- 1 **TITLE:**
- 2 THE INFLUENCE OF MATURITY STATUS ON MUSCLE ARCHITECTURE IN
- 3 SCHOOL-AGED BOYS
- 4
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30 ABSTRACT

31 Purpose: To determine the differences in muscle architecture of the lower limb in pre-, circaand post-peak height velocity (PHV) boys. Methods: Muscle architecture variables from both 32 33 the gastrocnemius medialis (GM) and vastus lateralis (VL) were derived from ultrasonographic 34 images in 126 school-boys. One-way ANOVA's using Bonferonni post-hoc comparisons were 35 employed to determine between-group differences and effect sizes were calculated to establish 36 the magnitude of these differences. Results: All muscle architecture variables showed significant small to large increases from pre- to post-PHV, excluding GM fascicle length (d =37 38 0.59 - 1.39; p < 0.05). More discrete between-group differences were evident as GM and VL 39 muscle thickness and physiological thickness significantly increased between pre- and circa-40 PHV (d > 0.57; p < 0.05); however only the VL muscle thickness and physiological thickness 41 increased from circa- to post-PHV (d = 0.68; p < 0.05). The post-PHV group also showed larger 42 GM pennation angles than the circa-PHV group (d = 0.59; p < 0.05). Conclusion: The 43 combined results show that maturation is associated with changes in muscle morphology. 44 These data quantify the maturity-related changes in muscle architecture variables provide a 45 reference to differentiate between training induced adaptations versus changes associated with 46 normal growth and maturation.

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#### 55 INTRODUCTION

56 Notable disparities in anatomy and physiology exist between children and adults, and there are 57 also clear differentiations between children and adolescents, mediated by growth and 58 maturation (23, 35). Force-producing capacities are lower in children when compared to 59 adolescents or adults, partly as a result of the architecture and size of the muscle (18, 20, 30) and specific muscle activation patterns (9). Throughout maturation, the neuromuscular system 60 61 develops in a non-linear manner, resulting in large variations in physical performances of children of similar chronological age (26). It is known that maturity leads to increased body 62 63 mass and fat free mass (23), however, the specific structural adaptations that occur naturally 64 throughout growth and maturation are yet to be fully understood. Very few studies have specifically examined how muscle size changes throughout childhood and adolescence, and 65 66 how the maturation process may result in specific architectural changes. Considering that 67 muscle architecture is an important factor when it comes to determining a muscle's function and influences both force production and contraction velocity (21), examining maturity-related 68 69 developmental changes in muscle architecture is warranted.

70 Muscle architecture is typically characterised by parameters such as fascicle length, 71 pennation angle and cross-sectional area (5). Despite fascicular arrangements significantly 72 influencing a muscle function (7), particularly the fascicle's force-length and force-velocity 73 characteristics (36), age- and maturity-related comparisons of muscle architecture measures 74 between adults, adolescents, and children are sparse. Research suggests that adults demonstrate 75 greater muscle thickness than children (19, 29) while older adolescents exhibit greater muscle 76 thickness relative to their younger peers (17, 18). Men have been shown to have longer fascicles 77 than boys, and this remains the same for fascicle length when made relative to thigh length 78 (20). Furthermore, 15-year-old adolescents have been shown to possess significantly longer 79 muscle fascicles than children, but do not differ from adults (17), implying that fascicle length 80 reaches adult levels at or before the age of 15 years. Pennation angle of the knee extensor 81 muscles seems to remain consistent from childhood through to adulthood (17), whereas the 82 pennation angle of the gastrocnemius medialis has been reported to increase from birth before 83 becoming stable following the adolescent growth spurt (3, 14). However, maturational status 84 was not quantified in these studies, and the developmental effects on pennation angle throughout maturation is still unknown. The existing literature has focused on changes based 85 86 on chronological age as opposed to grouping according to maturity status. Therefore, the manner in which muscle architecture changes throughout the non-linear process of 87 88 maturational development remains unclear.

89 Literature has demonstrated that individuals of the same chronological age can differ markedly with respect to biological maturity (1). Furthermore, significant inter-individual 90 91 variation exists for the level (magnitude of change), timing (onset of change), and tempo (rate 92 of change) of maturation. Therefore, it is important to quantify muscle architecture in relation 93 to maturity status as opposed to chronological age. Understanding how muscle architecture 94 variables differ between maturity groups would better enable researchers to distinguish 95 between changes caused by growth and maturation versus those mediated by physical training. 96 Additionally, the majority of previous studies have compared a single age group during childhood versus adults and given that maturation is non-linear, the comparison of two discrete 97 98 age groups does not highlight how muscle architecture changes throughout the fluctuating 99 process of maturation (17, 20, 30). Finally, it should be noted that the majority of previous 100 studies have not included children coincident with peak height velocity (PHV) which, due to 101 the rapid gains in height and mass, is regarded as a unique stage of development within 102 paediatric populations and can have significant effects on muscle architecture due to the 103 differential lengthening of bone and muscle during this period.

Therefore, the aim of the current study was to investigate differences in muscle thickness, pennation angle, and fascicle length of vastus lateralis and gastrocnemius medialis of pre-, circa- and post-PHV boys. We hypothesised that muscle thickness, pennation angle, and fascicle length would increase across maturity groups, with the largest differences evident between the circa- and post-PHV groups.

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## 110 METHOD

#### 111 **Participants**

112 One hundred and eighty-three male secondary school children in the United Kingdom 113 volunteered to participate in the study. Sample size was estimated a priori using statistical 114 software (G\*Power v 3.1) for the analyses of variance test, considering an effect size of 0.4, 115 significance level of 0.05, and statistical power of 0.95, for the primary outcome variables of 116 muscle thickness, pennation angle and fascicle length, a sample size of 102 was needed. All 117 participants were free from lower extremity injury and were involved in regular sport and 118 physical education-based activity programs, inclusive of strength and conditioning and games-119 based activities. Participants were asked to refrain from strenuous exercise at least 48 hours prior to testing. Written parental consent and participant's assent were then obtained for 120 participants under 18 years prior to involvement in the study, while ethical approval was 121 122 granted by the Cardiff School of Sport Research Ethics Committee.

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# 124 **Procedures**

125 Anthropometrics: Standing height (centimetres) and seated height (centimetres) were measured 126 using a stadiometer (SECA 321, Vogel and Halke, Germany), and body mass (kilograms) was 127 measured using a balance beam scale (SECA 770, Vogel and Halke, Germany). Biological 128 maturity was then determined using years from PHV (28). Maturity offset values were 129 calculated using the sex-specific equation for boys: -[9.236 + 0.0002708\*Leg Length and 130 Sitting Height interaction]-[0.001663\*Age and Leg Length interaction] + [0.007216 Age and 131 Sitting Height interaction] +  $[0.02292 \cdot \text{mass}$  by Height ratio]. Due to the error in the prediction 132 equation of approximately 6 months for boys (16), participants with a maturity offset of -1 to -0.5 years, and +0.5 to +1 years were removed from the study. Additionally, participants whose 133 134 maturity offset was outside -3 or +3 years were also removed from the study to maximise 135 accuracy. These selection criteria resulted in 57 participants being removed from the study. 136 The final sample size was 126 subjects, which was still above the required number from the 137 sample size analysis. This robust approach enabled the identification of three distinct maturity 138 groups: pre-PHV group (maturity offset of < -1), circa-PHV (maturity offset between -0.5 -139 0.5), and post-PHV group (maturity offset of >1). Descriptive statistics for each age group are 140 presented in *table 1*.

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## \*\*\*\*\*Table 1 inserted here \*\*\*\*\*

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144 Muscle architecture: Muscle structure of the gastrocnemius medialis (GM) and vastus lateralis 145 (VL) were measured with real time B-Mode ultrasonography (Vivid E9, GE Healthcare, Chalfont St Giles, UK) with a 45-mm linear array probe. Water soluble gel was applied to both 146 147 the ultrasound probe and the participants' skin for acoustic coupling, to enhance the contrast 148 of the images, and to avoid depression of the dermal layer. To measure the GM, participants 149 laid prone on a massage couch with the hip angle at approximately  $180^{\circ}$  relative to the trunk, 150 the knee set at approximately 180° relative to the thigh, and ankle positioned at approximately 151 90° relative to the shank. For the imaging of the VL, participants lay supine with the hip 152 positioned at 90° relative to the trunk, and legs fully extended. For both muscles, the ultrasound 153 probe was placed perpendicular to the skin, and the scanning surface was orientated until it was

154	positioned parallel to the muscle to collect sagittal plane images of each subject's dominant
155	leg. For the VL muscle, the image was taken at 50% of the distance between the greater
156	trochanter and the lateral epicondyle of the femur (33) and the GM image was taken at 30% of
157	the distance from the popliteal crease to the centre of the lateral malleolus (18). The subsequent
158	analysis was carried out using open-source image analysis software (Image J, National Institute
159	of Health, Bethesda, MD, USA) to calculate pennation angle, fascicle length and muscle
160	thickness. Figure 1 depicts how the muscle architecture characteristics were calculated. Muscle
161	thickness was measured as the distance from the superficial aponeuroses to the deep
162	aponeuroses. The thickness of the proximal, distal and middle of the muscle belly was assessed
163	in the image, and the average of these sites used for further analysis. Physiological muscle
164	thickness was calculated by the following equation: (muscle thickness <sup>2</sup> + [tan x pennation angle
165	x muscle thickness] <sup>2</sup> ) <sup>0.5</sup> (6). Pennation angle ( $\Theta_p$ ) was defined as the angle between the muscle
166	fascicles and the deep aponeuroses. As the entire fascicle was not visible in the ultrasound
167	image, it was calculated from the equation: fascicle length = muscle thickness (sin $\Theta_p$ ) <sup>-1</sup> (6).
168	Each ultrasound image was assessed on three occasions, with the average value for each
169	architectural variable used for analysis.
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171	*****Figure 1 inserted here *****
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173	Figure 1. Ultrasound image of the gastrocnemius medialis, highlighting how the muscle
174	architecture variables were calculated. MT: muscle thickness; PA: pennation angle;
175	FL: fascicle length
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178	Reliability

179 To establish intra-rater reliability of the ultrasonographic measurements, a sub-group of thirty-180 four school aged boys (14 = pre-PHV, 20 = post-PHV) attended a test session, where three 181 images were taken for each muscle. Subsequently, two levels of reliability were quantified for 182 each muscle architecture variable. *Within-image reliability* reflected the reliability of digitising an image and was assessed using three measurements of each variable in the final image taken 183 184 during the test session. Between-image reliability determined the reliability of taking accurate 185 images of the same muscle site, and was assessed by calculating an average measure from the three trials of each muscle architecture variable from each of the three images within the test 186 187 session.

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## 189 Statistical Analysis

190 Within-image and between-image reliability for each variable was determined using mean 191 coefficients of variation (CV%), intraclass correlation coefficients (ICC) and the change in 192 mean. Magnitudes of ICC were classified according to the following thresholds: Poor < 0.49; 193 Moderate 0.5 - 0.74; Good 0.75 - 0.89; Excellent > 0.9 [18]. All reliability statistics were 194 calculated using an online spreadsheet run through Microsoft Excel for Mac version 16.3 (11).

195 Descriptive statistics (means  $\pm$  standard deviations) were calculated for all muscle architecture variables for each maturity group. Differences in muscle architecture variables 196 197 between maturity groups and across both muscles were assessed using a 3 x 2 (maturity group 198 x muscle) mixed model ANOVA. Homogeneity of variance was assessed via Levene's statistic 199 and where violated, Welch's adjustment was used to correct the F-ratio. Post-Hoc analysis was 200 used to identify the groups that were significantly different to one another using either 201 Bonferroni or Games-Howell post-hoc analysis, where equal variances were and were not 202 assumed, respectively. Cohen's d effect sizes were also calculated, using pooled standard 203 deviations to establish the magnitude of any between-group differences using the following classifications: : trivial < 0.19; small 0.2 – 0.59; moderate 0.6 – 1.19; large 1.2 – 1.9; very large 205 2.0 – 3.9; > extremely large > 4.0 (12). The strength of relationships between muscle 206 architecture variables in the whole sample, and when sub-divided by maturity group (pre-PHV 207 *vs* circa-PHV *vs* post-PHV) were assessed using Pearson's correlation coefficient, and 208 interpreted as: < 0.2 no relationship; 0.2 – 0. 45 weak; 0.45 – 0.7 moderate; > 0.7 strong based 209 on previous recommendations (31). All significance values were accepted at  $p \le 0.05$  and all 210 statistical procedures were conducted using SPSS v.23 for Macintosh.

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## 212 RESULTS

# 213 Reliability of ultrasonography

Data for all architectural variables in both muscle sites showed excellent ICC and acceptable CV% for within-image reliability in both maturity groups (pre-PHV: ICC = 0.87 - 1.00; CV = 0.9 - 7.1%, post-PHV: ICC = 0.92 - 1.00; 0.6 - 3.1%). Apart from VL fascicle length in the pre-PHV cohort, all architectural variables ICC's for between-images indicated moderate to excellent reliability and acceptable CV % for both muscle sites in the pre-PHV (ICC = 0.59 - 0.97; CV = 3.3 - 11.1%) and post-PHV groups (ICC = 0.75 - 0.95; CV = 2.9 - 5.9%).

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#### 221 Between-group differences in muscle architecture

Muscle architecture variables for pre-, circa- and post-PHV groups are displayed in *figure 2ae*. For GM muscle thickness and physiological thickness, there was a moderate, significant difference between the pre- and post-PHV groups (d = 1.19 and 1.12; p < 0.05), respectively); and also small to moderate, significant differences between the pre- and circa-PHV cohorts (d= 0.96 and 0.57; p < 0.05, respectively). There were moderate, significant differences for GM pennation angle between the post-PHV and both the pre- and circa-PHV groups (d = 0.94 and 0.59, respectively; p < 0.05). However, there was no significant difference in GM fascicle 229 length between any groups. For VL muscle thickness and physiological thickness there were 230 moderate to large, significant differences between the pre- and post-PHV groups, pre- and 231 circa-PHV, and circa- to post-PHV cohorts (d = 0.68 - 1.40, respectively). For both VL 232 pennation angle and fascicle length, there were significant differences between the pre-PHV 233 and the post-PHV group (d = 0.59 and 0.66, respectively; p < 0.05), but no differences between 234 any other groups. When made relative to leg length, there was no difference between groups 235 for fascicle length for either GM or VL muscles.

With regards to differences in architectural variables between muscles, both VL muscle thickness and fascicle length were significantly greater than GM muscle thickness and fascicle length (p < 0.001) in the pre-, circa- and post-PHV groups. Contrastingly, for all maturity groups, GM pennation angle was significantly greater than VL pennation angle (p < 0.001).

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#### \*\*\*\*\*Figure 2 a-e inserted here \*\*\*\*\*

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Figure 2. Differences between pre-, circa-, and post-PHV groups in a) physiological muscle
thickness, b) muscle thickness, c) pennation angle, d) fascicle length, and e) relative
fascicle length

<sup>\*</sup>significant difference between groups (p < 0.05). d = Cohen's effect size.

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For both the GM and VL muscle, data showed significant, weak correlations between thickness and pennation angle for the whole group, and the pre- and post-PHV group (r = 0.28-0.43; p < 0.05). Additionally, the relationship between both GM and VL muscle thickness and fascicle length showed weak to strong, significant relationships across the whole group, and the three maturity groups (r = 0.37 - 0.76; p < 0.05). Finally, there were significant, moderate to strong, negative correlations between GM and VL pennation angle and fascicle length in all the groups (r = -0.51 to -0.79; p < 0.05).

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## 256 DISCUSSION

The initial hypothesis can be accepted, as the results showed an increase in GM and VL muscle 257 258 thickness, GM and VL pennation angle, and VL fascicle length from pre- to post-PHV. 259 However, the larger differences in muscle architecture variables were seen between the pre-260 and circa- PHV groups. In light of existing literature, this appears to be the first study to 261 compare muscle architecture variables across all maturity stages in in a sample of male youth. 262 While previous literature has compared circa- and post-PHV groups with adults, this is the first study to examine a pre-PHV cohort. The main findings of the current study showed that all 263 264 muscle architecture variables showed significant small to large increases from pre- to post-265 PHV with the exception of GM fascicle length. More discrete between-group differences were evident as GM and VL muscle thickness and physiological thickness significantly increased 266 267 between pre- and circa-PHV; however only the VL muscle thickness and physiological 268 thickness increased from circa- to post-PHV. The post-PHV group also showed larger GM 269 pennation angles than the circa-PHV group. Finally, for both the VL pennation angle and 270 fascicle length, there were small to moderate, significant differences between the pre-PHV and 271 post-PHV groups, but no differences between any other groups.

The current study showed that GM muscle thickness increases from pre-PHV to both circa-PHV and post-PHV. For both maturity groups, these changes in muscle thickness were larger than the typical error (CV%), and are therefore considered as meaningful changes likely due to maturation as opposed to merely reflecting noise in the measurement. The nonsignificant change between circa- and post-PHV groups would indicate that natural developments in GM muscle thickness may have started to stabilise around the time of PHV. 278 Growth of the muscle throughout maturation may occur due to increases in body mass 279 potentially through increases in skeletal mass, and internal organs intensifying the mechanical load on the skeletal system during everyday tasks (5). Additionally, changes in stature will 280 281 enhance muscle growth as the load or stretch applied to muscles during bone growth may act as a stimulus for increases in muscle size (37). Previous studies, albeit examining age-related 282 283 differences, have shown significant increases in GM muscle thickness between elementary 284 school children, junior school children, and adults (18). The differences between adults and 285 junior school children suggest that there may be further development of muscle size during 286 ageing, beyond that of adolescence. This could be due to large changes in activity profile and 287 body mass during adulthood driving increases in mechanical loading that underpin the mechanism of muscle growth (5). Data from Binzoni and colleagues (3) showed approximate 288 289 increases in GM muscle thickness of 7% per year, which was then confirmed in an empirical 290 study with children between the ages of 5 -12 years (2). Expressing results from the current 291 study by chronological age as opposed to maturity, shows an approximate 9% increase in GM 292 muscle thickness per year between 12 years to 14 years, but only a 5% increase per year 293 between 14 to 16 years. Cumulatively, these results support the notion of a slower rate of 294 growth in the GM muscle following PHV, which may infer that further maturity-related increases in lower limb strength (27) may be due to growth of different muscles or ongoing 295 296 changes in neural properties (9). For example, as children mature they have a greater potential 297 to produce force, as there is a reduced agonist- antagonist co-contraction, a greater ability to 298 recruit high-threshold type II motor units, and greater muscle activation rates (10).

299 Pennation angle in the GM showed a moderate, significant increase between pre- and 300 post-PHV, and a small, significant increase between circa- to post-PHV. The circa- to post-301 PHV changes were larger than the typical error (CV%), and can be considered as meaningful 302 changes; however, the change from pre- to circa-PHV was only greater than the smallest303 worthwhile change (SWC) and not larger than the typical error, so should be viewed with 304 caution. These changes suggest that larger changes in pennation angle occur following PHV, 305 which may be explained by the nature of muscle architecture adaptation. The current study 306 found significant, small relationships between GM muscle thickness and pennation angle in 307 the whole cohort, as well as in the pre- and post-PHV groups. A greater muscle thickness would 308 necessitate a larger pennation angle, and therefore pennation angle increases may happen in 309 response to a muscle increasing in size (7). This may explain the significant increases in GM 310 muscle thickness between pre- to circa-PHV and then the subsequent increase in GM pennation 311 angle between circa- and post-PHV, where changes in muscle size manifest prior to alterations 312 in pennation angle. Comparable results between younger children have been reported in 313 previous literature, where pennation angle has been shown to be similar between children aged 314 between 5-12 years (2). The current study found significantly greater GM pennation angle in 315 the post-PHV than in either the pre- or circa-PHV groups. The lack of difference between the 316 pre- and circa-PHV group suggest that GM pennation angle occurs later in maturation, 317 potentially following the preceding growth in muscle thickness. The larger pennation angle as 318 a result of maturation would likely improve the force-generating capabilities of a muscle (21), 319 through an increase in PCSA and muscle gearing (36), which would ultimately increase 320 explosive-type exercise commonly reported throughout maturation (22, 25, 35).

Muscle thickness in the VL significantly increased from pre- to circa-PHV and circato post-PHV, with between-group differences exceeding the typical error (CV%). This continuous increase in VL muscle thickness throughout maturation coincides with the significant increase in body mass reported between the three maturity groups, again suggesting the importance of mechanical loading on muscle growth. Interestingly, the increases in muscle thickness of the VL across the different maturity groups differ to the growth pattern of the GM, where changes between circa- to post-PHV were not significant. More growth occurs in the femur compared to the tibia during childhood and adolescence (13), where the tibia may slow in growth around the time of PHV, and the femur continues to grow further into adolescence. Considering that growth in skeletal structures provides a stimulus for adaptation of muscle tissue (37), the extended growth typically seen in the femur may explain the increases in VL muscle thickness between circa- and post-PHV that are not seen in the GM. Similar increases in muscle size for the VL have been reported between children and adults (20, 30), but these findings do not offer insights into the complex and fluctuating process of maturation.

Pennation angle and fascicle length of the VL increased significantly between pre- to 335 336 post-PHV, and while all were above the SWC, only the change in VL fascicle length was above 337 the typical error. Similar findings related to fascicle length have been reported in previous literature, whereby men were shown to have longer VL fascicles than boys (30). However, in 338 339 terms of VL pennation angle, findings from the current study differ to previous research, which 340 reported that pennation angle of the VL remained relatively stable between boys aged 11 years, 341 to 14 years and into adulthood (17), and that boys of approximately 9 years of age have similar 342 VL fascicle lengths to adults (30). The discrepancy in results between the current study and 343 previous research may be due to smaller sample sizes in the previous research. Intuitively, 344 greater increases in muscle thickness compared to pennation angle would require an increase in fascicle length. This concept is supported by the current study, where a large negative 345 346 relationship was observed between pennation angle and fascicle length in both muscles. 347 Therefore, it can be assumed from this study that the increases in VL thickness are greater than 348 pennation angle, resulting in increased VL fascicle length. However, it can then be assumed 349 that the increase in thickness and pennation angle are similar in the GM, resulting in no 350 difference in fascicle length between the three groups.

The cumulative findings of the current study indicate that maturation may trigger architectural adaptations within the GM and VL muscles that are likely to facilitate greater 353 force production and rate-of-force-development. Considering that muscle size is a major 354 predictor of maximum strength and power in children (34), the increases in muscle size with 355 maturation could be a major factor contributing to the improved capacity to produce force as 356 children transition through adolescence. Furthermore, considering that the VL muscle plays a major role in sprinting and jumping tasks (8), the increase in the thickness of this muscle may 357 358 be a key factor in the improvement in the physical performance measures reported during 359 childhood and adolescence (22, 25, 35). The larger pennation angle demonstrated by the more 360 mature participants would allow the muscle to produce greater force, as a larger pennation 361 angle will increase the physiological cross-sectional area, resulting in a greater number of 362 contractile elements attaching to the aponeurosis or tendon for a greater force transfer (15). The increase in pennation would also allow a muscle to take advantage of the "gearing" effect 363 364 during movement (36), which permits that muscle fascicles do not need to shorten to the same 365 extent as the overall muscle, permitting slower fascicle velocities in relation to whole muscle velocities (36). This enables the fascicles to operate on a more optimal region of the force-366 367 velocity curve and work at a favourable region of the force-length curve over a longer period, 368 maximising the force that the muscle can develop, without impacting on the capacity for rapid 369 movement production. Finally, the increased fascicle length due to the maturation process 370 would result in a greater muscle shortening velocity (21). The longer fascicles seen in the more 371 mature subjects would have a greater ability to produce force at higher velocities and over larger length ranges, as more sarcomeres in series results in greater cumulative length change 372 373 of a fascicle within a given time. The increased number of sarcomeres in series may also result 374 in each individual sarcomere operating at a lower relative velocity, meaning greater force can 375 be produced by each fascicle (7).

To our knowledge, this is also the first report on the between-image and within-image reliability of GM and VL architectural properties attained from ultrasound images recorded at 378 rest in pre- and post-PHV groups. Data indicated that the muscle architecture variables could 379 be reliably digitised at both within-image and between-image levels, with large to nearly 380 perfect ICC and CV values below the acceptance threshold (<10%). While reliability data on 381 ultrasonography-derived measures of muscle architecture remain scarce in youth populations, 382 when compared to results from a cohort of strength trained males (24), data from the current 383 study showed that the reliability of the digitising process both within- and between-images was 384 more varied in the pre-PHV cohort, and similar in the post-PHV cohort. The increased 385 variability in the pre-PHV cohort compared with older populations may be due to a larger fat 386 mass seen in this population (23). Adipose tissue has less degrees of reflection than muscle due 387 to different acoustic impedance (32). The greater fat mass seen in the younger children may weaken the generated reflection and reduce the quality of the ultrasound image (32), causing 388 389 more variance in the digitisation and determination of muscle architectural variables.

390 The current study has made a novel contribution to the developmental literature 391 surrounding paediatric muscle architecture. However, it is important to consider certain 392 limitations within the study. Considering the dimensions of the linear probe being used, it was 393 necessary to estimate fascicle length from measures of muscle thickness and pennation angle 394 using a previously published equation (6). The model assumes that fascicles are straight and 395 does not account for fascicule curvature, however this equation has been used in previous 396 studies (6) and has been shown to result in an error of only approximately 3% for relaxed 397 muscles with short fascicles (4). Additionally, fascicle length of the GM and VL was made 398 relative to leg length in the current study. However, for more accurate relative measures, the 399 GM fascicle length would need to be made relative to the shank length, and VL fascicle length 400 made relative to thigh length. The current study used a cross-sectional research design; 401 however, future research is now required to validate these findings by longitudinally measuring 402 changes in muscle architecture characteristics in youth of varying maturity levels. Furthermore,

while this study has gone someway into highlighting the differences in muscle architecture
during maturity, the influence that this has on physical performance in tasks such as sprinting
and jumping remains unknown.

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407 CONCLUSION

408 The current study showed that all muscle architecture variables showed significant 409 small to large increases from pre- to post-PHV, with the exception of GM fascicle length. These findings indicate that maturation underlies changes in muscle morphology, and that maturation 410 411 may trigger architectural adaptations within the GM and VL muscles that are likely to facilitate 412 greater force production and rate-of-force-development. This study establishes maturity-413 related changes in muscle architecture variables that provides researchers and practitioners 414 with reference data that can be used to differentiate between training induced adaptations 415 versus those realised as a consequence of growth and maturation. For example, the data showed 416 that an increase in 0.3 cm was observed in VL muscle thickness between pre- to circa-PHV; 417 therefore, any adaptations would need to exceed this value to confirm changes were not 418 attributable solely to the maturational process.

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