Fully automated algorithm estimates muscle fascicle length from ultrasound image

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Abstract

While ultrasound is a useful tool for visualizing muscle *in vivo*, traditional analysis involves substantial manual labor. Semi-automated algorithms have been introduced in recent years, reducing the amount of time required for extracting pennation angles and fascicle lengths from ultrasound images. Unfortunately, semi-automated algorithms still require some user actions and thereby subjective decision making. We here present a freely available, fully automated feature detection algorithm that involves Hessian filtering to highlight line-like objects within the ultrasound image. Hough transform is used to determine muscle fascicle angles and feature detection is used to determine the location and angle of aponeuroses. As a demonstration, we test the algorithm on ultrasound images obtained from vastus lateralis muscle in healthy individuals (N = 9) during isometric knee extension moment production (0 – 45 N-m) at three knee angles (15-25 deg). Pennation angle, muscle thickness and fascicle length vary with knee moment and knee angle in line with previous observations. Specifically, fascicle length decreases with larger knee moments and increases towards knee flexion. We expect the proposed algorithm to be useful for estimating muscle fascicle lengths during cyclic movements like human locomotion.

Introduction

Animal movement occurs primarily through rotations of bony segments about joints, causing length changes in muscle-tendon complexes. When such movement occurs passively, series elastic tissue length is constant such that all length change is taken up by muscle fascicles. When movement requires muscle force, series elastic tissue length varies as a function of force. Consequently, series elastic tissue can change length even when joint angle is constant, and the muscle-tendon complex is isometric. The same is true for muscle fascicle, since any series elastic length change will generally result in fascicle length change in the opposite direction when muscle-tendon complex is isometric. The effects of joint rotation and muscle force on fascicle length can be captured using ultrasonography.

In ultrasonography, the echoes of ultrasonic waves (1-18 MHz) are used to reconstruct a cross-sectional image of bodily tissues. An ultrasound probe sends and receives ultrasonic waves, which are reflected anywhere where there are acoustic impedance changes. If the reflection occurs deeper in the tissue, it takes longer for it to reach the receiver (assuming constant acoustic velocity). The intensity of the reflection sets the brightness in the image, while the time difference sets the location in the image. Muscle ultrasonography relies on the reflection of waves by sheets of collagenous tissue called perimysium, which surrounds the muscle fascicles. Over the last few decades, muscle ultrasonography has been used to determine the angle and length of these fascicles.

Traditionally, fascicle lengths have been manually estimated from ultrasound images using curvimeters (e.g. Fukunaga et al., 1997; Kuno & Fukunaga, 1995). While such methods can provide reproducible results, they can be time consuming. This becomes especially problematic when fascicles change length rapidly and ultrasound images need to be sampled at a high rate for longer periods of time (e.g. 43 Hz for 10 s during walking, Bohm et al., 2018). Technological advancements in recent years have enabled semi-automated ultrasound tracking, which can reduce required manual labour. Algorithms can be divided into feature detection and feature extraction algorithms (Marzilger et al., 2018).

Feature detection algorithms often use a Hough transform to detect line-like features (Ryan et al., 2019; Y. Zhou & Zheng, 2008), whereas feature extraction algorithms often rely on optic flow to extract change in orientation and/or position of features (Brennan et al., 2017; Cronin et al., 2011). These feature extraction algorithms have the disadvantage that they are time-dependent, as the estimated fascicle length at image (i) depends on the estimated fascicle length at image (i-1). Consequently, the length of the first image needs to be set manually, and the estimated length will be different when the same image would have been presented later. Since feature detection algorithms are generally time-independent, they do not have this disadvantage. Instead, they have the disadvantage that the user needs to make more judgement calls on how to detect features of interest in the ultrasound image. Current feature detection algorithms are not fully automated since they rely on manually selecting a region of interest (e.g. Marzilger et al., 2018) and/or manually detecting aponeuroses (Marzilger et al., 2018; Ryan et al., 2019). Altogether, although current ultrasound algorithms can reduce manual labor required for fascicle length estimation, there is room for improvement since they are not yet fully automated.

We here present a fully automated ultrasound algorithm that estimates muscle fascicle length from ultrasound images using feature detection. The algorithm applies a three-step method, with separate detection of aponeuroses and fascicles. We expect our algorithm to detect a decrease in fascicle length with knee moment and an increase with knee angle. Due to relatively constant knee moment arm across knee angles used in the experiment (15-25 deg)(Krevolin et al., 2004), we expect the effect of knee angle to be linear. Due to non-linear tendon force-length curve (Lichtwark & Wilson, 2008) and non-linear muscle thickness changes during contraction (Azizi et al., 2008), we expect the effect of moment to be non-linear. We therefore test for a quadratic effect of moment, and a linear effect of knee angle. We apply our algorithm on ultrasound images of human vastus lateralis, recorded during isometric knee extension moment production.

Methods

Human subjects produced isometric knee extension moment while ultrasound images were recorded. Muscle pennation angle, thickness and fascicle length were estimated using a fully automated algorithm and related to knee moment and knee angle.

Nine healthy participants (6 male, 3 female, leg length = 87.4 ± 4.9 cm, body mass = 70.6 ± 13.1 kg, mean ± s.d.) produced knee extension moment M_{knee} against an isometric dynamometer (Biodex, Biodex Medical Systems, NY, USA). Participants produced knee moment in stepwise manner (10 s per step, 0-45 N-m range) at three different knee angles (15, 20 and 25 deg). Images were obtained from vastus lateralis using ultrasound (5 cm probe, 11 MHz basic-mode; Logiq E9, General Electric, Fairfield, USA), recorded at 30 Hz and approximately synchronized with moment recordings (via a sync pulse). In a separate trial, maximal voluntary contraction (MVC) strength of quadriceps was determined at 20 deg and 70 deg knee angle. The latter angle is thought to be closer to the plateau of the knee extension moment-angle relationship (e.g. Kulig et al., 1984).

A three-step method was used to estimate pennation angle φ, muscle thickness T and fascicle length L_{fas} from the ultrasound image (see Fig. 2). Filtering was done separately for aponeurosis and fascicle detection. For muscle fascicles, the Hough transform was used to detect the most frequently occurring lines in the image. Fascicle length was determined from muscle thickness and pennation angle (see Fig. 1).

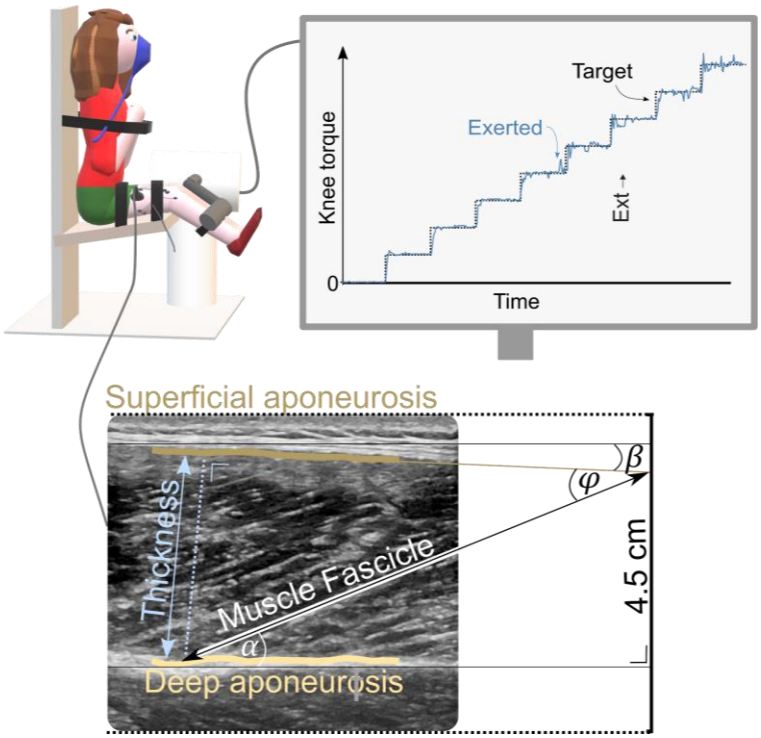

Figure 1: Experimental set-up. Upper plot: Human subject exerted knee extension torque against an isometric dynamometer in a step-wise manner, specified by a target that was displayed on a monitor in front of the dynamometer. Lower plot: Muscle thickness T, muscle fascicle angle α, superficial aponeurosis angle β and pennation angle φ were estimated from ultrasound image and used to calculate muscle fascicle length L_{fas}.

Ultrasound images were filtered using a MATLAB implementation of a Frangi-type Hessian filter (Kroon, 2020; Frangi et al., 1998). This filter uses eigenvalues of the Hessian of the image to highlight line-like structures with a certain thickness, thereby facilitating the identification of both muscle fascicles and aponeuroses (that are both line-like). The thickness range was 8-15 pixels for the aponeurosis and 1-3 pixels for the fascicles, resulting in filtered images referred to as aponeurosis image and fascicle image respectively. The ultrasound image was in the middle of the collected image (see Fig. 3). The boundaries of the ultrasound image were detected using edge detection and Hough transform specifically aimed at the horizontal and vertical angles. The outside edge of the image was cut out to remove boundary effects of the Hessian filter (20-30 pixels).

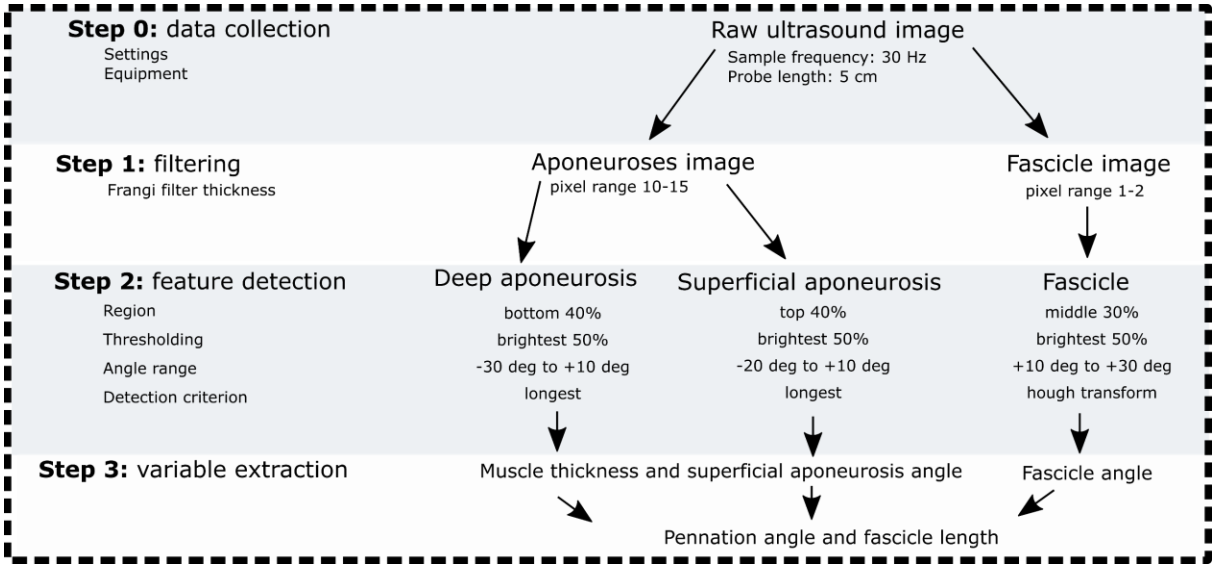

Figure 2: Overview of ultrasound algorithm. Raw ultrasound images were filtered with either a fine pixel range (fascicles) or a rough pixel range (aponeuroses). Aponeuroses and fascicles were identified using feature detection and used to estimate muscle thickness, superficial aponeurosis angle and fascicle angle. It was assumed that superficial aponeurosis was in the upper 40%, deep aponeurosis in the lower 40% and fascicles in the middle 30%. A different range of angles was considered for each category of objects (i.e. fascicle, superficial aponeurosis, deep aponeurosis).

The aponeurosis image was thresholded (brightest 50%, imbinarize, MATLAB) and split: the upper 40% was used for the superficial aponeurosis, while the lower 40% was used for the deep aponeurosis. Objects were filtered based on orientation (bwpropfilt, MATLAB). Angle ranges for the superficial and deep aponeuroses were -20 to +10 deg and -30 to +10 deg respectively. Here, positive angles are counter-clockwise with respect to the right horizontal, which points in proximal direction. The two longest objects were selected (bwpropfilt, MATLAB). If there was little difference in length between these two (<20% difference), the object closed to the fascicles was chosen (regionprops, MATLAB). If not, the longest was chosen. Images were discarded if they did not contain aponeurosis objects with a minimal principle axis length of 200 pixels or principle axis lengths that were less than 5 times longer than the secondary axis lengths. Holes in the deep aponeurosis were filled (imfill, MATLAB), as well as gaps smaller than 5 pixels. The deep aponeurosis boundary was defined as the first black pixel encountered in an upward search that started from the deepest white pixel on the deep aponeurosis object. The superficial aponeurosis boundary was defined as the deepest white pixel on the superficial aponeurosis object. Muscle thickness was defined as the number of pixels between the superficial and deep aponeurosis boundaries (see Fig. 1), averaged at 17 longitudinal locations. Superficial aponeurosis angle (β) was determined from the proximal half of the superficial aponeurosis boundary.

The fascicle image was thresholded (brightest 50%, imbinarize, MATLAB), the middle 30% was selected and used for (Sobel) edge detection (edge, MATLAB). The Hough transform was used to detect angles within the ultrasound image (hough, MATLAB). Angle and distance resolution were set to 1 deg and 1 pixel respectively. Angles within the range 10-30 deg relative to the superficial aponeurosis were considered. Fascicle angle α was defined as the weighted-average of the 10 most frequent occurring angles, with the number of occurrences as weight factors.

Pennation angle φ was defined as the difference between fascicle angle α and superficial aponeurosis angle β,

$$\varphi = \alpha - \beta$$

Muscle fascicle length L_{fas} was defined as thickness T divided by the sine of pennation angle φ (see Fig. 1),

$$L_{\text{fas}} = \frac{T}{\sin(\varphi)}$$

We tested for the effects of knee angle and knee moment on pennation angle, muscle thickness and fascicle length using a linear mixed effects model, with fixed effects for knee angle (linear), moment (quadratic), and random effects for between-subjects variability.

Results

A typical example of knee moment, pennation angle, muscle thickness and fascicle length during the experiment is shown in Fig. 3 (knee angle = 20 deg). Fascicle angle increased with 67% in this subject, from about 15 deg to about 25 deg. Since fluctuations in muscle thickness were smaller (~10%), changes in muscle fascicle length were mostly due to changes in muscle fascicle angle. Fascicle length decreased with about 67%, from about 9 cm to about 6 cm. As intended, knee moment increased from 0 to 45 N-m in 9 consecutive steps. Fascicle length estimates and moment measurements were used to construct a fascicle length-moment relationship (lower right plot, Fig. 3).

Across subjects, knee moment and knee angle influenced pennation angle, muscle thickness and fascicle length (see Fig. 4). Pennation angle increased with knee moment, from 15.8±1.2 deg (mean±s.d.) at rest to 21.1±1.1 deg at 45 N-m (averaged across knee angles). Pennation angle was on average 1.0±0.0 deg smaller at 25 deg knee angle than at 15 deg knee angle (averaged across knee moments). Pennation angle changed quadratically with knee moment (coefficients: -0.004±0.0005 [mean±s.e.] deg·N-m^{-2} and 0.29±0.02 deg·N-m^{-1}, p-values: $6\cdot10^{-14}$ and $1\cdot10^{-26}$) and decreased linearly with knee angle (slope: -0.10±0.02, p-value = $2\cdot10^{-5}$) (adjusted R^2 = 0.69). Muscle thickness initially increased from 2.6±0.1 cm at rest to 2.7±0.1 cm at 5.6 N-m and then decreased to 2.6±0.1 cm at 45 N-m (averaged across knee angles). Muscle thickness was on average 0.05±0.00 cm larger at 25 deg knee angle than at 15 deg knee angle (averaged across knee moments). Muscle thickness changed quadratically with knee moment (coefficients: $1.6\cdot10^{-4}$±$3.9\cdot10^{-5}$ cm·N-m^{-2} and 0.005±0.002 cm·N-m^{-1}, p-value = $1\cdot10^{-6}$) and increased linearly with knee angle (slope: 0.005±0.002 cm·deg^{-1}, p-value = 0.008) (adjusted R^2 = 0.90). Fascicle length decreased from 9.5±0.4 cm at rest to 7.1±0.2 cm at 45 N-m (averaged across knee angles). Fascicle length was on average 0.6±0.0 cm larger at 25 deg knee angle than at 15 deg knee angle (averaged across knee moments). Fascicle length changed quadratically with knee moment (coefficients: 0.001±0.0002 cm N-m^{-2} and -0.1±0.008 N-m^{-1}, p-values: $5\cdot10^{-15}$ and $5\cdot10^{-35}$) and increased linearly with knee angle (slope = 0.06±0.008 cm·N-m^{-1}, p-value = $7\cdot10^{-12}$) (adjusted R^2 = 0.85).

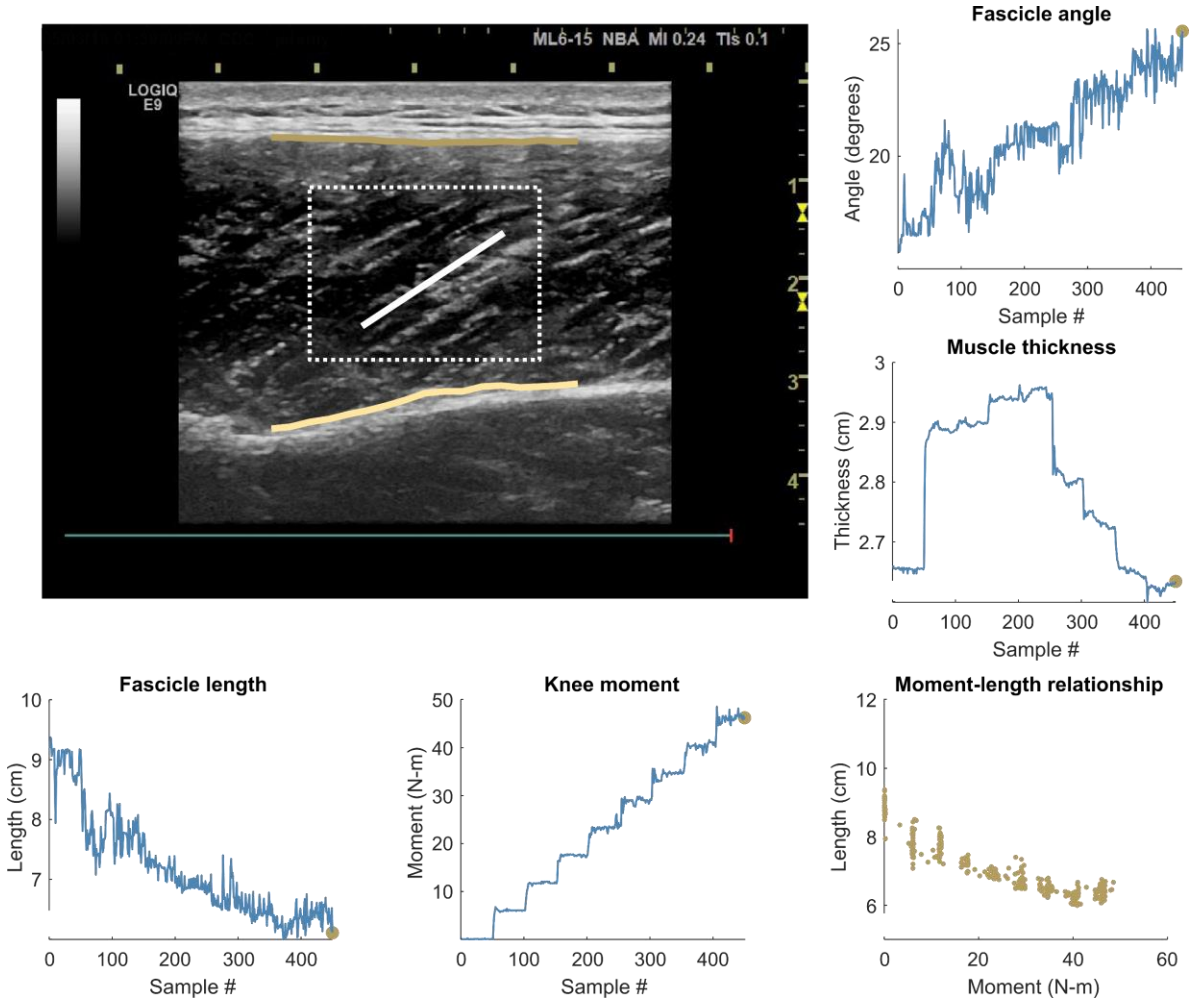

Figure 3: Typical example of outputs from ultrasound algorithm. Blue lines connect individual data points (brown dots). All subplots except for the lower right show the current data point only (i.e. the one corresponding to the image shown), the lower right shows all data points. Upper left: ultrasound image overlaid with identified aponeuroses (brown: superficial, yellow: deep). Dashed white box indicates the region considered for fascicle detection. Thick white line indicates the most frequently occurring line according to Hough transform (note that its angle does not equal fascicle angle, which is a weighted-average of the 10 most frequent occurring angles). Upper right: fascicle angle α increased during this trial. Mid right: thickness T increased rapidly from rest and decayed later in the trial. Lower left: fascicle length L_{fas} decreased during the trial. Lower mid: knee moment increased during the trial in a step-wise manner up to about 45 N-m. Lower right: fascicle length decreased with knee moment.

152

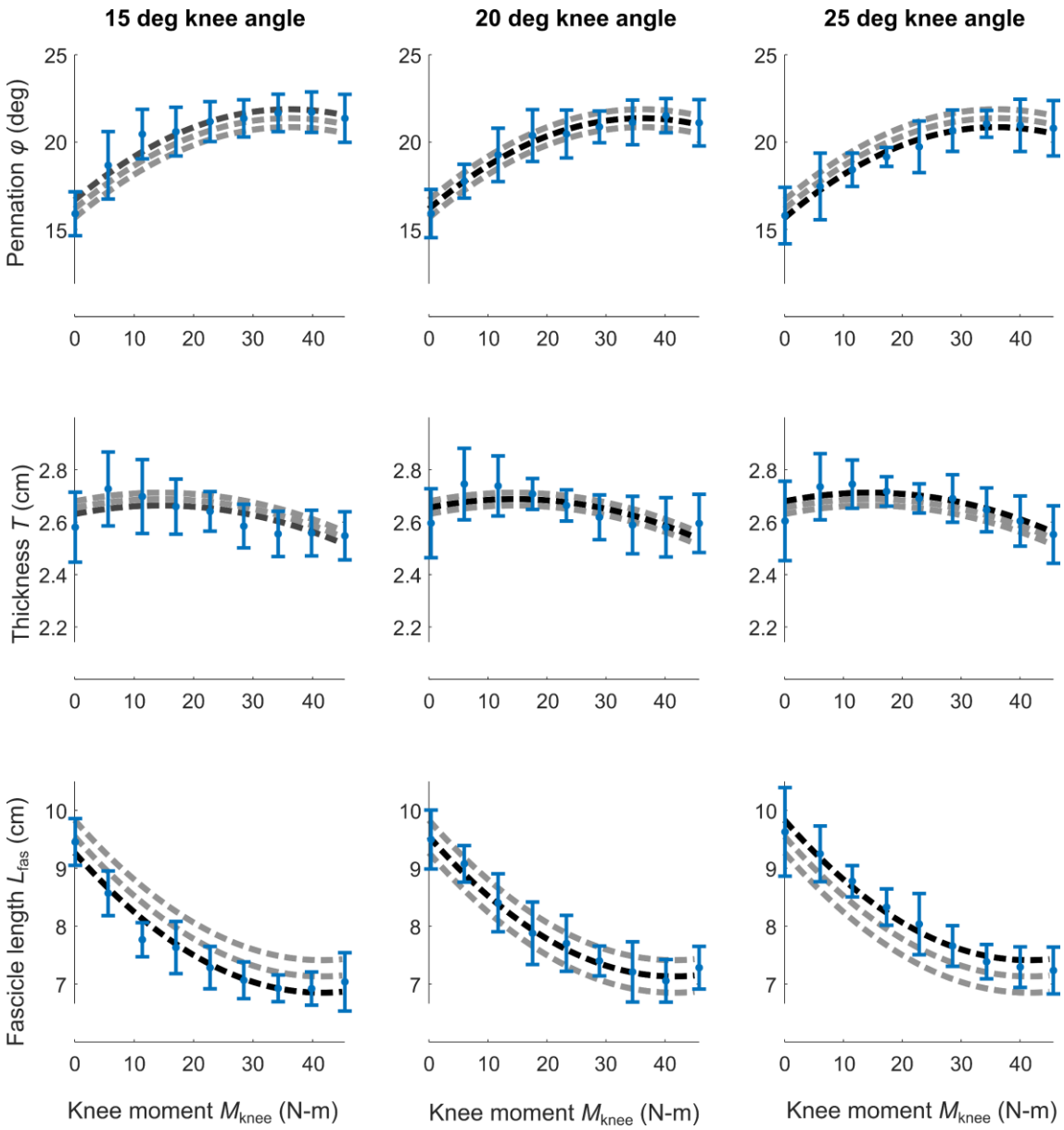

Figure 4: Effect of knee moment M_{knee} on pennation angle φ, muscle thickness T and fascicle length $L_{\text{fas.}}$ Blue dots and error bars indicate mean and s.d. across subjects. Dashed lines show the effect of knee moment according to mixed effects regression (black: current knee angle, grey: other knee angles). Upper plots: pennation angle changed quadratically with knee moment and decreased with knee angle. Middle plots: thickness changed quadratically with knee moment and increased with knee angle. Lower plots: fascicle length L_{fas} changed quadratically with knee moment and increased with knee angle.

Discussing

In the current study, human subjects produced isometric knee extension moment while ultrasound images were recorded. Muscle pennation angle, thickness and fascicle length were estimated using a fully automated algorithm. Knee moment and knee angle significantly affected pennation angle, thickness and fascicle length. In the following, the results will be discussed considering both underlying mechanisms and previous findings.

Fascicle length decreased from 9.5±0.4 cm at rest to 7.1±0.2 cm at 45 N-m, in line with reported previously under similar conditions (Fukunaga et al., 1997; Ichinose et al., 1997). Considering isometric muscle-tendon complex and moderate pennation angles (<25 deg), fascicle length changes should have induced series elastic tissue (SEE) length changes. SEE length changes larger than 1 cm seem inconceivable with the common Hill-type muscle model assumption that SEE elongates 4% of resting length at F_{max}, which would imply a maximal SEE length change of 0.64 cm when assuming a rest length of 16 cm (Soest & Bobbert, 1993). This discrepancy may be explained by the slack tendon hypothesis, referring to the idea that tendons become slack at small muscle-tendon complex lengths (Fukutani et al., 2017). Slack tendons could result in large muscle fascicle shortening at low forces, since any non-zero force will remove the slack. Muscle-tendon complex length was small in the current study, due to the small knee angle (15-25 deg flexion). In line with this slack tendon hypothesis, vastus lateralis fascicles shorten ~0.5 cm at 110 deg knee flexion when exerting 10% of maximal force, whereas they shorten ~3 cm for the same exertion at full knee extension (Fukunaga et al., 1997). A considerable portion of total fascicle shortening in the current study occured at the smallest moment values, substantiating the presence of slack tendon. In addition to substantial fascicle length changes, pennation angle and muscle thickness changed considerably at low moments. This is in line with the idea of variable gearing in pennate muscle (Azizi et al., 2008; Roberts et al., 2019). If increases in pennation angle changes are accompanied by increases in muscle thickness, the ratio of muscle-tendon complex shortening to muscle fascicle shortening (i.e. the architectural gear ratio, or AGR) is larger than 1. This favours muscle shortening over muscle force production. If increases in pennation angle changes are accompanied by decreases in muscle thickness, the AGR approaches 1. This favours muscle force production over muscle shortening. The current data suggests that human vastus lateralis muscle operates at high AGR when forces are small and at lower AGR when forces are high. The exemplifies the need for real-time muscle thickness tracking, enabled by automized aponeurosis detection.

The current algorithm is similar to the algorithm described by (G.-Q. Zhou et al., 2015), which uses a combination of Gabor and Hough transform. Both Gabor transform and Hessian filtering result in highlighting of line-like structures with a specified thickness. One of the differences between the two is that the angle of line-like structures is specified with Gabor transform, but not with Hessian filtering. Ryan and colleagues used a Frangi Hessian filter in combination with Hough transform for fascicle detection as in the current study, while relying on manual aponeurosis detection for muscle thickness estimation (Ryan et al., 2019). It may be insightful to compare manual aponeurosis detection against the automated aponeurosis detection as proposed in the current study. Marzilger and colleagues created a feature detection algorithm that does not rely on Hough transform (Marzilger et al., 2018). Disadvantages of such non-Hough feature detection algorithms include processing time and need for additional parameters. The current algorithm implements a similar strategy for aponeurosis detection, where their semi-automated method relies on manual aponeurosis detection. Despite their differences, feature detection algorithms all rely on detecting specific objects within the ultrasound image. Feature extraction algorithms belong to a different category, often relying on the change in optic flow within the image (Cronin et al., 2011). The advantage of such algorithms is that they use all the information in the image, rather than relying on specific objects that are suspect to noise. However, due to integrating a rate signal, optic flow algorithms are prone to drift and

initial conditions. Consequently, these algorithms are history dependent as the estimate in image (i) depends on the estimate in image (i-1). Feature detection algorithms have been used extensively for estimating fascicle lengths in human calf muscle, which have relatively short fascicles (e.g. Lichtwark & Wilson, 2006). Specifically, Ultatrack is a popular optic flow software that is freely available (Farris & Lichtwark, 2016). An issue with Ultratrack is that it relies on manually identifying a representative fascicle at the start of the analysis, which is not possible for longer fascicles like vastus lateralis (6-12 cm) when using regular ultrasound probes (4-5 cm). This issue may be solved using wider probes (Brennan et al., 2017), but these wider probes are not always available and may hamper movement flexibility.

The current algorithm does not work well on non-uniform fascicles and curved fascicles. In both cases, object angle will differ across the image. Subtle changes in the image will determine which population of fascicles dominates the image and thereby influence the estimated fascicle angle from the Hough transform. While absolute fascicle length may be difficult to estimate under such circumstances, fascicle length changes could be obtained from optic flow. Therefore, optic flow algorithms can potentially outperform the current algorithm under these circumstances. However, these circumstances are unlikely to occur during moderate moment production. The amount of muscle fascicle curvature is thought to increase towards higher force levels (Rana et al., 2014), which are unlikely to occur in vastus lateralis during locomotion, which is submaximally active throughout most of the stride cycle in both walking and running (Bohm et al., 2018). Altogether, despite some limitations for fascicle detection in extreme conditions, the current algorithm should provide valid estimations during moderate force production as during locomotion.

Since the current algorithm is fully automated, many images can be analysed in a relatively short amount of time. This is important for ultrasound recordings during locomotion, since a high image collection rate and a long image collection duration may be desired. Image collection rate needs to be high because fascicle length can change rapidly, especially during faster movements like running. It is important that lengths changes are detected in time, because delay can cause synchronization error with forces, which can result in erroneous work loops. Whereas image collection duration needs to be at least 1 s to allow for capturing a full step, one likely wants to collect several steps to average out step-to-step variability. A 5 s collection at 30 Hz results in 150 images, which would take about 150 minutes for manual detection (assuming 1 minute per image). In contrast, the algorithm presented here only needs 1.54 s per image on a modern laptop (i5 processor), so that the same analysis could be done within 4 minutes. Since 57% of total processing time is spend on Frangi filtering, one could filter once and do a fast sensitivity analysis on the feature detection. Altogether, implementation of the current algorithm for cyclic movements like locomotion may have several advantages over both manual detection and semi-automated detection.

Since various algorithms are currently available, the question arises which to use under which circumstances. In order to answer this question, it would help if algorithms would be openly accessible so that they can be tested. Ultrasound simulation software programs like Field II (field-ii.dk) and/or open access ultrasound data may be used to compare the accuracy and validity of algorithms.

Conclusion

We here present a fully automated ultrasound algorithm that provides estimates of fascicle lengths and pennation angles, comparable to previous observations and in line with underlying mechanisms. We expect that the algorithm may be applied for ultrasound analysis during cyclic movements, including human locomotion.

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Competing interests

The authors declare no competing or financial interests

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Data availability

Ultrasound algorithm and typical example of ultrasound data is available on:
https://github.com/timvanderzee/ultrasound-automated-algorithm

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