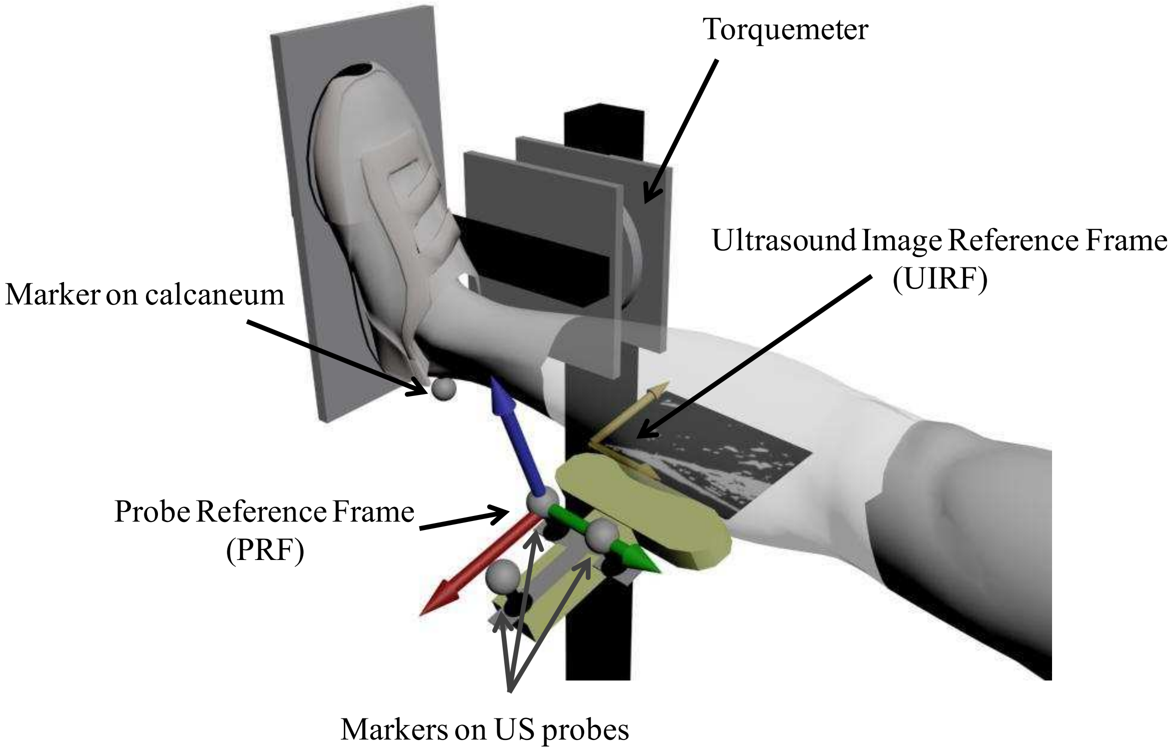

Fig. 1. Representation of the reference frames used in this study for estimating the *in vivo* tendon strain. Three markers positioned on ultrasound probe were used to define the Probe Reference Frame (PRF) while the Ultrasound Image Reference Frame was accessible directly from the US acquisition device.

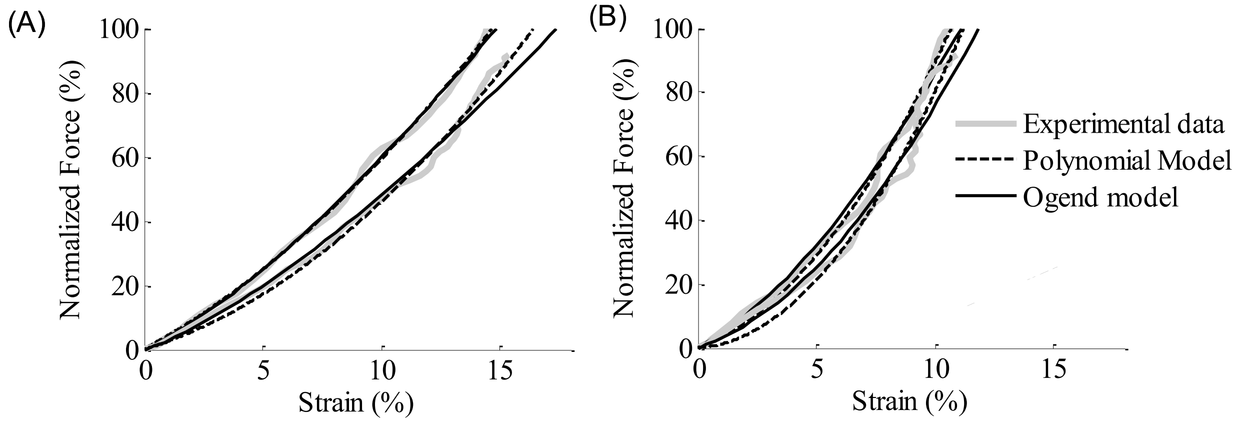

Fig. 2. *In vivo* tendon force strain relationships for one subject estimated from method (A) 1-UIRF and (B) 2-PRF obtained on C_{max} in grey and $C_{1.5}$ in black. Experimental data correspond to the grey line and polynomial and Ogden model to the continuous and dashed lines, respectively. Note the similar fitting between polynomial and Ogden on (A) and (B).

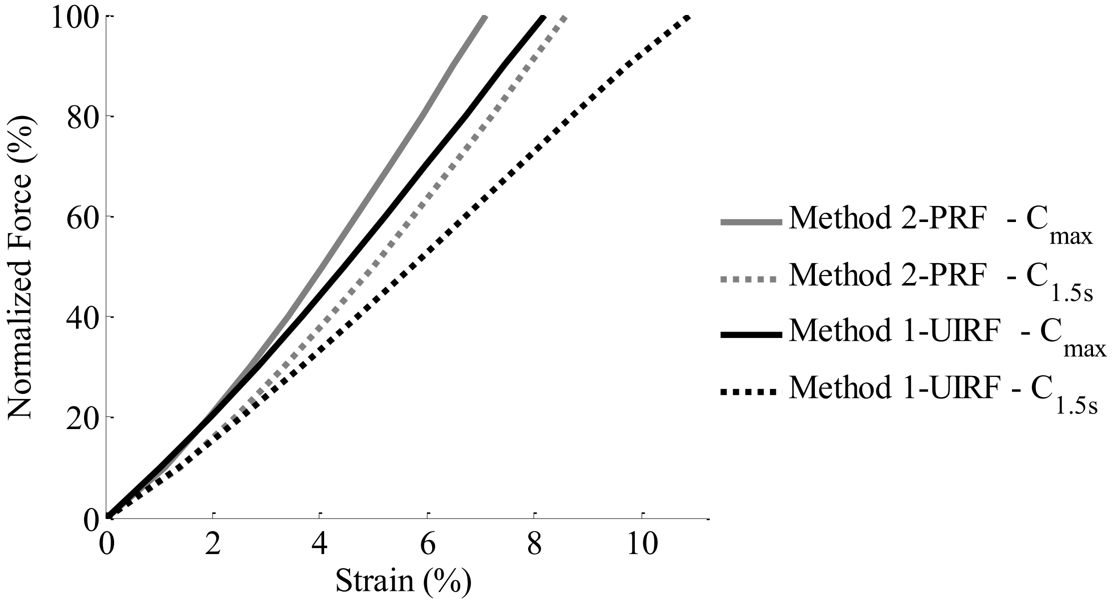

Fig. 3. *In vivo* tendon force-strain relationships averaged over eight subjects for C_{max} and $C_{1.5}$ for polynomial model according to method PRF and UIRF. Note the significant difference on the tendon strain estimates depending on the method used and the loading rate effect.

Table 1. Summary of experimental conditions

	C_{max}	$C_{1.5}$
Maximal moment (N.m)	139± 21	116± 24
Ankle plantarflexion rotation (°)	5.8 ± 2.4	4.3 ± 1.7
Ramp-up time duration (s)	0.58 ± 0.20	1.39 ± 0.10
Initial tendon length	208± 31	206± 31

Table 2. Maximal tendon elongation, maximal tendon strain, and fitting results according to method, model, and condition (mean ± SD).

	Method 1-UIRF				Method 2-PRF			
	Ogden		X^2		Ogden		X^2	
	C_{max}	$C_{1.5}$	C_{max}	$C_{1.5}$	C_{max}	$C_{1.5}$	C_{max}	$C_{1.5}$
Tendon elongation *† (mm)	16.4±3.6	20.7±5.4	16.4±3.6	21.9±4.9	14.6±2.5	17.0±3.6	14.4±2.3	17.4±3.7
Tendon strain *† (%)	8.2±3.1	10.4±3.9	8.2±3.1	10.9±3.4	7.2±1.9	8.4±2.5	7.1±1.8	8.6±2.4
RMS	3.3±2.1	2.4±0.9	2.8±1.1	2.3±0.8	5.2±1.8	3.5±2.2	4.4±1.3	3.0±1.2
R^2	0.99±0.02	0.98±0.01	0.99±0.01	0.99±0.01	0.97±0.02	0.97±0.04	0.98±0.01	0.98±0.01

*and † reveal significant difference between method 1 & 2 and condition C_{max} & $C_{1.5}$, respectively.