Correlations between hamstring muscle architecture, maturation and anthropometric measures in academy soccer players

Running Head: Hamstring architecture and anthropometry

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Abstract

Purpose: Muscle architecture is associated with motor performance and muscle injury. While muscle architecture and knee-flexor eccentric strength change with growth, the influence of anthropometric measures on these properties is rarely considered. This study aimed to investigate the relationship between hamstring muscle architecture and knee-flexor eccentric strength with anthropometric measurements.

Methods: Sixty male footballers (16.6±1.05years) from the U16, U17 and U19 teams of an elite soccer club were included in this study. Fascicle length, pennation angle and muscle thickness of the bicep femoris long-head (BFlh) and semimembranosus (SM) muscles were measured in both legs using ultrasound. Knee-flexor eccentric strength, height, body mass (BM), leg length, femur length and peak-height velocity (PHV) were measured within 1-week of the ultrasound images. A stepwise regression, and one-way analysis of variance (ANOVA) tests were used to evaluate the effects of age, maturity and anthropometric measurements on muscle properties.

Results: Variance within BFlh and SM muscle thickness ($r<0.61$), SM pennation angle ($r<0.58$) and knee-flexor eccentric strength ($r=0.50$) were highly related to BM. We observed no significant correlations between muscle architecture and age ($p>0.29$). However, moderately greater BFlh muscle thickness was shown for the post-PHV compared to the PHV group (ES±90%CL;0.72±0.49).

Conclusions: In conclusion, weak correlations between muscle architecture and anthropometric measurements suggests that other factors (i.e., genetics, training regime) influence muscle architecture. The moderate effect of maturity on BFlh muscle thickness strongly suggests post-PHV hypertrophy of the BFlh muscle. Our results confirmed previous findings that eccentric knee-flexor strength is influenced by BM.

Keywords:

Musculoskeletal Imaging, Fascicle Length, Pennation Angle, Muscle Thickness, Maturity Offset

Introduction

Physical arrangement of muscle fibres, referred to as muscle architecture, is one of the primary drivers of muscle function.[1] It is theoretically accepted that shorter muscle fibres present a lower maximum shortening velocity which likely influence the contraction velocity at the entire muscle level.[2] In contrast, the angle between the fascicle's line of action and the aponeurosis (*i.e.*, pennation angle) is considered proportional to the number of sarcomeres in parallel and the maximum muscle force-generating capacity.[2]

In addition to its influence on motor performance, muscle architecture has been associated with muscle strain injuries.[3] Longer muscle fascicles are assumed to juxtapose more sarcomeres in series and will exhibit less strain per sarcomere than shorter fascicles for a given muscle-tendon unit strain and stiffness.[4] During the stance and late-swing phases of sprinting, studies using computational musculoskeletal modeling showed hamstring muscles produce very-high forces and withstand large amounts of negative work over repeated strides.[5,6] The application of such singular high-magnitude or repeated stress may exceed the local mechanical limits of the hamstring muscle tissue.[7] In line with such theoretical background, recent evidence suggests that short *bicep femoris* long head (BFlh) fascicle length (<10.56cm) are 4.1 times more likely to sustain a future hamstring injury compared to athletes with longer fascicles.[8] This risk is increased when short BFlh fascicles are associated with low-levels of eccentric knee-flexor strength. This has led to practitioners identifying athletes of greatest hamstring injury risk based on their BFlh fascicle lengths and knee-flexor eccentric strength.[3,9] Hamstring injuries are increasingly prevalent within soccer populations and account for 12% of all injuries sustained by soccer players.[10] Therefore, the ability to identify soccer players at risk of injury through the investigation and development of hamstring muscle architecture and eccentric strength is becoming increasingly common.[11]

Muscle architecture measures are commonly determined via B-mode ultrasound images and computed using trigonometry approaches that assume pennate muscle as a parallelogram.[12] However, muscle fibres do not consistently present a homogeneous architectural arrangement along their whole length.[12] Further, fascicles are often curved, which is not accounted for when fascicles are represented by a straight line.[13] Therefore, future works have been encouraged to adopt more appropriate ultrasound methods (e.g., extended field of view (EFOV) imaging) that would enable the exploration of muscles with long fascicles and spatially heterogeneous architecture (*i.e.*, BFlh).[14,15]

While BFlh architecture have been considered as a surrogate of hamstring muscle heads, different muscles have distinct structural arrangements and mechanical properties.[16] It is therefore important to monitor the geometry of the other hamstring muscle heads to capture a better global understanding of the hamstring muscle group.[16] The fascicles of the *semimembranosus* (SM) muscle head are pennate thus, possess a similar architecture to that of the BFlh while the fascicle length of a muscle with a fusiform arrangement (*i.e.*, *semitendinosus*) is likely related to femur length and player size,[16] which is largely related to biological age. Further, maturity leads to increased body mass (BM) and fat-free mass.[17] However, little is known regarding how the maturation process influences muscle architecture. Literature suggests that muscle thickness and fascicle length increase while pennation angle remains stable throughout maturation.[18,19] One study showed an increase in *gastrocnemius medialis* and *vastus lateralis* (VL) muscle thickness, pennation angle, and VL fascicle length from pre-to-post peak height velocity (PHV).[17] Overall, this data suggests that muscle architecture changes with growth. However, the influence BM and height may respectively have on muscle architecture and knee-flexor eccentric strength is rarely considered.[20] Such investigations may contribute (i) to differentiate between training-induced adaptations versus normal maturation-induced changes and (ii) to identify which anthropometrical measurements share relationships with hamstring muscle architecture.

The main aim of this study was to evaluate relationships between hamstring muscle properties involved in motor performance and injury risk (*i.e.,* fascicle length, pennation angle, muscle thickness, knee-flexor eccentric strength) and anthropometric measurements within a population of highly trained youth soccer players. Additionally, this study aimed to identify age and maturation effects on hamstring architecture and knee-flexor eccentric strength.

Methods

Participants

Sixty male footballers (16.6±1.05years, 174.4±7.4cm, 63.8±9.2kg) including 17 participants from the Under-16 (15.3±0.4years, 170.9±10.1cm, 59.9±11.2kg), 17 participants from the Under-17 (16.4±0.4years, 174.7±7.5cm, 65.2±9.7kg) and 26 participants from the Under-19 (17.5±0.7years, 177.5±4.5cm, 66.3±6.8kg) teams of an elite French Ligue-1 football club partook in this study. Participants, who represented all football playing positions (goalkeepers = 5, defenders = 21, midfielders = 20, forwards = 14), joined the club at 13-years old thus, their experience of professional football training ranged from 2-5 years depending on their age. Since all athletes competed in the same sport, potential confounding effects of different training regimes on muscle architecture were unlikely. Each team followed the same training model established by the football club in which each training day focused on the development of the same specific attributes (i.e., strength, aerobic, technical, tactical) and all teams played matches of 90 minutes. Participants completed an average of five 90-minute training sessions, one game and one rest day per-week.

Design

During the first 3 weeks of preseason training, athletes had muscle architecture measurements taken for the BFlh and SM hamstring muscles from both legs on one occasion using ultrasound. Peak knee-flexor eccentric strength measurements using a Nordbord (Vald Performance, Queensland Australia) and anthropometric measurements (BM, height, sitting height, leg length and femoral length) were taken within one week of the ultrasound scans.

Ultrasound scans were taken pre-training to minimize any acute effects soccer-specific training may have on knee-flexor muscle architecture. As the preseason starts dates were staggered by one week (i.e., U17 started one week later than Under-19 and Under-16 started 2 weeks later than Under-19) the ultrasound scans were all completed within 1-week of each players' preseason start date to minimize any knee-flexor muscle architecture adaptations induced by soccer-specific training following the off season.

All knee-flexor eccentric strength testing was undertaken pre-training following 24 hours of rest without physical exertion. Each player completed a standardized warm-up protocol before completing the testing. The warm-up consisted of 5 minutes of light cycling (>80 - <120 watts) on a static gym bike, 2x 10 repetitions of unilateral Swiss ball rollouts per leg and 3 submaximal Nordic hamstring exercise repetitions. Verbal encouragement was provided to the players throughout the testing procedure.

Athlete monitoring procedures, data collection and anonymized data publication, were accepted as part of the participant's contractual agreement with the football club. In addition, the club gave consent for the research and publication of its findings thus, additional ethical approval was not required for this study however, it complied to the declaration of Helsinki.[21] Participants were excluded from the study if there were injured at the time of testing or were absent due to illness.

Methodology

Ultrasound Imaging and Processing

The BFlh and SM of each leg were scanned in lying prone position using an ultrasound device (Aixplorer V11, Supersonic Imagine, Aix-en-Provence, France) and transducer (2–10MHz, SL10-2, Supersonic Imagine). The proximal (*i.e.*, conjoint tendon of the BFlh and *semitendinosus* muscles) and distal musculo-tendinous junctions were located. Cross-sectional images were acquired in the transversal plane to determine the path of the BFlh muscle. Scans were then performed longitudinally along this path in EFOV panoramic mode in order to not extrapolate the nonvisible part of the fascicle.[13] This mode uses algorithms that stitches successive frames together to obtain panoramic images. Scans were performed by following fascicles in the superficial compartment from the proximal insertion following the fascicle's plane while trying to manipulate the transducer, so the fascicles remained continuous while superficial and deep aponeuroses were visible. The path of the best image was marked over the skin to position the transducer during EFOV scans. Scans were performed along the muscle midline until reaching the distal portion, at approximately 90% of the femur length (Fig.1A). The transducer was moved slowly and continuously along the marked path with a constant pressure from the distal to the proximal musculo-tendinous junctions while transducer orientation was continuously adjusted to stay in the fascicle plane. Total scan time was 10–15s per scan. This was repeated for each muscle until two images with clear, visible fascicles and aponeuroses were obtained.

The muscle insertion, superficial aponeurosis, deep aponeurosis, and fascicles were then manually digitised using the ImageJ multipoint tool (ImageJ v.1.48; Bethesda, MD). For SM and BFlh muscles, superficial and deep aponeurosis and four clearly visible fascicles spreading throughout the muscle distally, medially, and proximally were required. As fascicles cannot consistently be imaged over their entire course, each fascicle was digitized over ten points of its clearly visible (*i.e.*, echogenic) portions (Fig.1B). The same procedure was used to label aponeuroses. The x-y coordinates of all labelled points were recorded (Fig.1C) and fitted using a second-degree polynomial order separately applied for each aponeurosis and fascicle (Fig.1D) using custom-written scripts (Origin 2020, OriginLab, USA). The coefficients of the polynomial fit were used to produce a dataset over an evenly distributed 10-points comprised: (i) between the insertions of each fascicle on the superficial and deep aponeurosis; (ii) between the most distal and proximal digitized points for each aponeurosis. Fascicle length was computed as the sum of the distance between each pair of points. Pennation angle was calculated as the average of the angle computed at 25, 50, 75 and 100% of the fascicle length to accommodate for angle spatial variability throughout the muscle. This procedure was applied for each fascicle and values were averaged across images and fascicles totaling 40 measured fascicles per player.

Muscle thickness was measured from the superficial to the deep aponeurosis using the average of three measurements at 33%, 50% and 66% of the length of the muscle, commencing at the upper insertion to evenly represent the muscle belly.

Standardized typical error ± 90% confidence internals of fascicle length measurements averaged between both legs for the BFlh was 0.19 ± 0.06 with a coefficient of variation ± 90% confidence internals of 2.9 ± 0.9.

Standardized typical error ± 90% confidence internals of fascicle length measurements averaged between both legs for the SM was 0.18 ± 0.06 with a coefficient of variation ± 90% confidence internals of 3.2 ± 0.9.

The fascicles of the semitendinosus hamstring muscle head have a fusiform arrangement where they run in parallel with the aponeuroses' throughout the length of the muscle thus, it was not possible to accurately identify the origin and endpoint of the fascicles and therefore not possible to accurately measure the muscle architecture using the EFOV ultrasound technique.[16] In addition, the BFlh and SM muscles are more commonly injured than the semitendinosus thus, are of greater interest within this context.[16]

Insert Figure 1 here.

Femoral length

A Cescorf aluminum large-bone anthropometer sliding caliper (60cm) was used to measure femoral lengths. Participants stood in a neutral position and measurements were taken from the superior point on the greater trochanter of the femur to the superior point on the lateral border of the head of the tibia. Measurements were repeated 3-times and averaged on each leg to ensure precision. Standardized typical error ± 90% confidence internals of femoral length measurements averaged between both legs was 0.04 ± 0.01 with a coefficient of variation ± 90% confidence internals of 0.25 ± 0.1.

Biological maturity

Biological maturity was determined using years from PHV. Participants were categorized into three maturity groups–1.Pre-PHV (Maturity offset=<-1), 2.PHV (Maturity offset=-1<x>1), 3.Post-PHV (Maturity offset=>1).[17] Maturity offset was calculated using the following equation for measuring sex-specific PHV for males:

Maturity Offset=-29.769+0.0003007*leg length and sitting height interaction-0.01177*age and leg length interaction+0.01639*age and sitting height interaction+0.445*leg by height ratio.[22]

In this equation, interactions refer to the multiplication of the two factors involved (*i.e.*, leg length and sitting height interaction=leg length*sitting height).

No participants qualified for the pre-PHV category; thus, comparisons were performed between the circa-PHV (n = 15, age = 15.4 years, height = 166.2 cm, body mass = 53.8 kg, maturity offset = 0.4 au) and post-PHV (n = 45, age = 16.6 years, height = 177.2 cm, body mass = 67.1, maturity offset = 2.1 au) categories.

The Mirwald equation for calculating maturity offset exhibited similar levels of accuracy and reliability as other equations designed to calculate biological age.[23]

Knee-flexor eccentric strength

Knee-flexor eccentric strength was assessed using a Nordbord. Data was collected using a protocol previously outlined by Opar et al.[24] Participants regularly carried out eccentric hamstring strength exercises, including the Nordic hamstring exercise twice weekly within their normal training programs so were familiar with this test.

Anthropometric Measurements

Participant standing and sitting height were measured using a portable SECA 213 stadiometer (SECA, Hamburg, Germany). Participants were instructed to remove any footwear and stand on the highlighted foot markers on the base of the stadiometer with their hips, back and back of the head all in contact with the stadiometer support and to keep the head within the Frankfort horizontal plane (e.g., upper ear canal aligned with lower orbitals). The stadiometer arm was lowered until it made contact with the most superior part of the head. The participant was then asked to take a deep breath in a nd hold this while the measurement was taken. This measurement was repeated three times and the average calculated to ensure a consistent measurement was taken.[25]

To measure sitting height, the same protocol was used but the height was measured between a platform where the base of the stadiometer was placed. The participants were instructed to sit on the stadiometer base (on the platform) and the stadiometer arm was lowered to the most superior point of the participant's head where the measurement was then taken once the participant had taken a deep breath.[26] Body mass was calculated using a HD-366 Digital Weight Scale (Tanita, IL, US) and were collected following previously outlined procedures.[25]

Statistical Analysis

Data are presented as Mean±SD, 90% Confidence Intervals. Data which were not normally distributed were log transformed to minimise bias from non-uniformity error. One-way analysis of variance (ANOVA) tests were performed to analyse the variance of muscle architecture with age and maturity offset. Models were created using R software (v3.4.1, R Foundation for Statistical Computing) using the MASS package step function (v7.3) to perform a backward elimination stepwise-regression to analyse relationships between muscle architecture and knee-flexor eccentric strength as dependent variables with anthropometric measures as independent variables. Significance was set at $p<0.05$ for ANOVA and stepwise-regression tests. Linear-regressions were performed using Excel (Microsoft, Washington, USA) where the *r*-value was calculated to analyse the strength of relationships between muscle architecture and anthropometric measures. The magnitude of the correlation coefficients were calculated using Hopkin's scale: ≤0.1 (very small), 0.1-0.3 (small), 0.3-0.5 (moderate), 0.5-0.7 (high), 0.7-0.9 (very-high), ≥0.9 (almost perfect).[27] Using a custom Excel spreadsheet,[28] effect sizes were calculated to determine the magnitude of differences between age-groups (U16vsU17, U16vsU19, U17vsU19) and maturity offset groups (circa PHV vs Post-PHV) using Hopkin's scale: 0.2 (Small), 0.6 (Moderate), 1.2 (Large), 2.0 (Very-Large).[27] The likelihood to obtain a meaningful change was assessed with Magnitude based decision framework where the following thresholds were used: 25-75% (possible), 75-95% (likely), 95-99% (very-likely), >99% (almost-certain).[27] The effect was reported as unclear if the probabilities of the effect being positive and negative were >5%.[27] The effects were otherwise reported as the observed value.[27]

Results

Age effect on muscle architecture

We observed no significant correlations between BFlh fascicle length (*p*=0.98), SM fascicle length (*p*=0.94), BFlh pennation angle (*p*=0.62), SM pennation angle (*p*=0.29), BFlh muscle thickness (*p*=0.72) and SM muscle thickness (*p*=0.72) with age group (Table 1).

Insert table 1 here.

Maturity offset effect on muscle architecture

Moderately greater BFlh muscle thickness was shown for the post-PHV category compared to the PHV category (ES±90%CI;0.72±0.49). *Unclear* SM muscle thickness differences were observed between PHV and post-PHV groups (0.16±0.67;Table 2).

Unclear differences for BFlh (0.29±0.70) and SM (-0.02±0.55) fascicle lengths were also found between PHV and post-PHV groups. Additionally, PHV and post-PHV differences in pennation angles were *unclear* for BFlh (0.31±0.78) and SM (0.07±0.65) muscles.

Insert table 2 here.

Relationship between muscle architecture and anthropometry

The relationships between muscle architecture measures and femoral length are shown in figure 2. Small correlations were found between muscle architecture and femoral length (*r*<0.12).

Table 3 shows the relationships between muscle architecture and anthropometric measures. The predictor model was able to account for 24% of the variance observed in BFlh muscle thickness (*r*=0.49) and 37% in SM muscle thickness (*r*=0.61) with BM significantly related to both measures (*p*=0.001).

Weak correlations were found between the BFlh fascicle length and predictor model (*r*=0.34) where BFlh fascicle length was significantly related to femoral length (*p*=0.033) and leg length (*p*=0.012). Strong correlations were found between SM pennation angle and the anthropometric predictors (*r*=0.58). Within this model, SM pennation angle was significantly related to BM (*p*<0.001) and femoral length (*p*=0.025).

Insert figure 2 here.

Insert table 3 here.

Relationships between muscle architecture, anthropometry and knee-flexor eccentric strength

Mean ± SD knee flexor eccentric strength test scores for each team were: U16; 281 ± 80 N, U17; 336 ± 75 N and U19; 295 ± 69 N respectively. Regarding knee flexor eccentric strength test scores in relation to maturation status, the circa-PHV and post PHV groups scored as follows: circa-PHV; 281± 37 N, post-PHV; 309.2 ± 85 N. No significant relationships were observed between knee-flexor eccentric strength and BFlh and SM architecture (Table 4). A large relationship was observed between knee-flexor eccentric strength and BM (p=0.001,r=0.50).

Insert table 4 here.

Discussion

The main aim of this study was to evaluate relationships between hamstring muscle properties involved in motor performance, injury risk and anthropometric data in a substantial cohort of 60 highly trained male youth footballers. Three main findings originate from this study: (i) there were small relationships between hamstring muscle architecture and anthropometric measures, with leg length most notably accounting for 3.8% of the variance in BFlh fascicle length, (ii) knee-flexor eccentric strength was strongly related with BM, (iii) significantly higher BFlh muscle thickness was shown for post-PHV compared to the PHV category.

We observed weak relationships between muscle architecture and anthropometric measurements. Only a slight portion of the BFlh fascicle length variance was explained by anthropometric measurements with leg length accounting for 3.8% of this. Additionally, femoral length accounted for 4.5% of SM pennation angle variance. This suggests that muscle architecture depends, in part on lower-limb segment size. Musculoskeletal imaging is therefore required to accurately appraise individual hamstring muscle architecture to integrate these properties for training individualization purposes. Previous research has shown BFlh muscles with fascicles shorter than 10.56cm are 4.1 times more likely to sustain a future hamstring injury compared to athletes with longer fascicles.[8] As anthropometric measurements cannot be accountable for all variance observed in muscle architecture, musculoskeletal imaging is required to accurately measure static-resting fascicle length and pennation angle to estimate exposure to injury risk. Additionally, these findings suggest it is not necessary to control for athlete size when analysing team muscle architecture data. Furthermore, regular monitoring of muscle architecture using ultrasound can provide information regarding training effects on muscle development and can be used to monitor accelerated changes during earlier growth periods.[15] This may be useful to assess rehabilitation process effectiveness for injured players that may be exposed to persistent atrophy[29] and reduced force-generating capacity[30] as reported in BFlh after muscle injury. Thus, hamstring architecture during injury rehabilitation can be compared to baseline-levels to evaluate how rehabilitation processes counterbalance injury-mediated changes in muscle geometry. Therefore, it has been suggested that where possible, a specialised figure with expertise in musculoskeletal imaging should be introduced into teams interested in implementing such approaches.[15]

Our results suggest it is not possible to solely use basic anthropometric measurements to estimate hamstring architecture. This finding is of importance for investigations using computational models to explore behaviors (*i.e.*, dynamic-length changes) of hamstring muscle fibres during ecological motor tasks such as running or sprinting[5,6]. Such approaches use cadaveric data or MR images to implement muscle-tendon unit length to be able to estimate BFlh fibre length changes. However, our results strengthen that approaches based on generalized models may not be sufficiently personalized to reflect actual hamstring muscle fascicle dynamics, which requires direct assessment of muscle geometry. In this context and given that other factors strongly influence muscle architecture (*i.e.*, genetics, training level), direct measurements from musculoskeletal imaging are required to obtain accurate appraisal of muscle architecture. Similarly, to previous research, where factors such as eccentric hamstring training and exposure to sprint running were assessed as to how they affected hamstring muscle architecture,[11] future studies may investigate this further to investigate which specific training factors influence hamstring architecture within highly trained, youth football players.

Our findings showed the presence of a strong relationship between knee-flexor eccentric strength and BM measured using the Nordic hamstring exercise, with BM accounting for 25% of knee-flexor eccentric strength variance. These results reflect those of a previous study in which BM accounted for 30% of knee-flexor eccentric strength variance.[20] To account for the effect of BM, allometric scaling could be utilized to correct Nordic hamstring test scores to provide relative strength to BM outputs.[20] As our study shows a quarter of knee-flexor eccentric strength is accounted for by BM, it is suggested to use allometric scaling to remove BM influence. Allometric scaling allows for the minimisation of the effect of BM on Nordic hamstring scores, thus, it is possible to assess true changes in knee-flexor eccentric strength.[20] Additionally, it allows accurate comparisons to be made between athletes within the same team regardless of BM to assess individual athlete performance against baseline values to potentially improve performance and minimise injury risks. However, knee-flexor eccentric strength did not share any further meaningful relationships with other anthropometric or hamstring architecture measurements. This did not support previous studies where greater knee-flexor eccentric strength was observed with greater lever-arm (e.g., femoral length) and hamstring fascicle length.[2,16] Numerous factors influence knee-flexor eccentric strength (*i.e.*, muscle volume, neural drive, muscle activation, specific tension, force-length relationship, force-velocity relationship, inter-muscle coordination). It is therefore possible the influence these factors had, while they may have been small, accumulated to account for a proportion of the remaining variance observed for knee-flexor eccentric strength in this cohort.

No meaningful differences in muscle architecture were observed between age-groups. However, only a 6.6cm height difference was observed across all age-groups and only a 1.1kg BM difference between the U17 and U19 age-groups. This modest range of variability across ages may partly explain why hamstring muscle architecture did not share a relationship with age-group. Regarding relationships between muscle architecture and maturity offset, moderately greater BFlh muscle thickness was found for the post-PHV group compared with the PHV group. This is supported by previous research where muscle thickness of the *gastrocnemius medialis* and *vastus lateralis* increased for the post-PHV group compared to the PHV and pre-PHV groups.[17] Previous research reported similar increases in muscle size and hypertrophy for post-PHV participants.[31] This finding is likely to reflect the growth-induced development of contractile material which translated into the amount of force the footballers could produce. However, further research is required to investigate why maturity offset impacted BF but not SM muscle thickness. The growth-related muscle hypertrophy observed in this study was associated with no changes in pennation angles or fascicle length of BFlh and SM. This did not support previous reports of significantly larger pennation angle and fascicle length for post-PHV individuals.[17] Together with previous findings, this study highlights the importance of using maturity offset to make comparisons between adolescent athletes rather than age. Research has shown athletes at different stages of maturity respond more effectively to different training programs.[32,33] Therefore, it is paramount to ensure athletes of the same age but different stages of maturity are not compared under the same protocols[17,18] so training can be individualized to certify the safe, constructive development of each athlete.

Regarding the limitations associated with this study, the participants all play and train within the same club. Thus, other clubs and cohorts of participants may display different relationships due to differences in training regime and exposure to strength training. Therefore, it is important that these results are not generalized across youth football and data would need to be collected on an individual club basis to explore this research within different populations. One limitation of this study may be that no pre-PHV athletes were present within the testing cohort thus, it was not possible to compare muscle architecture between all three maturity offset categories as performed in previous studies.[17,31] This investigation should be further expanded to include younger age-groups within the cohort to evaluate the differences in muscle architecture, anthropometric and knee-flexor eccentric strength measures between pre-PHV, PHV and post-PHV groups. However, the present sample is relatively large compared to existing literature. Using ultrasound scans to assess muscle geometry could be a relevant area of common work between performance and clinical staff to expand hamstring architecture, anthropometric and knee-flexor eccentric strength reference values and improve understanding of hamstring muscle geometry in youth athletes. Additionally, it is important to note that we did not completely control the participant's training regimen. While slight differences in training contents may exist across age groups, all players trained in the same environment, likely resulting in negligible subsequent effect on hamstring muscle architecture. This assumption is strengthened by previous works from our group showing modest changes in hamstring muscle architecture even after high-volume eccentric training.[34] Finally, the authors would like to acknowledge recent criticisms of magnitude based decision framework and they understand other statistical methods may be preferred.[35] However, we believe robust statistical methods have been used to analyze the data and provide accurate results from which to draw appropriate conclusions.

Practical Applications

- Controlling for anthropometric measures is not necessary when screening hamstring muscle architecture.
- BM allometric scaling is required to make between-player Nordic hamstring strength comparisons.
- Using maturity offset measures for screening may allow practitioners to better differentiate muscle adaptations induced by maturation or training effects however, more data is required to investigate this further.
- Ultrasound neuromuscular imaging of the hamstring muscle group may work in conjunction with traditional neuromuscular testing batteries to provide further information regarding hamstring muscle architecture.

Conclusions

In conclusion, this study showed that muscle properties are poorly associated with anthropometric measurements, suggesting it is not necessary to control for anthropometric measurements when considering muscle variables as part of a screening process for injury prevention strategies. Therefore, musculoskeletal imaging is required to accurately measure muscle architecture within a practical environment. Additionally, this study supports previous findings regarding the large influence of BM on knee-flexor eccentric strength performance thus, allometric scaling should be used when between-player comparisons are required.[20]

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Figures

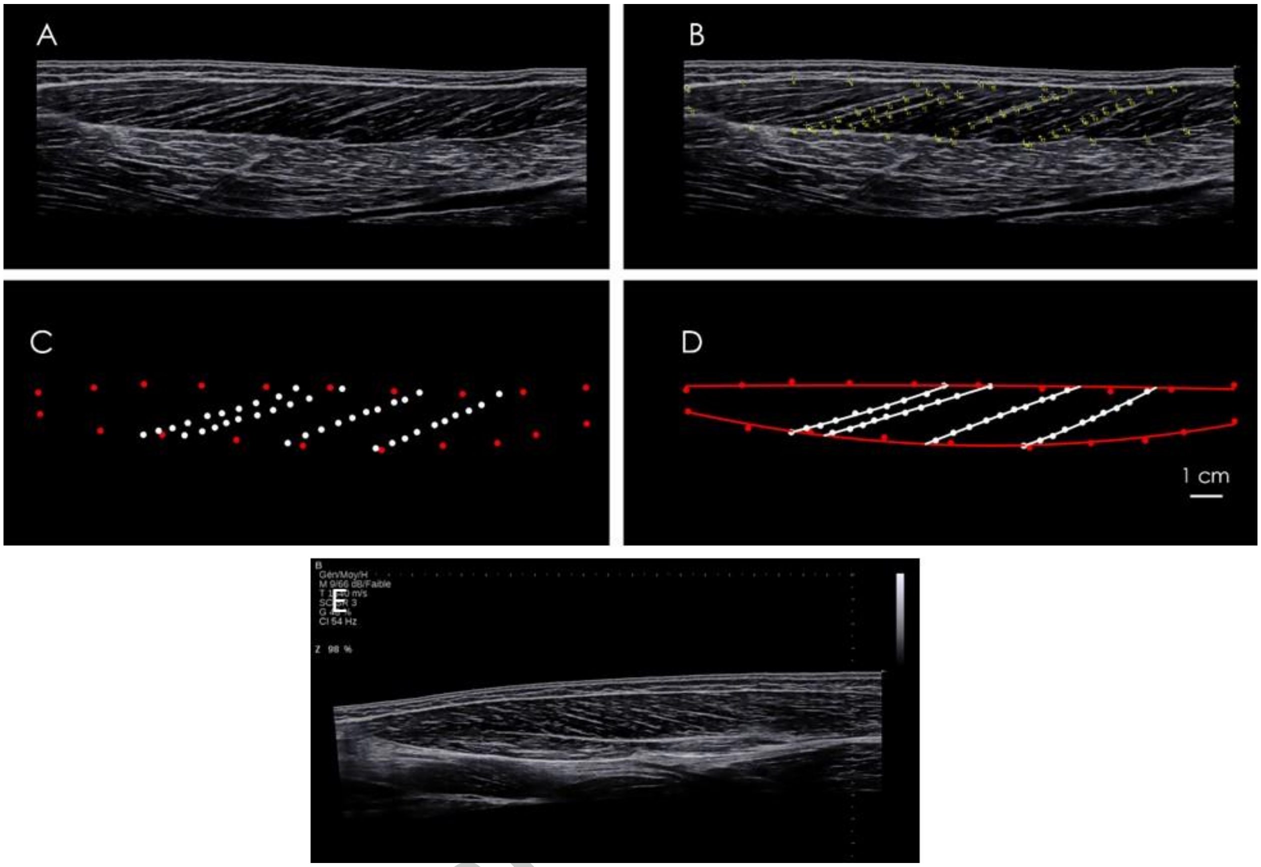

Figure 1. Typical example of the methods employed to extract muscle architecture from a ultrasound extended field of view scan of the semimembranosus muscle (A). Superficial and deep aponeuroses, and clearly visible fascicles (between 3 to 5 for each scan, 4 in the current example) are first labelled over 10 points from the ultrasound image using Image J software (B). The x-y coordinates of the label dots are recorded (C). The coordinates of the two aponeuroses and fascicles are then fitted using a second-degree polynomial order function to finally compute the fascicle length and pennation angle (D). Fascicle length was computed as the sum of the distance between each point of the fit. Pennation angle was computed as the average of the pennation angle computed at 25, 50, 75 and 100% of the fascicle length between the two aponeuroses. (E). An example of an image captured that was of slightly lower quality. The lower aponeurosis is more difficult to identify towards the proximal and distal ends of the image.

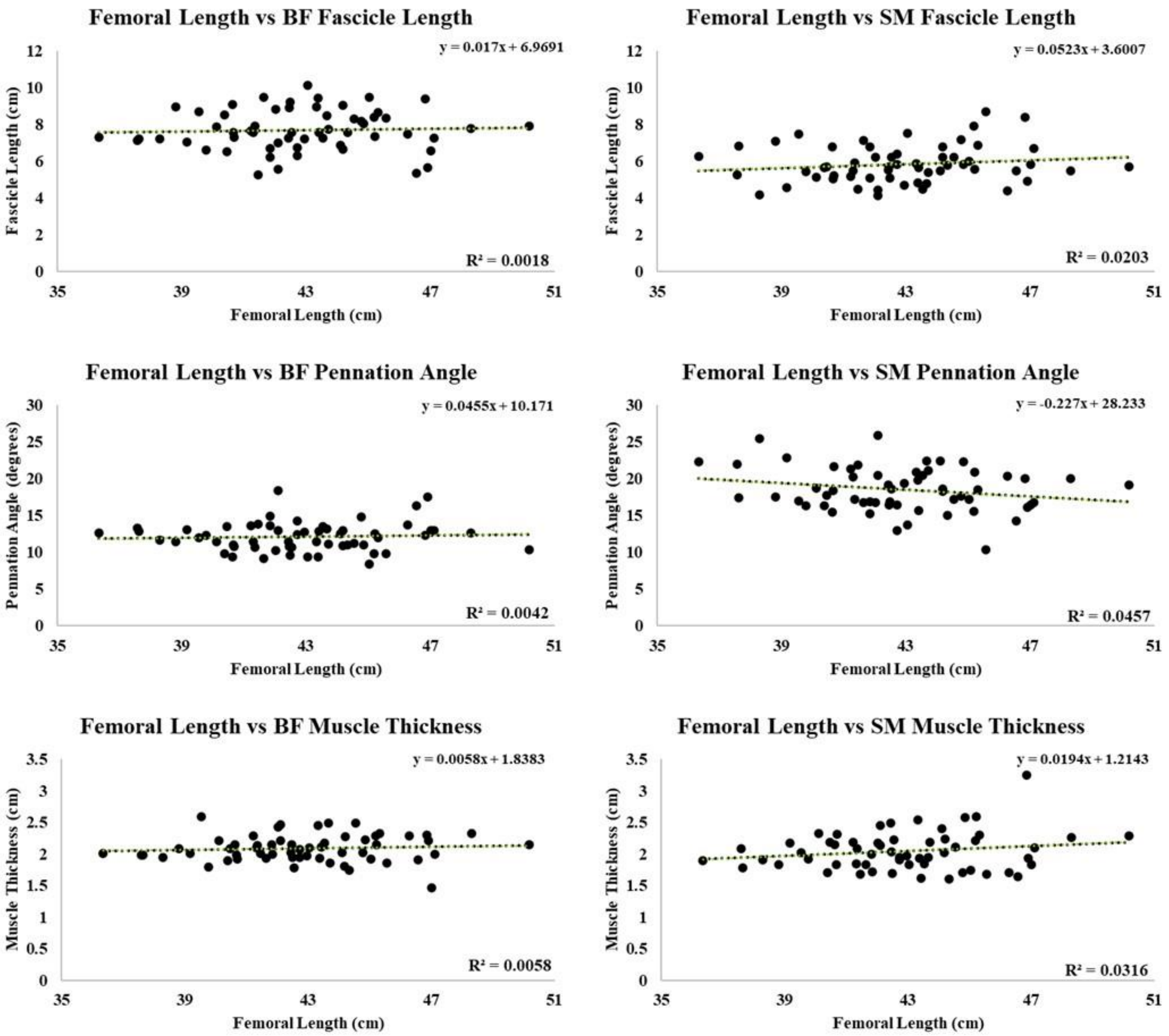

Figure 2. Linear regression graphs showing the relationships between the femoral length of the participants compared to muscle architecture measures (fascicle length, pennation angle and muscle thickness) for the *bicep femoris* long head (BFlh) and *Semimembranosus* (SM) muscles.

Tables

	Mean±SD			ANOVA	Effect Size±90%CI; Likelihood (%)					
Fascicle Length	U16	U17	U19	*p*-value	U16 vs U17		U16 vs U19		U17 vs U19	
Bicep Femoris (cm)	7.79 ± 0.98	7.73 ± 1.20	7.72 ± 1.22	0.98	-0.08 ± 0.42	13/56/31	-0.18 ± 0.52	11/42/47	-0.10 ± 0.63	21/40/39
Semimembranosus (cm)	5.93 ± 1.20	5.81 ± 1.04	5.83 ± 0.92	0.94	-0.08 ± 0.58	21/43/36	-0.13 ± 0.61	18/40/43	-0.05 ± 0.61	24/42/34
Pennation Angle										
Bicep Femoris (°)	11.71 ± 1.52	12.22 ± 2.15	12.33 ± 2.36	0.62	0.21 ± 0.40	52/43/4	0.38 ± 0.46	74/24/2	0.16 ± 0.66	46/36/17
Semimembranosus (°)	18.27 ± 3.52	19.41 ± 3.14	18.01 ± 2.26	0.29	0.38 ± 0.58	70/25/5	0.06 ± 0.64	35/40/25	-0.32 ± 0.54	6/29/65
Muscle Thickness										
Bicep Femoris (cm)	2.09 ± 0.23	2.07 ± 0.16	2.13 ± 0.30	0.72	-0.05 ± 0.36	12/64/24	0.09 ± 0.64	39/40/22	0.15 ± 0.72	45/34/21
Semimembranosus (cm)	2.03 ± 0.23	2.13 ± 0.39	2.08 ± 0.42	0.72	0.25 ± 0.55	57/35/9	-0.04 ± 0.56	24/46/31	-0.29 ± 0.72	13/29/58

Table 1. Comparison between muscle architecture measures (fascicle length, pennation angle and muscle thickness) of different age groups; Analysis of variance (ANOVA), effect size ± 90% CI and the likelihood to obtain a meaningful change. Magnitude based decisions (Likelihood) is reported as the percentage probability that the observed differences are negative/trivial/positive compared to the measure by which the comparison is being made.

	Mean±SD		ANOVA	PHV vs Post-PHV	
Fascicle Length	PHV	Post-PHV	*p*-value	Effect Size±90%CI	Likelihood (%)
Bicep Femoris (cm)	7.61 ± 0.77	7.72 ± 1.20	0.74	0.29 ± 0.70	59/29/12
Semimembranosus (cm)	5.85 ± 1.12	5.84 ± 1.01	0.99	-0.02 ± 0.55	25/47/28
Pennation Angle					
Bicep Femoris (°)	11.53 ± 1.66	12.29 ± 2.03	0.22	0.31 ± 0.78	60/27/13
Semimembranosus (°)	18.33 ± 3.77	18.55 ± 2.74	0.82	0.07 ± 0.65	37/40/23
Muscle Thickness					
Bicep Femoris (cm)	2.01 ± 0.14	2.11 ± 0.23	0.12	0.72 ± 0.49	96/4/0
Semimembranosus (cm)	1.96 ± 0.22	2.07 ± 0.32	0.25	0.16 ± 0.67	46/36/18

Table 2. Comparison between muscle architecture measures (fascicle length, pennation angle and muscle thickness) of different maturity offset groups; Analysis of variance (ANOVA), effect size ± 90% CI and the likelihood to obtain a meaningful change. Magnitude based decisions (Likelihood) is reported as the percentage probability that the observed differences are negative/trivial/positive compared to the measure by which the comparison is being made.

		p - value				
Fascicle Length	R-squared	Body Mass	Femoral Length	Height	Leg Length	Age
Bicep Femoris	0.119	0.359	0.033*	0.856	0.012*	0.843
Semimembranosus	0.101	0.318	0.561	0.418	0.409	0.395
Pennation Angle						
Bicep Femoris	0.053	0.291	0.109	0.435	0.104	0.614
Semimembranosus	0.341	<0.001*	0.025*	0.053	0.754	0.623
Muscle Thickness						
Bicep Femoris	0.244	0.001*	0.971	0.942	0.543	0.099
Semimembranosus	0.376	<0.001*	0.342	0.54	0.637	0.037*

Table 3. Stepwise regression results to identify relationships between anthropometric measures (body mass, femoral length, height, leg length and age) and muscle architecture measures (fascicle length, pennation angle and muscle thickness) for the *bicep femoris* long head (BFlh) and *semimembranosus* (SM) muscles.

		p - values								
	R-squared	Body Mass	Age	Height	BFlh FL	SM FL	BFlh PA	SM PA	BFlh MT	SM MT
Knee-flexor eccentric Strength (N)	0.316	0.001*	0.369	0.079	0.082	0.321	0.088	0.873	0.135	0.084

Table 4. Stepwise regression results to identify relationships between knee-flexor eccentric strength and both anthropometric (body mass, age, height) and muscle architecture measures (BFlh = *bicep femoris* long head, SM = *semimembranosus*, FL = fascicle length, PA = pennation angle, MT = muscle thickness).