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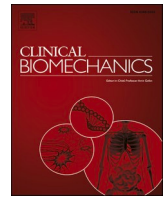
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Medial gastrocnemius muscle fascicle function during heel-rise after non-operative repair of Achilles tendon rupture

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ABSTRACT

Background: To better understand muscle remodelling in dynamic conditions after an Achilles tendon rupture, this study examined the length of medial gastrocnemius muscle fascicles during a heel-rise at 6- and 12-months after non-operative ATR treatment.

Methods: Participants (15 M, 3F) were diagnosed with acute Achilles tendon rupture. Medial gastrocnemius subtendon length, fascicle length and pennation angle were assessed in resting conditions, and fascicle shortening during bi- and unilateral heel-rises.

Findings: Fascicle shortening was smaller on the injured side (mean difference [95% CI]: -9.7 mm [-14.7 to -4.7 mm]; -11.1 mm [-16.5 to -5.8 mm]) and increased from 6- to 12 months (4.5 mm [2.8–6.3 mm]; 3.2 mm [1.4–4.9 mm]) in bi- and unilateral heel-rise, respectively. The injured tendon was longer compared to contralateral limb (2.16 cm [0.54–3.79 cm]) and the length decreased over time (-0.78 cm [-1.28 to -0.29 cm]). Tendon length correlated with fascicle shortening in bilateral ($r = -0.671$, $p = 0.002$; $r = -0.666$, $p = 0.003$) and unilateral ($r = -0.773$, $p \leq 0.001$; $r = -0.616$, $p = 0.006$) heel-rise, at 6- and 12-months, respectively. In the injured limb, the change over time in fascicle shortening correlated with change in subtendon length in unilateral heel-rise ($r = 0.544$, $p = 0.02$).

Interpretation: This study showed that the lengths of the injured tendon and associated muscle can adapt throughout the first year after rupture when patients continue physiotherapy and physical exercises. For muscle, measures of resting length may not be very informative about adaptations, which manifest themselves during functional tasks such as unilateral heel-rise.

1. Introduction

The Achilles tendon (AT) is the most frequently ruptured tendon in the body and incidence is rising (Holm et al., 2015; Lantto et al., 2015). An Achilles tendon rupture (ATR) can result in long term deficits, characterized by tendon elongation, plantar flexor atrophy and decreased heel-rise height (Agres et al., 2020; Hullfish et al., 2019a; Silbernagel et al., 2012; Stäudle et al., 2022), with the latter being a clinically important factor in ATR treatment and an important marker of patient recovery (Baxter et al., 2019; Olsson et al., 2014).

The AT is a complex structure that consists of the three triceps surae muscle subtendons, namely the soleus, lateral gastrocnemius and medial gastrocnemius (MG) (Edama et al., 2015; Pękala et al., 2017), of which

the MG is most often examined. Tendon elongation and changes in morphological muscle properties, such as pennation angle and fascicle length, are important parameters for patient function (Baxter et al., 2018; Peng et al., 2017; Svensson et al., 2019). Alterations in muscle-tendon properties appear to be linked (Hullfish et al., 2019b) and related to the maximum height achieved during a heel-rise test (Baxter et al., 2019; Silbernagel et al., 2012). The lack of tension caused by the extra tendon slack is thought to stimulate muscle remodelling (Hullfish et al., 2019b). Computer simulations showed that increased tendon length and decreased MG muscle fascicle length are negatively correlated with single leg heel-rise height (Baxter et al., 2019). Moreover, decreased muscle fascicle length has been associated with decreased peak plantar flexor torque (Hullfish et al., 2019b). The excursion

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capacity of fascicles is an important factor in force production and muscle tendon unit (MTU) function (Peng et al., 2017). Therefore, long term plantar flexion force deficits after ATR might be explained by morphological changes in MG (Baxter et al., 2018; Hullfish et al., 2019a), such as decreased fascicle length (Hullfish et al., 2019b; Nicholson et al., 2020; Peng et al., 2017; Svensson et al., 2019), that occur during the initial immobilization phase. Focusing on the interplay between tendon and gastrocnemius muscle remodelling is thus important (Hullfish et al., 2019b).

To date, research is limited, and it is not yet known how MG muscle remodelling manifests itself in dynamic conditions, such as during a heel-rise, after non-operative ATR repair. Therefore, to better understand the functional recovery after ATR, the aim of this study was to examine the length of the medial gastrocnemius fascicles during a heel-rise at 6- and 12-months follow up in non-operatively treated patients. Further, the objective was also to examine the association between MG fascicle and MG subtendon lengths.

It was hypothesized that after non-operative ATR repair, the MG muscle in the injured limb would have a smaller capacity to shorten than the uninjured side in agreement with changes found in passive conditions (Hullfish et al., 2019b; Peng et al., 2017; Svensson et al., 2019). Moreover, an increase of both the resting length and fascicle shortening during heel rise in the course of rehabilitation was expected with the inter-limb difference becoming smaller over time. Furthermore, it was expected that tendon and fascicle length would be associated.

2. Methods

2.1. Study design and participants

This study is part of a larger clinical cohort study “non-operative treatment of Achilles tendon Rupture in Central Finland: a prospective cohort study – NoARC” (trail registration: NCT03704532). The study was approved by the research ethics committee of the Central Finland Health Care District (2 U/2018) and all participants gave written informed consent.

Participants were recruited through the Central Hospital of Central Finland. Recruitment procedures have been previously described (Khair et al., 2022b). Patients were included if diagnosed by a medical doctor within 14 days of the injury, with 2 out of 4 positive tests based on American Academy of Orthopaedic Surgeons (AAOS) criteria (Kou, 2010) and excluded if there was a re-rupture. After recruitment, participants were asked to come to the biomechanics lab at the Faculty of Sport and Health Sciences, University of Jyväskylä (Jyväskylä, Finland) at both 6- and 12-months post-rupture. Data was collected between July 2018 and December 2021.

2.2. Treatment protocol

All participants were treated non-operatively in combination with an early mobilization rehabilitation protocol. After diagnosis, the foot was placed in a full equinus cast, which was replaced, after 2 weeks, by an open cast with the ankle in 20°-degree plantarflexion and 1 cm heel wedge, allowing walking and toe movement. At week 4 post-rupture, walking and full weightbearing were, if possible, encouraged and a custom-made functional cast orthosis with a 1 cm thick heel wedge was provided. After 8 weeks of treatment, the orthosis was removed, and a physiotherapist prescribed a progressive rehabilitation program. Swimming, walking, cycling, and underwater running were recommended. The first goal of training was to regain normal pain free walking. Slow running at 6 months and maximum plantarflexion at 8 to 9 months were recommended, if muscle strength was close to the strength of the contralateral limb (Khair et al., 2022a; Reito et al., 2018). Participants were asked about compliance to the physiotherapy program and return to physical activities during the laboratory visit.

2.3. Muscle and tendon resting lengths

MG fascicle length, defined as resting fascicle length, and pennation angle were measured using an Aloka Alpha-10 ultrasonography system (Aloka, Tokyo, Japan) with a 6-cm linear probe (UST-5712; 7.5 MHz). Participants laid in a prone position with the foot hanging freely at the end of the plinth so that the ankle joint was at a natural resting angle. Two images were taken at 50% of muscle belly length at a mediolateral location where fascicles could be best visualized. The best quality image was chosen for analysis of fascicle length and pennation angle. Images were analysed with ImageJ software version 1.50e (U. S. National Institutes of Health, Bethesda, Maryland, USA) and the average of two measurements was calculated for further analysis. Fascicle endpoints were manually selected and pennation angle was analysed as the angle between fascicle and lower aponeurosis. The intra-rater reliability, based on a subsection of 11 of the images, had an intraclass correlation (ICC_{2,1}) of 0.82 (90% CI 0.82–0.97) and 0.86 (90% CI 0.71–0.94) for fascicle length and pennation angle, respectively. The inter-rater reliability for fascicle length and pennation angle was found to be 0.80 (90% CI 0.57–0.91) and 0.74 (90% CI 0.43–0.90), respectively.

MG subtendon length was acquired in the same prone position as described above. A 3.6-cm linear probe (UST-5411; 7.5 MHz) was used to find the distal end of the MG-MTJ and the proximal end of the calcaneus. Both locations were marked with a pen and the distance between the two points was measured with a measuring tape (Khair et al., 2022a). This method has been shown to have an ICC of 0.99 (90% CI 0.97–0.99) (Khair et al., 2022b).

2.4. Fascicle length during heel-rise

Participants performed single heel-rises bilaterally and unilaterally at both the 6- and 12- months measurements. A 6-cm linear probe (96-element –7 MHz – EchoBlaster 128; Telemed, Vilnius, Lithuania or LV8-5 N60-A2; 5–8 MHz – ARTUS EXT-1H, Telemed, UAB) was placed on the mid-belly of the MG on the injured side, to have a representative measure of the average fascicle length change during heel-rise (Lichtwark et al., 2007). After the probe placement was secured, the participant was asked to perform a bilateral heel-rise, from the ground, as high as possible, slowly at their own pace. Minor chair support with two fingers was allowed to help maintain balance. The participant was given some time to rest before being asked to perform a single, unilateral heel-rise, starting with the injured limb. The same protocol was repeated for the un-injured limb. The task was repeated twice and the greatest amount of shortening or the video with the best image quality was taken for further analysis. Fascicle lengths during the tasks were tracked using a previously validated (Cronin et al., 2011) semi-automated approach in Matlab 2020b (The MathWorks Inc., USA). Fascicle shortening was calculated as the difference between the standing fascicle length and the length when standing on toes. The standing fascicle length was defined as the average length of 1 s before initiation of the contraction. If contraction started within the 1st second, the averaging window was shortened based on visual determination of the start of contraction. When standing on toes, the average length of the medial gastrocnemius fascicle was calculated within ± 0.5 s of the shortest length (Fig. 1). The ICC_{2,1} for intra-rater reliability (Hopkins, 2000) was found to be 0.97 (90% CI: 0.93–0.99).

2.5. Statistical analysis

IBM SPSS statistics 26 (IBM Corp., Armonk, NY) was used for statistical analysis. Two-way repeated measures ANOVA was used to check the interaction and main effects for the limb condition (injured and uninjured) and time points (6 and 12 months). When there was no interaction, a paired sample *t*-test was performed for the main differences between limbs and over time. A Pearson's correlation coefficient was calculated between the absolute values of MG subtendon length and

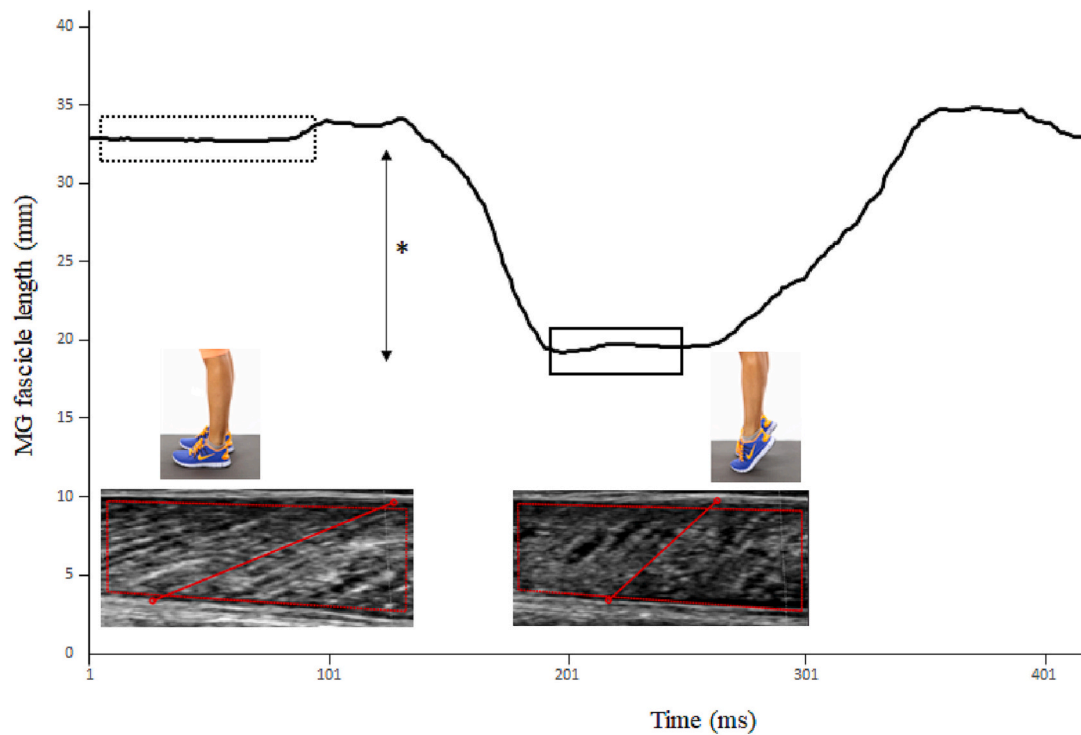


Fig. 1. Example of raw data of medial gastrocnemius muscle fascicle length changes during heel-rise test. Data was analysed from the ultrasound images shown in bottom. Shortening length (*) was calculated as fascicle length when on toes (solid box) subtracted from length during standing quietly (dashed box). When participant was standing quietly, the length was taken as average of the dotted box and when standing with toes.

shortening length, and between subtendon length and resting fascicle length. A correlation between the resting fascicle length and the standing fascicle length was calculated. Furthermore, correlations for the length changes over time were calculated between shortening length and MG subtendon length, and MG subtendon length and resting fascicle length. Correlations were interpreted as weak (0.1), moderate (0.4) or strong (0.7) (Schober et al., 2018). Level of significance for all tests was set at $\alpha = 0.05$.

3. Results

3.1. Participants

In total, 22 participants met initial eligibility criteria, 4 were excluded because of surgical treatment ($n = 2$) or because of a previous contralateral ATR ($n = 2$). Data from 18 (15 M, 3F) participants were

successfully collected at 6 (± 1.4) and 12 (± 1.0) months and were included in the analysis. At 6-months, fourteen participants (77.7%) had continued instructed physiotherapy and 15 (83.3%) reported participation in physical activities. At 12-months, five participants (27.7%) reportedly had continued physiotherapy exercises while seventeen (94.4%) were physically active in strength training, endurance, or team sports. Participant characteristics were as follows (means [SD]): age, 40.6 [11.6] years; height, 175.4 [7.2] cm; and body mass, 80.2 [12.3] kg. The means (SD) for resting fascicle length, pennation angle, MG subtendon length, standing fascicle length and shortening length are shown in Table 1.

3.2. Resting fascicle length and pennation angle

For resting fascicle length, a main effect for limb condition ($F = 19.607, p < 0.001$) was observed, with the injured limb displaying a

Table 1
Descriptives of the main outcome variables.

	limb condition	time		Difference	95% C.I.	P-value
		6 months	12 months			
Resting fascicle length (mm)	uninjured	49.26 (± 8.09)	47.96 (± 6.85)	-1.29 (± 4.01)	-3.29 - 0.71	0.190
	injured	39.28 (± 6.79)	38.31 (± 6.39)	-0.96 (± 5.39)	-3.64 - 1.72	0.459
Resting pennation angle (deg)	uninjured	23.51 (± 4.06)	24.45 (± 4.51)	0.94 (± 2.71)	-0.41 - 2.28	0.161
	injured	25.67 (± 4.51)	28.04 (± 5.82)	2.37 (± 4.12)	0.03 - 4.42	0.026
MG subtendon length (cm)	uninjured	18.18 (± 2.3)	17.49 (± 2.27)	-0.69 (± 1.16)	-1.26 - -0.11	0.022
	injured	20.43 (± 2.92)	19.56 (± 2.45)	-0.87 (± 1.72)	-1.73 - -0.02	0.046
Unilateral standing fascicle length (mm)	uninjured	53.56 (± 11.92)	54.13 (± 12.20)	0.58 (± 12.10)	-5.45 - 6.60	0.843
	injured	41.52 (± 13.09)	40.73 (± 14.42)	-0.78 (± 10.01)	-5.76 - 4.19	0.744
Bilateral standing fascicle length (mm)	uninjured	51.76 (± 12.60)	54.90 (± 11.80)	3.14 (± 9.33)	-1.50 - 7.78	0.171
	injured	41.43 (± 12.50)	43.62 (± 10.47)	2.19 (± 8.75)	-2.16 - 6.54	0.303
Shortening unilateral heel-rise (mm)	uninjured	22.62 (± 7.02)	26.17 (± 8.17)	3.54 (± 6.19)	0.46 - 6.62	0.027
	injured	11.85 (± 8.56)	14.66 (± 9.60)	2.81 (± 4.27)	0.69 - 4.94	0.012
Shortening bilateral heel-rise (mm)	uninjured	22.73 (± 7.04)	27.91 (± 6.97)	5.18 (± 6.04)	2.17 - 8.18	0.002
	injured	13.71 (± 7.72)	17.60 (± 9.26)	3.88 (± 4.03)	1.88 - 5.89	0.001

Differences are mean values (\pm SD). * = significant result ($\alpha = 0.05$) tested with a paired sample t-test. C.I. = confidence interval.

shorter resting fascicle length in the prone position (mean difference [95% CI]: -9.8 mm [-14.3 to -5.3 mm]). There was no main effect of time or limb x time interaction.

Concerning the resting pennation angle, we did not observe main effects of limb condition or limb x time interaction. However, main effect of time was found ($F = 8.101$, $p = 0.007$) with pennation angle increasing over time (mean difference [95% CI]: 1.7 deg. [0.5 – 2.8 deg.]).

3.3. MG subtendon length

MG subtendon length had a significant main effect of time ($F = 10.228$, $P = 0.003$) and limb condition ($F = 7.327$, $P = 0.011$) but no interaction. The injured limb showed a longer MG subtendon compared to the contralateral limb (mean difference [95% CI]: 2.16 cm [0.54 – 3.79 cm]). Over time, there was a decrease in the subtendon length (mean difference [95% CI]: -0.781 cm [-1.28 to -0.29 cm]).

3.4. Standing fascicle length and shortening length

For bilateral standing fascicle length there were no significant effect of time or limb x time interaction but main effects of limb condition were found for both unilateral ($F = 10.645$, $P = 0.003$) and bilateral ($F = 8.718$, $P = 0.006$) standing fascicle lengths. The injured limb displayed a shorter standing fascicle length for uni- and bilateral heel-rise (mean difference [95% CI]: -12.7 mm [-20.6 to -4.8 mm] and -10.8 mm [-18.2 to -3.4 mm], respectively).

During bilateral heel-rise, there was no statistically significant time-limb interaction for fascicle shortening, but main effects of limb condition ($F = 15.49$, $P < 0.001$) and time ($F = 28.03$, $P < 0.001$) were found. MG fascicle shortening was significantly smaller on the injured side compared to the contralateral limb (Mean difference [95% CI]: -9.7 mm [-14.7 to -4.7 mm]). An increase in MG fascicle shortening was observed over time (Mean difference [95% CI]: 4.5 mm [2.8 – 6.3 mm]) (Fig. 2a). There was no statistically significant time-limb interaction for unilateral heel-rise. Main effects for limb condition ($F = 17.65$, $P = 0.001$) and time ($F = 12.87$, $P < 0.001$) were found. Mean differences [95% CI] were, respectively, -11.1 mm [-16.5 to -5.8 mm] and 3.2

mm [1.4 – 4.9 mm] (Fig. 2b).

3.5. Correlations

Resting fascicle length and changes in this parameter were not significantly correlated with MG subtendon length or its change for either the uninjured or injured limb. Resting fascicle length was significantly (moderately) correlated with bilateral and unilateral standing fascicle lengths at both 6 ($r = 0.502$, $P = 0.002$; $r = 0.583$, $P < 0.001$) and 12 months ($r = 0.510$, $P = 0.001$; $r = 0.506$, $P = 0.002$), respectively.

MG subtendon length and MG fascicle shortening were negatively correlated in both bilateral ($r = -0.671$, $P = 0.002$) and unilateral ($r = -0.773$, $P \leq 0.001$) heel-rise at 6 months and 12 months ($r = -0.666$, $P = 0.003$; $r = -0.616$, $P = 0.006$; bilateral and unilateral, respectively) on the injured side. No significant correlations were observed for the contralateral limb.

For unilateral but not bilateral heel-rise, a significant positive correlation was observed between the change in fascicle shortening and the change in MG subtendon length over time on the injured side ($r = 0.544$, $P = 0.02$).

4. Discussion

This study investigated inter-limb differences and the effect of time on MG fascicle length changes during a heel-rise after non-operative ATR repair. We found that MG fascicle shortening during heel-rise increased from 6 to 12 months after ATR, reflecting improved capacity to work. However, the between-limb differences remained, the injured limb having persistently shorter resting MG fascicle length and longer MG subtendon length. Moreover, we observed correlations between MG fascicle shortening and tendon length reflecting the functional interaction between muscle and tendon.

The injured limb displayed on average a 10 mm smaller MG fascicle shortening during both uni- and bilateral heel-rise, compared to the contralateral limb. Since the MTU length remains the same, this could possibly be the result of shorter MG fascicles and increased tendon

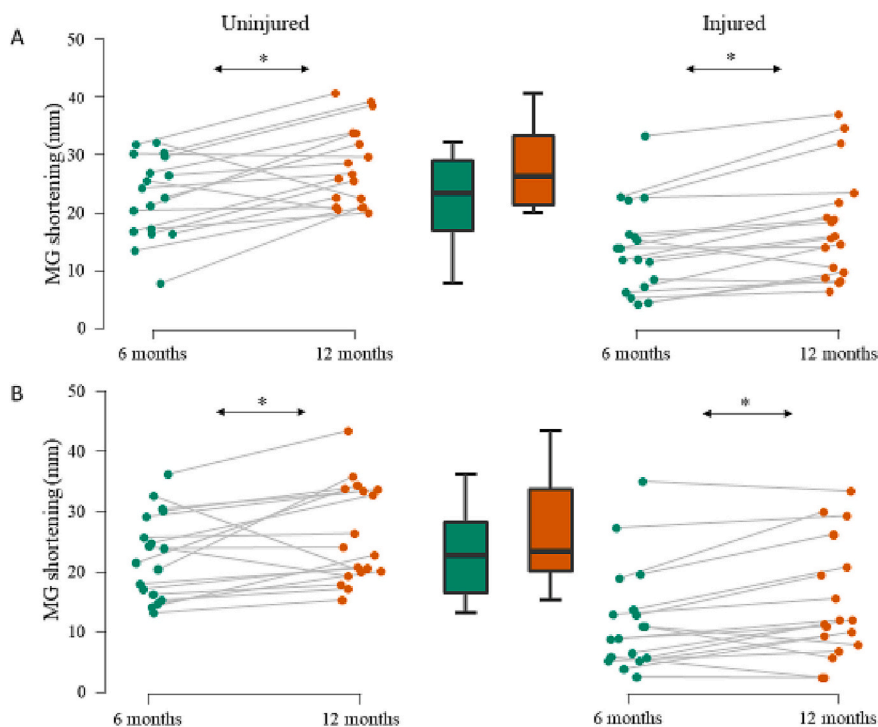


Fig. 2. Medial gastrocnemius (MG) muscle fascicle shortening of each individual during bilateral (A) and unilateral (B) heel-rise from uninjured (left) and injured limb (right) at 6 months (green) and 12-months (orange) time points. For both tasks, there were significant main effects of time and limb condition. * = significant difference over time, tested with paired sample *t*-test. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

length at the injured side. Indeed, we found a shorter resting fascicle length in the injured limb, as observed in previous studies (Agres et al., 2020; Baxter et al., 2018; Hullfish et al., 2019a; Peng et al., 2017), which could be the result of increased tendon length (Svensson et al., 2019). We also observed MG subtendon elongation, whereby the tendon was on average 2 cm longer 1-year post-rupture. This is in accordance with previous literature showing that AT resting length is longer up to 14 years post-surgery (Agres et al., 2015; Heikkinen et al., 2017) and is associated with functional deficits during dynamic conditions, such as walking and heel-rise performance (Khair et al., 2022a; Silbernagel et al., 2012).

During the rehabilitation process from 6 to 12 months, we observed a significant increase in MG fascicle shortening in both uni- and bilateral heel-rise. To the best of our knowledge, this has not been reported previously. Interestingly, the increase over time in fascicle shortening was also found on the healthy side, which possibly could be the result of the reduction in fascicle length post-rupture because of the decreased overall activity during the early phases of rehabilitation. Nevertheless, the difference between limbs persisted and, although fascicle shortening increased, deficits on the injured side remained. It was expected that the increase in fascicle shortening would be the result of an increase in resting fascicle length. However, no significant changes in resting fascicle length between 6- and 12-months was observed. An earlier study by Agres et al. (2020) reported no differences in fascicle length between 8 and 16 weeks after surgical repair of ATR, and together with present findings calls for novel rehabilitative approaches to induce positive muscle fascicle adaptations.

Thus, although fascicle shortening seems to increase over time, the same does not apply for the resting fascicle length. This suggests that some other mechanism is responsible for the functional improvement during rehabilitation. It is known that skeletal muscle, although sarcomeres have a preferred range, is extremely plastic (Lieber et al., 2017). It is possible that the fascicles and sarcomeres adapt to their new length during rehabilitation, resulting in a greater excursion and shortening capacity. Another possible explanation may be the shorter and stiffer tendon (Stäudle et al., 2021, 2022), which gives the fascicles more room to operate. Indeed, we observed a decrease in MG subtendon length over time. Similarly, Kangas et al. (2007) showed that after initial tendon elongation, there is a slight decrease in tendon length 6 weeks post-operation, regardless of rehabilitation protocol. Thus, there seems to be an interplay between AT elongation and MG fascicle length.

Indeed, fascicle shortening was negatively correlated with MG subtendon length, suggesting a longer subtendon in combination with a smaller fascicle shortening. A possible explanation for this interplay could be the loss of sarcomeres in series (Baker and Matsumoto, 1988; Williams and Goldspink, 1978). Non-operative ATR treatment starts with immobilization of the affected limb in a shortened position (Khair et al., 2022a; Reito et al., 2018). Animal studies have shown that this could decrease the amount of sarcomeres in series (Tabary et al., 1972; Williams and Goldspink, 1978), which can lead to a shorter fascicle length. Due to this functional length change of the muscle, the sarcomeres are shifted to a suboptimal position where force production is hindered (Williams and Goldspink, 1978). To adjust to this change and restore their optimal range, it is assumed that sarcomeres are removed (Baker and Matsumoto, 1988; Hinks et al., 2022; Williams, 1990; Zöllner et al., 2015), thereby changing the working range of the sarcomeres in the force-length relationship (Agres et al., 2020) and limiting the capacity to shorten. Another mechanism that seems to help compensate for the opposing effects after ATR repair is a change in tendon stiffness (Hullfish et al., 2019a; Stäudle et al., 2021, 2022). Stäudle et al. (2021) showed with computer simulations that a stiffer MG tendon makes the slope of the muscle force-length relationship steeper, bringing it back towards control levels, but as a result the muscle cannot produce force at more plantar-flexed joint angles, leaving the force production capacity compromised (Khair et al., 2022a).

For the unilateral heel-rise, a positive correlation was found between

the change in fascicle shortening and change in MG subtendon length over the 6-months follow-up. However, this correlation must be interpreted with caution, since the association relied on limited number of individuals with large changes in tendon length.

Lastly, in our study pennation angle did not differ between limbs, which is in contrast with previous literature (Hullfish et al., 2019a). However, the study by Hullfish et al. (2019b) only looked at the short-term effects (up to 4-weeks post-injury). In the long-term, it is thought that pennation reverts back to values comparable to the contralateral limb (Peng et al., 2017), which would explain why no differences were found in our study.

This study also had limitations. Firstly, heel-rise height and speed were not controlled for. It is possible that the increase in fascicle shortening allowed participants to reach a higher heel-rise height and thus improved function. Measuring this could have given a better picture of the effects on functional outcomes. Second, the two-dimensional ultrasound image cannot cover out of plane movements that can occur during muscle contractions thereby potentially causing errors when assessing the amount of fascicle shortening. Furthermore, although participants were asked during the lab protocol about physical activity, little is known about the type of physical therapy exercises performed. Although beyond the scope of this article, this information could have led to better understanding of exactly which exercises lead to the observed improvements.

5. Conclusion

The longer MG subtendon length and smaller MG muscle fascicle shortening during heel-rise persisted in injured compared to uninjured limb throughout the year after Achilles tendon rupture. However, the fascicle shortening increased during the rehabilitation period from 6 to 12 months in both the uninjured and injured limbs. Concurrently, MG subtendon lengths decreased reflecting adaptability of the tendon. The study showed that resting fascicle length may not change although the shortening capacity of muscle can increase when recovering from Achilles tendon rupture.

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Author contributions

Conception TF, NJC; Acquisition KvD, RMK, MS, TF; Analysis KvD, RMK, MS; Interpretation KvD, RMK, MS, TF; Drafting KvD; Revising KvD, RMK, MS, NJC, TF; Approval and accountability KvD, RMK, MS, NJC, TF.

Declaration of Competing Interest

None of the authors have conflicts of interest.

Data availability

Data used in this manuscript is available for viewing with reasonable request from the corresponding author.

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