



Soleus muscle and Achilles tendon compressive stiffness is related to knee and ankle positioning

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ABSTRACT

Changes in fascicle length and tension of the soleus (SOL) muscle have been observed in humans using B-mode ultrasound to examine the knee from different angles. An alternative technique of assessing muscle and tendon stiffness is myometry, which is non-invasive, accessible, and easy to use. This study aimed to estimate the compressive stiffness of the distal SOL and Achilles tendon (AT) using myometry in various knee and ankle joint positions. Twenty-six healthy young males were recruited. The Myoton-PRO device was used to measure the compressive stiffness of the distal SOL and AT in the dominant leg. The knee was measured in two positions (90° of flexion and 0° of flexion) and the ankle joint in three positions (10° of dorsiflexion, neutral position, and 30° of plantar flexion) in random order. A three-way repeated-measures ANOVA test was performed. Significant interactions were found for structure × ankle position, structure × knee position, and structure × ankle position × knee position ($p < 0.05$). The AT and SOL showed significant increases in compressive stiffness with knee extension over knee flexion for all tested ankle positions ($p < 0.05$). Changes in stiffness relating to knee positioning were larger in the SOL than in the AT ($p < 0.05$). These results indicate that knee extension increases the compressive stiffness of the distal SOL and AT under various ankle joint positions, with a greater degree of change observed for the SOL. This study highlights the relevance of knee position in passive stiffness of the SOL and AT.

1. Introduction

The soleus (SOL) and gastrocnemius (GAST) are two of the most studied muscles due to their close anatomical and functional relationships during plantar flexion in walking, running, and balance tasks (Alizadehsaravi et al., 2020; Franz and Thelen, 2016; Knaus et al., 2020). The SOL is the largest muscle of the triceps surae; The muscular fibres of the SOL insert in a broad aponeurosis, lies immediately adjacent to the aponeurosis of the GAST on the anterior surface (Dalmau-Pastor

et al., 2014; Olewnik et al., 2020; Shan et al., 2019). Distally, SOL and GAST are connected to the calcaneus through the Achilles tendon (AT). The origin of the GAST and SOL are anatomically distinct, arising from above and below the knee respectively (Dalmau-Pastor et al., 2014; Olewnik et al., 2020; Shan et al., 2019). The AT comprises various layers (Ballal et al., 2014; Finni et al., 2017; Handsfield et al., 2017); the deep layer is composed of the tendon of the lateral GAST and SOL, while the superficial layer is composed of the medial GAST. The AT is the thickest and strongest tendon in the human body and demonstrates a high

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capacity for transmitting tensile forces between the uniarticular SOL and biarticular GAST muscles (Finni et al., 2017; Maas et al., 2020).

Knee and ankle joint positions have been used to study the mechanical properties and interactions of musculoskeletal tissues (e.g., muscle, tendon, fascia) in uni and biarticular plantar flexor muscles (Bojsen-Møller et al., 2010; Diong and Herbert, 2015; Le Sant et al., 2017; Tian et al., 2012; Yaman et al., 2013). In clinical practice, the passive knee flexo/extension with the ankle in dorsiflexion/plantar flexion position is usually used to differentiate gastrocnemius tightness from an achilles tendon contracture in clinical populations, for example, as witnessed in individuals with cerebral palsy using the Silfverskiöld Test (Barouk and Barouk, 2014; Singh, 2013). While the effects of knee joint position on the tension of AT and biarticular plantar flexor (i.e., GAST) are intuitive, the influence of the connective tissue on the mechanical interaction between the knee joint and uniarticular SOL muscle continues to be debated (Maas, 2019; Sandercock and Maas, 2009; Tijs et al., 2018). For instance, using tetanus stimulation, no or little intermuscular force transmission between SOL and GAST has been reported in cats (Sandercock and Maas, 2009). The transmission of forces between adjacent muscles is known as intermuscular myofascial force transmission (Huijing, 2007). Using B-mode ultrasound, a significant change in SOL and GAST muscle fascicle lengths was observed in humans when the knee angle was manipulated, suggesting an intermuscular force transmission between these muscles (Tian et al., 2012). Using shear wave elastography, it has been observed that knee extension increases the SOL stiffness distally (Ateş et al., 2018; Liu et al., 2020), but not proximally (Ateş, Andrade, 2018). This suggests that SOL stiffness might be site-dependent (Ateş et al., 2018).

An alternative non-invasive technique to assess muscle and tendon stiffness is myometry, which estimates the mechanical properties of tissues, such as the index of stiffness, by estimating the resistance of tissue to deformation in response to an applied force (Pruyn et al., 2016). Myometry involves use of a portable hand-held device, which is cheaper compared to B-mode ultrasound and shear wave elastography, and does not require the presence of a specialized technician. The myometry device exerts a single, quick mechanical impulse (time pulse 15 ms, force 0.4 N) over a constant precompression force (0.18 N) applied to the subcutaneous tissue layer above the muscle or tendon under assessment (Schneider et al., 2014). This technique has been reported to be reliable and valid for assessing superficial muscle and tendon stiffness in both clinical and laboratory settings (Pruyn et al., 2016; Schneebeli et al., 2020). Myometry has been shown to be sensitive in discriminating changes in biarticular GAST muscle and AT stiffness with different knee and ankle configurations (Huang et al., 2018). However, the measurement of stiffness in the SOL during different ankle and knee joint positions has not yet been conducted through the use of myometry. The available scientific evidence has shown that epimuscular myofascial forces could occur through the connective tissue network that runs parallel to the SOL fasciculus (Tijs et al., 2018), which suggests that myometry could be appropriate to detect changes on compressive stiffness of the SOL muscle during knee extension.

This study aimed to estimate the compressive stiffness of the distal SOL and AT using myometry in various knee and ankle joint positions. We hypothesized that knee extension increases the stiffness of the SOL and AT under various ankle joint positions. We also used a parasagittal cadaveric dissection of one leg to support our hypothesis from an anatomical perspective. Understanding the effect of passive knee extension on the compressive stiffness of SOL muscle using myometry could provide new insights into the anatomical and biomechanical interactions of the triceps surae muscle, which could then be transferred to clinical populations.

2. Methods

2.1. Participants

The inclusion criteria were as follows: 18–30 years old; moderate level of physical activity, as measured by the short version of the International Physical Activity Questionnaire (Silva-Batista et al., 2013); and a body mass index < 30. The exclusion criteria comprised: musculoskeletal injury in the six months before measurements; scoliosis; spinal surgeries; current degenerative or musculoskeletal disease affecting the ankle, knee, or hip; and any cardiovascular or respiratory disease, and performed strenuous exercise within 48 h of testing (Huang et al., 2018). For participant characteristics, see Table 1.

This study was approved by the Northern Metropolitan Health Service Ethical Committee. Before commencing the study, participants were informed about the purpose and content of the investigation. Each participant's signature provided informed consent.

2.2. Protocol

Each participant took part in one assessment session. Volunteers were instructed to refrain from physical activity that was more intense than their daily activities for 48 h prior to the session. Height, weight, physical activity level, and leg dominance of each participant were registered at the beginning. During the session, participants were asked to lie prone on a therapeutic bed. A dermatograph pencil was used to mark the two assessment areas (SOL and AT) (see below). Participants were then instructed to “stay as calm and relaxed as possible” and were also informed that their leg was going to be passively positioned in the respective position prior to any action. Knee positions (joint angles: 90° of flexion and 0° of flexion) and three ankle joint positions (joint angles: 10° of dorsiflexion, neutral position, and 30° of plantar flexion) were used in various combinations (e.g., 90° of knee flexion with 30° of ankle plantar flexion) while myometry measurements were taken for the target structures (SOL and AT). One researcher (KCM) positioned the ankle and knee configurations with the aid of a standard 8” Universal Goniometer with a sensitivity of 2 degrees (Prestige Medical, Northridge, California), while the second researcher (NAV) performed the stiffness assessments using the myometer (see below). All measurements were assessed on the dominant leg. All combinations of knee and ankle positions were selected in random order, with two measurements collected per configuration; the average of the two measurements was used for further analysis. All assessments were performed within approximately 45 min.

2.3. Equipment and Variables

2.3.1. Myometry

The Myoton-PRO device (Myoton SA, Tallinn, Estonia) was used to measure the passive stiffness at two points (distal SOL and AT, see below) of the dominant leg of each participant. The Myoton-PRO device provides a stiffness value (Newton/meter; N/m) that reflects the resistance of the soft tissue to deformation induced by an exterior, low force, quick-release mechanical impulse applied under constant pre-load. The

Table 1
Characteristics of participants.

Variables	(n = 26)
Age (years)	22 [21–25]
Height (cm)	174.5 ± 4.9
Weight (kg)	71.6 ± 8.2
IPAQ	Moderate (n = 23); high (n = 7)

Short version of the International Physical Activity Questionnaire (IPAQ). Mean ± standard deviation for normal distribution. Median [range] for no normal distribution.

standard 3 mm diameter probe was placed perpendicularly to the skin's surface, directly over the muscle or tendon to be analyzed (Fig. 1). The compressive stiffness value is calculated as the maximum acceleration of oscillation divided by the maximum displacement of the tissue; larger values of this index indicate stiffer tissues. A set of ten mechanical impulses were performed at 1 s intervals, and the mean value from each set of ten was used for analysis.

The SOL was measured distally from GAST, 2 cm medial to the midline of the leg at the end of the medial GAST (Fig. 1) (Schoenrock et al., 2018). As shown in Fig. 1, the AT was measured 4 cm above the calcaneal tuberosity (Liu et al., 2018). The values obtained for each measurement are considered to be valid with an error of less than 3 %, while an interreliability greater than 0.84 has been shown for AT and GAST measurements in various ankle positions (Chang et al., 2020; Schneebeli et al., 2020; Feng et al., 2018). The minimal detectable change in TA stiffness has been estimated at 45 Newton/meter (N/m) (Liu et al., 2018). The resting stiffnesses of the AT and GAST muscle belly, as measured by Myoton-PRO, have been reported to be related ($r = 0.54\text{--}0.67$) to Young's modulus values quantified by shear wave elastography, indicating that the Myoton-PRO can be used to assess the mechanical properties of the GAST muscle and AT under resting conditions (Feng et al., 2018; Lee et al., 2021). It should be noted that the compressive stiffness assessed by myometry may not be interpreted only as muscle stiffness as several tissues may be involved (see Fig. 2).

2.3.2. Cadaveric dissection

To illustrate the interface between the GAST and SOL, a cadaveric dissection was performed. A single supracondylar amputation of a deceased 60-year-old male was used (Anatomical Bank, Human Anatomy Laboratory, Faculty of Medicine, University of Chile, Santiago, Chile). Parasagittal anatomical slices were prepared to expose the connective tissue linkages between the GAST and SOL, as well as the sheath surrounding the neurovascular tract.

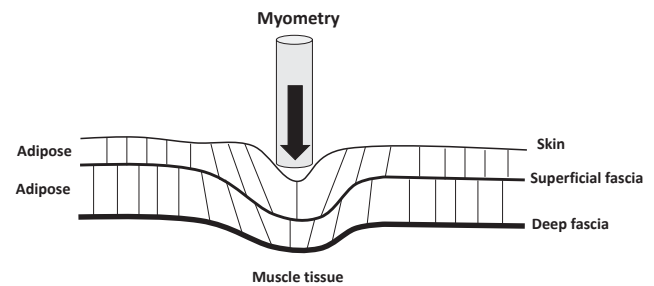


Fig. 2. Schematic sagittal view of compressive stiffness using the myometry. Note that several tissues may be involved during compressive stiffness assessment.

2.4. Statistical analysis

The sample size was calculated using G*Power v3.1.9.2 with an effect size of 0.67, alpha error of 0.05, and statistical power of 0.8. Thus, the minimum number of subjects required for the study was determined to be 21 (Huang et al., 2018).

Data recorded with the Myoton-PRO were analyzed with IBM SPSS statistics (v. 25). Normality was assessed via the Shapiro-Wilk test. A three-way repeated measurements ANOVA test considering the factors of structure (SOL and AT), ankle position (10° of dorsiflexion; neutral position; and 30° of plantar flexion), and knee position (90° of flexion and 0° of flexion) was performed. The Greenhouse-Geisser correction was used if the assumption of sphericity, as checked by Mauchly's test, was violated. The eta-squared (η^2) for ANOVA was used to examine the effect size. An η^2 value less than 0.06 was classified as "small", 0.07–0.14 as "moderate", and greater than 0.14 as "large". In addition, Cohen d for paired samples was used as an indicator of the effect size. A Cohen d value less than 0.2 was classified as "trivial", 0.2–0.5 as "small", 0.5–0.8 as "moderate", and greater than 0.8 as "large" (Cohen, 1992). For all tests, statistical significance was defined as $p < 0.05$.

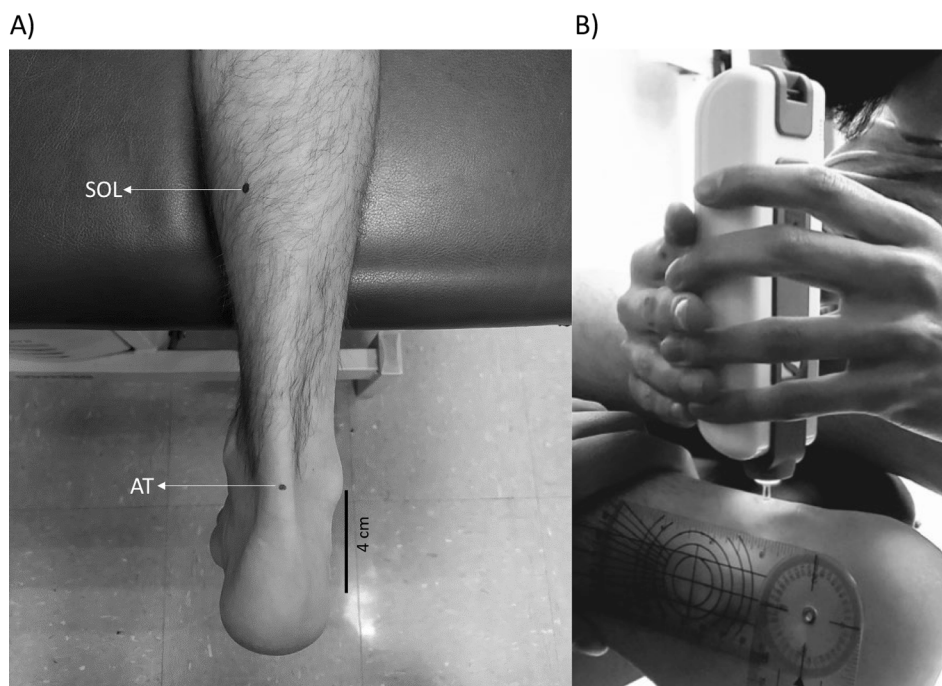


Fig. 1. (A) Sites where the soleus muscle and Achilles tendon were measured. The soleus was measured distally at the end of the medial gastrocnemius, 2 cm medial to the midline of the leg. The Achilles tendon was measured 4 cm above the calcaneal tuberosity. (B). Example of myometry assessment of the Achilles tendon.

3. Results

3.1. Compressive stiffness

Twenty-six males were recruited (Table 1). The recorded values of stiffness and their 95 % confident intervals of differences between knee positions are shown in Fig. 3, Fig. 4, and Table 2, respectively.

Significant interactions were found when considering structure × ankle position ($F_{(1.3, 33.3)} = 140.7$; $p < 0.001$; $\eta^2p = 0.85$) and structure × knee position ($F_{(1.0, 25.0)} = 20.2$; $p < 0.001$; $\eta^2p = 0.45$), as well as when considering all three factors structure × ankle position × knee position ($F_{(1.5, 38.6)} = 7.12$; $p = 0.005$; $\eta^2p = 0.22$).

The AT ($p < 0.05$, effect size > 0.6 , moderate to large) and SOL ($p < 0.001$, effect size > 3.0 , large) showed significant increases in stiffness with knee extension over knee flexion for all ankle positions tested (Table 2, Fig. 3 and Fig. 4). When the ankle was positioned in a neutral position or dorsiflexion, the mean changes in stiffness as the knee position changed were larger for the SOL compared to the AT (neutral position, p -value 0.008, $d = 0.93$; dorsiflexion position, $p < 0.001$, $d = 1.35$) (Fig. 5). Regarding the AT, the mean differences in stiffness as knee position varied were similar across all ankle positions (Fig. 5).

3.2. Anatomical dissection

Based on a qualitative description of a cadaveric sample with a parasagittal cut, it was possible to identify several structures that may be related to the observed results. The proximal portion of the SOL, partially covered by the GAST, shares their aponeurotic laminae (Fig. 6). The distal portion of SOL, where the myometry measurements were taken, is more superficial and accessible and closely linked with the aponeurotic lamina which is continuous with the AT (Fig. 6). The neurovascular tract directly connects i) distally with the GAST and SOL

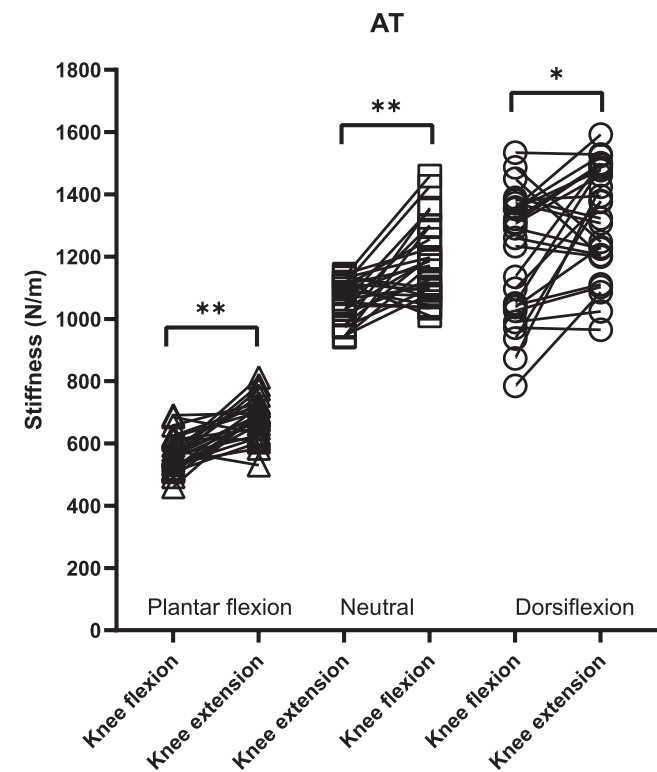


Fig. 3. Stiffness changes in the Achilles tendon measured in three ankle positions (30° of plantar flexion, [triangles]; neutral position [squares]; or 10° of dorsiflexion [circles]) and two knee positions (0° of flexion or 90° of flexion). * $p < 0.05$, ** $p < 0.001$.

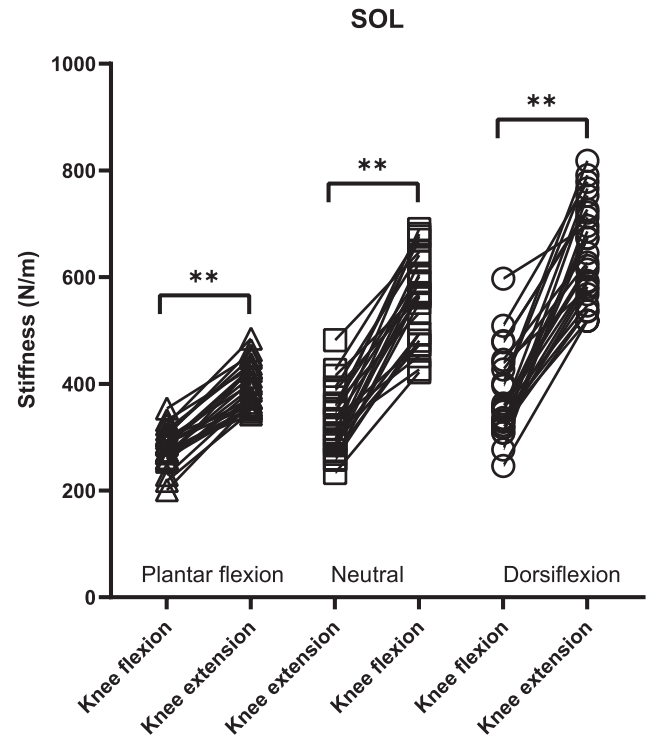


Fig. 4. Stiffness changes in the soleus muscle measured in three ankle positions (30° of plantar flexion [triangles]; neutral position [squares]; or 10° of dorsiflexion [circles]) and two knee positions (0° of flexion or 90° of flexion). * $p < 0.05$, ** $p < 0.001$.

epimysium and ii) proximally to the deep fascia of the hamstring muscles and femoral periosteum (Fig. 6).

4. Discussion

Through the myometry assessments, we found that: i) AT demonstrated higher compressive stiffness values than SOL for all measurements; ii) stiffness of AT and SOL were significantly higher when the knee was extended, independent of ankle positioning; iii) as knee position changed, changes in stiffness of the SOL were larger than those of the AT when the ankle was positioned in a neutral and dorsiflexed position. Overall, these results confirmed that knee extension increases the stiffness of both the distal SOL and AT in combination with various ankle joint positions. At the same time, the magnitude of these changes depends upon both the structure being assessed and the position of the ankle. These results show that myometry could potentially be used to complement orthopedic tests, such as the Silfverskiöld Test (Molund et al., 2018).

4.1. Effects of knee angle on the stiffness of the SOL and AT

This study has demonstrated that the compressive stiffness of both the distal SOL and AT are affected by changes in knee and ankle position. Our results align with recent studies using shear wave elastography and B-ultrasound (Ateş et al., 2018; Liu et al., 2020; Tian et al., 2012). However, our results should not be extrapolated to the entire SOL, due that intermuscular mechanical interactions of SOL are not homogeneous (Ateş et al., 2018). Our results can be explained by different factors. In our study, the stiffness of the SOL was measured in the distal region of the GAST due to its accessibility (Fig. 6). This region of SOL is near the musculotendinous junction, which is exposed to higher stresses resulting in increased stiffness (Liu et al., 2020). Other more proximal in-series structures, such as the hamstring muscle, fascia and neurovascular tract (see anatomical implication), may increase the length of GAST

Table 2
Differences of stiffness between knee positions (n = 26).

Structure	Ankle position	Mean difference between knee positions (N/m)	95% IC of differences (N/m)	p-value	Effect size
AT	Plantarflexion	115.9	[81.1 to 150.7]	<0.001	1.31 (large)
	Neutral	119.0	[61.2 to 176.8]	<0.001	0.74 (medium)
	Dorsiflexion	116.1	[34.8 to 197.4]	0.007	0.62 (medium)
SOL	Plantarflexion	116.2	[102.9 to 129.4]	<0.001	3.58 (large)
	Neutral	235.8	[206.2 to 265.3]	<0.001	3.29 (large)
	Dorsiflexion	274.7	[237.1 to 312.2]	<0.001	3.02 (large)

Achilles tendon, AT. Soleus muscle, SOL. 95 % Confidence Interval, 95 % CI.

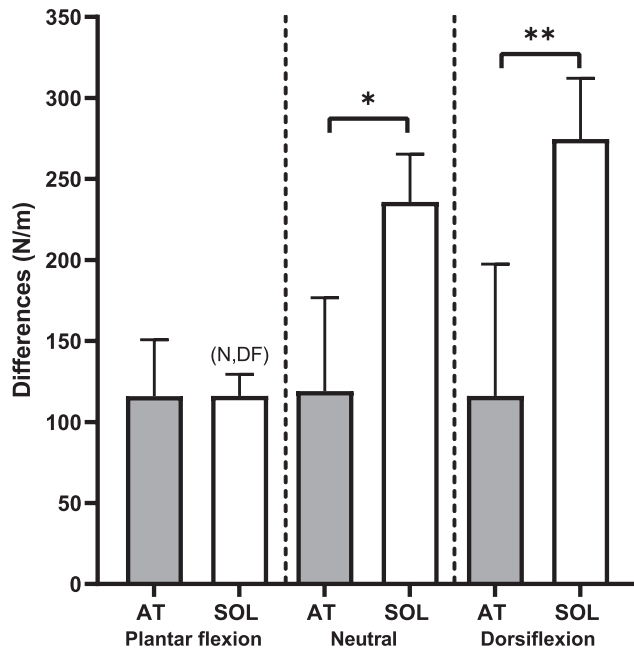


Fig. 5. Differences in stiffness in the Achilles tendon (AT) and soleus muscle (SOL) in the two knee positions (extended or flexed) for each position of the ankle. * $p < 0.05$, ** $p < 0.001$. A significant difference ($p < 0.05$) was identified between the stiffness in the SOL during plantar flexion with the ankle in the neutral (N) or dorsiflexed (DF) positions. Data are expressed as means and 95 % confidence interval.

proximally (Cruz-Montecinos et al., 2015; Huijing, 2009; Maas, 2019; Wilke et al., 2020; Wilke and Tenberg, 2020). Because GAST and SOL are connected to the AT, the GAST lengthening may increase the stiffness of AT transmitting passive tension to distal SOL.

At the AT, we observed that the changes in compressive stiffness relating to knee position were similar for all ankle positions (Fig. 4). Even though the SOL is a monoarticular ankle muscle, it demonstrated more significant variations in stiffness than the AT in relation to changes in knee position in neutral and dorsiflexed positions. There are two main explanations for these results. First, the distal SOL may be associated with higher mechanical compliance (or elastic deformation) compared to AT (Liu et al., 2020). In response to knee extension, the SOL may decrease their stiffness proximally but increase distally (Ateş, Andrade, 2018), where the aponeurosis of SOL is thicker than the proximal region (Shan et al., 2019). This may serve to dissipate the stiffness from the proximal to the distal sites. Second, the transmission of myofascial forces during distal joint rotation may occur, mainly via the connective tissue network without significant changes in fascicle length (Tijs et al., 2018). Considering the above, the stiffness value of distal SOL and AT assessed by myometry may reflect a complex mechanical behavior in response to knee positioning.

4.2. Anatomical considerations

Several anatomical structures may be involved in the force transmission between the knee and ankle joints (e.g., fascia, neurovascular tracts, muscle and tendon tissues). For instance, the crural fascia may play an important role in load transmission of the leg (Prilutsky et al., 2011; Stahl and Nichols, 2014). The crural fascia separates the leg muscles into three different compartments (anterior, posterior, and lateral). These compartments can transmit forces between them and potentially transfer forces to the ankle and knee (Benjamin, 2009; Stecco et al., 2009). In the posterior compartment, the crural fascia is attached along the entire area of the medial margin of the tibia. Force transmission directed proximally (cranially) may occur via the continuity between the SOL and the deep crural fascia, located posterior to the MG muscle belly at the level of the leg, which continues its ascent throughout the popliteal and posterior femoral region (Cruz-Montecinos et al., 2015; Wilke et al., 2020; Wilke and Tenberg, 2020). Note that distally, the crural fascia is attached to the transverse intermuscular septum and surrounding the AT (Edama et al., 2015). Similarly, the anterior and posterior (relative to the anterior/intramuscular aponeurosis of the SOL) muscle fibers extend throughout the muscle, accompanying the posterior aponeurosis (insertion lamina), which merges into the AT. The anterior (intramuscular) aponeurosis also merges with the AT to further transmit the force from the AT into the distal SOL (Finni et al., 2017; Franz and Thelen, 2016; Maas et al., 2020). Hence, knee extension not only may lengthen the GAST but also increase the tension of crural fascia, aponeurosis of distal SOL and AT.

The fascia is a continuous with the neurovascular tracts. As shown in Fig. 6, the neurovascular tract provides connections with adjacent and distant muscles (Huijing, 2003, 2009). Knee extension may also increase stiffness of the neurovascular tract (Huijing, 2003, 2009), changing the relative position of SOL, which would stretch the epimuscular connections increasing distal SOL stiffness (Ateş et al., 2018). At the same time, the distal portion of SOL may be affected by changes in ankle position (Ateş et al., 2018; Liu et al., 2020). The above described support our hypothesis from an anatomical perspective.

4.3. Methodological considerations

Compared to shear wave elastography, the myometry has shown a moderate to good correlation in GAST muscle stiffness at rest and under muscle contraction (Lee et al., 2021). Comparing to US and shear wave elastography, the passive knee extension alongside myometric assessment of SOL and AT can be used in clinical context since it is simple and easy to apply. However, the compressive stiffness assessed by myometry may be affected by several anatomical and physiological factors, such as subcutaneous fat tissue; joint position; resting condition of muscles; type of muscles assessed (i.e., deep, architecture, volume, type of fiber); gender; hormonal status; body position (i.e., gravity); and aging (Agyapong-Badu et al., 2016; Huang et al., 2018; Lee et al., 2013; Schneider et al., 2014; Viir et al., 2006). The subcutaneous adipose tissue can affect the interpretation of compressive stiffness, since it can act as a low-pass filter by attenuating the mechanical impulse response (Lee et al., 2021). All these factors should be controlled in research and clinical settings. In our case, the SOL was measured in a supine position,



Fig. 6. (A) Soleus (SOL) in the parasagittal plane; unfilled arrowhead: SOL. A white asterisk indicates a reference zone where the myometry measurements were taken. (B) Neurovascular tract in the popliteal region (parasagittal slice). Unfilled arrowhead: hamstring; white arrowhead: medial gastrocnemius muscle; and white arrow: neurovascular tract. This image shows how the neurovascular sheath directly connects i) distally with the GAST and SOL epimysium and ii) proximally to the deep fascia of the hamstring muscles and femoral periosteum.

distally at the end of the medial GAST, where the SOL muscle is more accessible, and the subcutaneous fat tissue was expected to have little or no effect on the compressive stiffness (Fig. 6).

4.4. Limitations

Although myometry is a sensible and reliable technique for assessing muscle stiffness, care should be taken when interpreting results for the SOL (Lee et al., 2021). Several factors might be involved in the compressive stiffness value. For instance, since measurements of the SOL (distal portion) were taken directly over the skin, the subcutaneous fat and aponeurosis tissue may have been involved in the interaction observed. Further, our results were limited to only the distal portion of the SOL due to the limitation of myometry. Furthermore, muscle activity with electromyography was not assessed during combinations of knee and ankle positions; thus, some participants might not have reached a resting state, potentially affecting the measurements. In addition, our study only considered the mechanical aspect of the stiffness. The muscle and tendon stiffness may also be regulated by the nervous system. Finally, this study included only men to control the potential hormonal effect on connective tissue; future research should consider comparative studies between genders.

In terms of applications, the present results may help transfer this type of measurement (myometry combined with passive knee extension) to the clinical population with plantar flexion contractures (e.g., cerebral palsy, plantar fasciitis). Future studies are needed in the clinical population with plantar flexion contractures to understand the

biomechanical and functional implications of these results.

5. Conclusion

These results indicate that knee extension increases the compressive stiffness of the distal SOL and AT under various ankle joint positions, with a greater degree of change observed for the SOL. This study highlights the relevance of knee position in passive stiffness of the SOL and AT.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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