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Published version

BALSHAW, Thomas G, MADEN-WILKINSON, Tom, MASSEY, Garry J and FOLLAND, Jonathan P (2021). The Human Muscle Size and Strength Relationship. Effects of Architecture, Muscle Force and Measurement Location. *Medicine & Science in Sports & Exercise*.

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Title page

Title:

THE HUMAN MUSCLE SIZE AND STRENGTH RELATIONSHIP: EFFECTS OF ARCHITECTURE, MUSCLE FORCE AND MEASUREMENT LOCATION

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Abbreviated title for running head:

MUSCLE SIZE AND STRENGTH RELATIONSHIP

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Key words

Magnetic resonance imaging

Quadriceps femoris

Muscle volume

Physiological cross-sectional area

1 **Abstract**

2 **Purpose:** This study aimed to determine the best muscle size index of muscle strength
3 by establishing if incorporating muscle architecture measurements improved the human muscle
4 size-strength relationship. The influence of calculating muscle force, and the location of
5 anatomical cross-sectional area (ACSA) measurements on this relationship were also
6 examined.

7 **Methods:** Fifty-two recreationally active males completed unilateral isometric knee
8 extension strength assessments and MRI scans of the dominant thigh and knee to determine
9 quadriceps femoris (QF) size variables (ACSA along the length of the femur, maximum ACSA
10 [ACSA_{MAX}] and volume [VOL]) and patellar tendon moment arm. Ultrasound images (2 sites
11 per constituent muscle) were analyzed to quantify muscle architecture (fascicle length,
12 pennation angle), and when combined with VOL (from MRI), facilitated calculation of QF
13 effective PCSA (_{EFF}PCSA) as potentially the best muscle size determinant of strength. Muscle
14 force was calculated by dividing maximum voluntary torque (MVT) by the moment arm and
15 addition of antagonist torque (derived from hamstring EMG).

16 **Results:** The associations of _{EFF}PCSA ($r=0.685$), ACSA_{MAX} ($r=0.697$), or VOL
17 ($r=0.773$) with strength did not differ, although qualitatively VOL explained 59.8% of the
18 variance in strength, ~11-13% greater than _{EFF}PCSA or ACSA_{MAX}. All muscle size variables
19 had weaker associations with muscle force than MVT. The association of strength-ACSA at
20 65% of femur length ($r=0.719$) was greater than for ACSA measured between 10-55% and 75-
21 90% ($r=-0.042-0.633$) of femur length.

22 **Conclusions:** In conclusion, using contemporary methods to assess muscle architecture
23 and calculate _{EFF}PCSA did not enhance the muscle strength-size association. For
24 understanding/monitoring muscle size, the major determinant of strength, these findings

- 25 support the assessment of muscle volume, that is independent of architecture measurements,
- 26 and was most highly correlated to strength.

27 **Introduction**

28 Muscular strength, the maximum voluntary torque (MVT) a muscle group can produce,
29 influences the performance of athletic events (1) and functional activities of daily life (2), is a
30 risk factor for muscle injury (3), and is implicated in the development and progression of joint
31 degeneration (i.e. osteoarthritis; (4, 5)). Knowledge of the factors underpinning strength and
32 strength change are important in order to understand, assess/monitor and potentially modify
33 the most important determinants. Whilst it is well known that muscle size is a key determinant
34 of maximum strength (6–8), the most important muscle size determinant of strength is unclear.
35 In fact the relationship of muscle size and strength may be influenced by both muscle size and
36 strength measurements, with little investigation of the inclusion of muscle architecture
37 assessment in the measurement of muscle size, the effect of calculating muscle force as the
38 index of strength, which accounts for joint-level confounders (moment arm and antagonist co-
39 activation), or the influence of muscle size measurement location (relative to segment length).

41 Theoretically, physiological cross-sectional area (PCSA) most accurately reflects the
42 number of sarcomeres/myosin-actin cross-bridges arranged in parallel and thus able to generate
43 tension between the tendons (9), and would be expected to be the strongest determinant of
44 maximum strength (7, 10). However, studies comparing three common measures of muscle
45 size have reported either maximum anatomical cross-sectional area ($ACSA_{MAX}$; (7)), muscle
46 volume (VOL; (11)- elbow extensors), or PCSA ((11)- elbow flexors) to be the best correlate
47 of strength. This inconsistency might be explained by the methodological limitations in the
48 measurement of architecture and thus PCSA within these studies. Whilst $ACSA_{MAX}$ and VOL
49 can be accurately assessed with T1-weighted magnetic resonance imaging (MRI), considered
50 the gold standard technique (12), PCSA cannot be assessed directly by MRI alone. PCSA is
51 typically calculated by dividing VOL (from MRI) by fascicle length (L_F), and further corrected

52 for the loss of force transmission to the aponeurosis/tendon in pennate muscle (i.e. pennation
53 angle, θ_P), sometimes referred to as effective PCSA ($_{EFF}PCSA$). Therefore, PCSA relies on
54 both the determination of VOL (via MRI) and the precise measurements of muscle architecture
55 (L_F and θ_P , via ultrasound).

56

57 Previous studies comparing associations between strength and different muscle size
58 indices (i.e. $ACSA_{MAX}$ vs. PCSA vs. VOL), have used the following muscle architecture (L_F
59 and θ_P) approximations for the calculation of PCSA: estimation from cadaveric data (7);
60 estimation based on muscle length ((11)–elbow flexors); or ultrasonographic measurements at
61 one site in one constituent muscle ((11)–elbow extensors). It is known that muscle architecture
62 varies both between and along constituent muscles of a muscle group (13, 14), therefore
63 performing multiple ultrasonographic measurements along the length of each constituent
64 muscle may better account for such variations. Furthermore, L_F has commonly been assessed,
65 and hence PCSA calculated, with relatively short ultrasound arrays that visualise ~30-50% of
66 L_F (40 mm array (15, 16); 45 mm (17); 60 mm (18)) and consequently, require prediction of
67 the remainder of the fascicular path in order to estimate L_F . This limitation can be minimized
68 by use of a longer ultrasound array, thereby reducing the proportion of overall fascicle length
69 that is extrapolated. Moreover, these issues in the assessment of L_F and θ_P , may provide a
70 methodological explanation for the variable findings as to the best muscle size index of
71 strength. It is conceivable that more careful architecture, and thus also PCSA, measurements,
72 could result in PCSA being the superior determinant of muscle strength and demonstrate the
73 utility of incorporating muscle architecture measurements within the assessment of human
74 muscle size. Finally, muscle thickness assessed with ultrasound imaging is often considered a
75 convenient, but somewhat crude index of muscle size. Perhaps surprisingly the relationship

76 between muscle thickness and strength has not been compared to that of MRI-derived measures
77 of muscle size (ACSA, PCSA and VOL) and strength.

78

79 The theoretical basis for a close association of cross-sectional area measurements
80 (ACSA and PCSA) and muscular strength, as opposed to VOL, is based on the assumption of
81 longitudinal transmission of force along the muscle from the cross-section of greatest area and
82 thus the largest amount of contractile material aligned in parallel. However, lateral force
83 transmission may provide an alternative means of transferring force from intermediate points
84 along muscle fibres (19), allowing a single muscle fibre to act as a series of independent force
85 generators each able to transmit force to the aponeuroses/tendons via radial structural proteins,
86 costameres, the sarcolemma and extra-cellular matrix (20); see Fig. 11 of reference (21).
87 Lateral force transmission might favor a stronger relationship between VOL, which
88 incorporates length, and strength compared to cross-sectional areas.

89

90 The assumption of predominantly longitudinal force transmission also underpins the
91 notion that the best measure of ACSA in relation to strength is the cross-section with the
92 greatest amount of contractile material in parallel (i.e. ACSA_{MAX}, usually calculated as the sum
93 of maximum ACSAs from each constituent muscle, typically occurring at different points
94 along the limb (7, 22, 23)). However, it has been suggested based on ultrasound measurements
95 that for the quadriceps femoris (QF) muscle group relatively proximal ACSA, may be more
96 strongly associated with strength (24) and strength gains after resistance training (25) than mid-
97 muscle measurements. To date there has been no MRI study of ACSA at set intervals along the
98 length of the muscle/bone in relation to strength or how location/length specific ACSA
99 measurements compare to ACSA_{MAX}.

100

101 The association between muscle size variables (i.e. $EFFPCSA$, $ACSA_{MAX}$ and VOL) and
102 joint-level function (i.e. strength/MVT) may be somewhat diluted/confounded by joint
103 neuromechanical factors such as the leverage (moment arm) of the agonist muscles, as well as
104 co-activation and thus opposing torque from the antagonist muscles. It is possible that muscle
105 size variables could be very highly correlated with muscle force, as is the case for isolated
106 animal muscles ($r=0.99$; (8)), and thus explain a greater proportion of the variance in muscle
107 force than is the case for joint-level MVT. However, this has not been investigated.

108

109 Therefore, the aim of this study was to determine the effect of incorporating muscle
110 architecture measurements, calculating muscle force, and $ACSA$ measurement location on the
111 human muscle size-strength relationship. Specifically by comparing: (i) the relationships of
112 four distinct QF muscle size measures ($ACSA_{MAX}$ and VOL from MRI, $EFFPCSA$ from the
113 combination of MRI with ultrasound measurements of architecture, and muscle thickness from
114 ultrasound) with knee extensor strength in a large cohort of healthy young men; (ii) the
115 association of these muscle size measures with muscle force as opposed to joint-level MVT;
116 and (iii) the relationship of strength to $ACSA$, according to the site of $ACSA$ measurements
117 along the femur. We hypothesised that: rigorous muscle architecture measurements (L_F and/or
118 θ_P), at eight sites throughout the QF, in combination with high resolution MRI, would facilitate
119 $EFFPCSA$, rather than $ACSA_{MAX}$ or VOL , being the most powerful determinant of knee
120 extension MVT; that higher associations would be found between muscle size indices with
121 muscle force, than with MVT, once joint-level confounders were accounted for; and that the
122 MVT- $ACSA$ relationship would be influenced by the location of the $ACSA$ measurement along
123 the femur length.

124

125

126 **Materials and Methods**

127 *Participants*

128 Fifty-two young, healthy men (age 25 ± 2 years, height 1.76 ± 0.07 m, body mass $72 \pm$
129 9 kg) free from musculoskeletal injury provided written informed consent prior to participation
130 in this study that was approved by the Loughborough University Ethical Advisory Committee.
131 All participants were recreationally active (2200 ± 1355 metabolic equivalent min/wk; assessed
132 with the short format International Physical Activity Questionnaire; (26)), were not completing
133 any form of systematic physical training, and had not completed lower-body strength training
134 for >18 months. Only male participants were included in order to prevent the potential
135 confounding influence of sex differences in specific tension (force per muscle area: in vivo-
136 (27, 28); and in vitro [i.e. single muscle fibre specific tension]- (29)) on the muscle strength
137 and size relationships investigated in this study.

138

139 *Overview*

140 Participants completed a familiarisation session and two neuromuscular function
141 measurement sessions of their dominant leg, 7-10 days apart at a consistent time of day (starting
142 between 1200 and 1900), and an imaging session (within ± 7 days of the second function
143 measurement session). Familiarisation involved participants completing knee extension and
144 flexion maximum voluntary contractions (MVCs) to become accustomed to these assessments.
145 Function measurement sessions involved assessment of unilateral isometric knee extension
146 strength (i.e. MVT) and antagonist co-activation (hamstrings EMG), as well as knee flexion
147 MVCs for EMG normalization. Musculoskeletal imaging of the dominant limb involved
148 acquisition of magnetic resonance T1-weighted axial plane images (1.5 T) of the QF to assess:
149 ACSA along the length of each constituent muscle, VOL, and in combination with ultrasound-
150 derived muscle architecture measurements, $_{EFF}PCSA$. Ultrasonographic images were recorded

151 at two locations along the length of each constituent muscle of the QF to assess L_F , θ_P , and
152 muscle thickness using a 92-mm wide transducer that was typically able to visualise ~80-90%
153 of total L_F (see Fig. 1A). Sagittal plane MRI scans of the knee were also acquired and analyzed
154 to determine patellar tendon moment arm (PT_{MA}) which was used (along with antagonist EMG
155 during knee extension) to calculate muscle force.

156

157 *Torque recording*

158 Knee extension and flexion torque was recorded whilst participants were seated on a
159 rigid custom-made isometric dynamometer (see Fig. 6B of reference (30)) with knee and hip
160 angles of 115° and 126° (180° = full extension), respectively. This knee joint angle was selected
161 as the angle of peak torque (31). Extraneous bodily movement was minimized by fastening
162 adjustable straps across the pelvis and shoulders. An S-beam strain gauge with a low baseline
163 noise range ($<0.1\%$ MVT; Force Logic, Swallowfield, UK) mounted to the dynamometer was
164 positioned posterior and perpendicular to the tibia and then secured around the participant's
165 leg at ~15% of tibial length (distance from lateral malleolus to knee joint space) above the
166 medial malleolus with an ankle strap (35 mm width reinforced canvas webbing). The analog
167 force signal from the strain gauge was amplified ($\times 370$) and sampled at 2,000 Hz using an
168 external analog-to-digital (A/D) converter (Micro 1401; CED, Cambridge, UK) and recorded
169 with Spike 2 computer software (CED, Cambridge, UK). In offline analysis, force data were
170 low-pass filtered at 500 Hz using a fourth-order, zero-lag Butterworth filter (32), gravity
171 corrected by subtracting baseline force, and multiplied by lever length, the distance from the
172 knee joint space to the center of the ankle strap, to calculate torque values.

173

174

175

176 *EMG recording*

177 Surface EMG was recorded from the medial and lateral hamstring muscles using a
178 wireless EMG system (Trigno; Delsys Inc., Boston, MA). Hamstring EMG measurements were
179 performed to allow estimation of muscle force during knee extension MVT (see “Moment arm
180 and calculation of muscle force” section of methods below). Prior to sensor placement skin
181 preparation (shaving, abrading, and cleansing with 70% ethanol) was conducted. Single
182 differential Trigno Standard EMG sensors (Delsys Inc., Boston, MA; fixed 1-cm interelectrode
183 distance) were then positioned on the medial (semitendinosus and semimembranosus) and
184 lateral (biceps femoris long head) hamstring muscles using adhesive interfaces at 45% of thigh
185 length (above the popliteal fossa). The location of the sensors was determined by palpating the
186 borders of the biceps femoris long head and the medial hamstrings (semitendinosus and
187 semimembranosus) respectively. Each sensor was then placed at 50% of the mediolateral
188 muscle width and parallel to the presumed orientation of the underlying fibres. The isometric
189 dynamometer had a section of the seat removed to accommodate the placement of Hamstring
190 EMG sensors on the skin without compression. EMG signals were amplified at source (x300;
191 20- to 450-Hz bandwidth) before further amplification (overall effective gain, x909), and
192 sampled at 2,000 Hz via the same A/D converter and computer software as the force signal, to
193 enable data synchronization. In offline analysis, EMG signals were corrected for the 48-ms
194 delay inherent to the Trigno EMG system.

195

196 *Knee extension maximum voluntary contractions.*

197 Following a brief warm-up of the dominant leg knee extensors [3 s contractions at 50%
198 (x3), 75% (x3), and 90% (x1) of perceived maximum] participants performed 3-4 MVCs with
199 the instruction to “push as hard as possible” for 3-5 s. MVCs were separated by ≥ 30 s of rest.
200 Biofeedback was provided by placing a horizontal cursor on the torque-time curve with a

201 horizontal cursor indicating the greatest torque obtained within that session, and verbal
202 encouragement was provided during all MVCs. Knee extensor MVT was defined as the
203 greatest instantaneous torque achieved during any MVC during that measurement session.
204 RMS EMG from each of the hamstring sites during knee extension MVT (i.e. over a 500-ms
205 time period, 250-ms either side of instantaneous knee extension MVT) was normalized to that
206 measured during knee flexion MVT (knee flexion EMG_{MAX} ; see below) and then averaged
207 across the two hamstring sites (normalized antagonist HEMG). Knee extension MVT had a
208 between-session within-participant coefficient of variation (calculated as: $[SD \div mean] \times 100$)
209 value of 3.0%.

210

211 *Knee flexion maximum voluntary contractions*

212 Knee flexion MVCs were then performed in the same manner as knee extension MVCs,
213 following prior warm-up contractions. Knee flexion MVT was defined as the greatest
214 instantaneous torque achieved during any MVC during that measurement session. RMS
215 hamstring EMG for a 500-ms epoch at knee flexion MVT (250-ms either side) was analyzed
216 for each site (knee flexion EMG_{MAX}); this approach was intended to capture muscle activity
217 measures during the 500-ms period with the highest mean torque during the plateau phase of
218 the 3-5 s MVC where torque is relatively stable.

219

220 *Fascicle length, pennation angle and muscle thickness*

221 L_F , θ_P and muscle thickness of the constituent QF muscles (vastus lateralis (VL), vastus
222 intermedius (VI) vastus medialis (VM), and rectus femoris (RF)) were measured using a B-
223 mode ultrasonography machine (EUB-8500, Hitachi Medical Systems UK Ltd,
224 Northamptonshire, UK) and a 92-mm, 5-10 MHz linear-array transducer (EUP-L53L) coated
225 with water soluble transmission gel. To match the knee joint angle during MVCs, these images

226 were collected whilst participants sat at rest in the same isometric dynamometer and joint
227 configuration used for maximum strength testing. Images were captured at rest at two sites
228 along the length of each constituent muscle of the QF. Specifically, ultrasound images were
229 recorded at 50% of superficial medio-lateral muscle width at the following locations along the
230 length of the femur from the knee joint space: 30% and 50% of femur length (VI), 50 and 70%
231 of femur length (VL); 20% and 40% of femur length (VM); 55% and 75% of femur length
232 (RF). The locations of these ultrasound recordings were largely adopted from prior research
233 incorporating multiple ultrasound measurements along the length of each constituent muscle
234 of the QF (33). The transducer was positioned parallel to the long axis of the thigh and
235 perpendicular to the skin, the position of the transducer was then subtly adjusted to align with
236 the plane of the fascicles at each site so that an image with the deep and superficial aponeuroses
237 and the trajectory of several fascicles was clearly identifiable with minimal pressure applied to
238 the dermal surface. Video output from the ultrasound machine was transferred to a computer
239 (via an S-video to USB converter) and images recorded using ezcap video capture software.
240 Images were subsequently imported into public domain software and analyzed (Image J, v1.48,
241 National Institutes of Health, Bethesda, USA).

242

243 θ_p at each individual recording site was measured as the angle of insertion of the muscle
244 fascicles into the deep aponeurosis (Fig. 1A), except for the VI where θ_p was measured as the
245 angle between the proximal end of each fascicle and the femur. L_F was measured as the length
246 of the fascicular path between the insertions into the superficial and deep aponeurosis (Fig.
247 1A). When the fascicular path extended beyond the acquired image the missing portion of the
248 fascicle was estimated by extrapolating linearly the fascicular path and the aponeurosis (18,
249 34). L_F and θ_p at each measurement site were taken as the mean of three individual fascicles.
250 L_F and θ_p of each constituent muscle was averaged across the two measurement sites of that

251 muscle. Overall QF L_F and θ_P were calculated as the mean of the four constituent muscles.
252 Muscle thickness at each measurement site (i.e. 2 per muscle) was quantified as the mean of
253 the distance between the deep and superficial aponeurosis at each end, and the middle of the
254 image before being averaged across sites within each muscle (Fig. 1A). Finally, the muscle
255 thickness for each constituent muscle was summed to quantify overall QF muscle thickness.

256

257 *MRI scan and analysis procedures for muscle size and patellar tendon moment arm*

258 **Muscle size variables.** Participants reported for their MRI scan (1.5 T Signa HDxt,
259 GE) having not completed any strenuous physical activity in ≥ 36 h, and had received prior
260 instruction to arrive in a relaxed state having eaten and drunk normally. Upon arrival
261 participants sat quietly for 15 min prior to their MRI scan. Participants lay resting supine at a
262 knee joint angle of 163° (180° = full extension) whilst MR imaging was conducted. A receiver
263 8-channel whole body coil allowed axial T1-weighted images (time of repetition/time to echo
264 550/14, image matrix 512 x 512, field of view 260 x 260 mm, pixel size 0.508 x 0.508 mm,
265 slice thickness 5 mm, inter-slice gap 0 mm) of the dominant leg to be acquired from the anterior
266 superior iliac spine to the knee joint space in two overlapping blocks. Oil filled capsules placed
267 on the lateral side of the thigh allowed alignment of the blocks during analysis (OsiriX
268 software, Version 6.0, Pixmeo, Geneva, Switzerland). The constituent muscles (VM, VL, VI,
269 and RF) were manually segmented in every third image (i.e. every 15 mm; Fig. 1B) starting
270 from the most proximal image in which the muscle appeared. The number of images (slices)
271 manually segmented along the length of each constituent muscle was (mean \pm SD): VL, $24 \pm$
272 2 (range 21-28); VI, 24 ± 1 (range 21-28); VM, 23 ± 2 (range 20-26); RF, 22 ± 1 (range 20-
273 25).

274

275 The ACSA values measured along the length of each muscle were expressed relative
276 to femur length (defined by the number of axial slices between the proximal greater trochanter
277 (100% femur length) and the knee joint space (0% femur length)). Cubic spline interpolation
278 (1000 point; GraphPad Prism 6; GraphPad Software) was then used to quantify ACSAs for
279 each constituent muscle at 5% intervals along the length of the femur, and overall QF ACSA
280 at each 5% interval calculated by summation. QF ACSA_{MAX} was calculated as the summation
281 of the maximal measured ACSA from each constituent muscle and the location relative to
282 femur length was also recorded.

283

284 The volume of each constituent QF muscle was calculated as the area under the ACSA-
285 femur length curve following cubic spline interpolation, and the constituent muscle volumes
286 summed for overall QF VOL. The $_{EFF}PCSA$ of each individual QF muscle was calculated as:
287 muscle volume (cm³) divided by L_F multiplied by the cosine of θ_P . L_F and θ_P were mean values
288 from two ultrasound measurement sites along the length of each individual QF muscle. Overall
289 QF $_{EFF}PCSA$ was calculated via the summation of the individual QF muscle $_{EFF}PCSA$ s.

290

291 **Moment arm and calculation of muscle force.** Immediately after thigh imaging, a
292 lower extremity knee coil was used to acquire sagittal images (time of repetition/time to echo
293 480/14, image matrix 512 x 512, field of view 160 x 160 mm, pixel size 0.313 x 0.313, slice
294 thickness 2 mm, inter-slice gap 0 mm) of the knee joint. Sagittal plane images were used to
295 determine PT_{MA} , the perpendicular distance from the patellar tendon line of action to the tibio-
296 femoral contact point, which was approximately the mid-point of the distance between the
297 tibiofemoral contact points of the medial and lateral femoral condyles (Fig. 1C). The tibio-
298 femoral contact point was used as an approximation of the joint centre of rotation (35). Due to
299 constraints in the size of the knee coil, sagittal images were acquired in an extended knee

300 position ($\sim 163^\circ$). PT_{MA} length was then corrected to that during MVC by estimating moment
301 arm at 115° from previously published data fitted with a quadratic function (36) scaled to each
302 participant's measured moment arm length at 163° . Internal muscle force was subsequently
303 calculated as follows: (external knee extensor MVT + estimated antagonist knee flexor torque)
304 \div corrected PT_{MA} . Antagonist knee flexor torque at knee extension MVT was estimated by
305 expressing the antagonist HEMG amplitude during knee extensor MVT relative to the knee
306 flexor EMG_{MAX} (normalized antagonist HEMG) and multiplying by the knee flexor MVT
307 (assuming a linear relationship between EMG amplitude and torque).

308

309 *Statistics*

310 All statistical analyses were conducted using SPSS Version 26.0 (IBM Corp., Armonk,
311 NY, USA), unless stated. Significance was defined as $P < 0.05$. MVT (knee extension and
312 flexion) and normalized antagonist HEMG from each of the duplicate test sessions was
313 averaged for each participant to produce criterion values for the calculation of muscle force
314 and statistical analysis. Knee extension MVT, muscle force, QF size (VOL, $ACSA_{MAX}$,
315 $EFFPCSA$, and muscle thickness), PT_{MA} , QF L_F and QF θ_P were tested for outliers using the
316 Grubbs' test, also referred to as the ESD method (extreme studentized deviate;
317 <https://www.graphpad.com/quickcalcs/grubbs1/> (36)), and no outliers were detected. Values
318 presented for group level results are Mean \pm SD. Data normality was assessed using the Shapiro
319 Wilk test. MVT was the only variable found to not be normally distributed and was transformed
320 (log 10) to meet parametric statistical testing requirements of normality prior to further
321 statistical analysis. Pearson's product moment bivariate correlations were conducted between
322 MVT / muscle force and the different measures of overall QF size (VOL, $ACSA_{MAX}$, $EFFPCSA$,
323 and muscle thickness). To statistically assess differences between Pearson's product moment
324 bivariate correlations (e.g. MVT vs. QF VOL compared to MVT vs. QF $ACSA_{MAX}$, MVT vs.

325 QF VOL compared to muscle force vs. QF VOL, or MVT vs. ACSA at 50% of femur length
326 compared to MVT vs ACSA at 70% of femur length) an online resource
327 (<http://comparingcorrelations.org>; (37) was used to implement Meng et al.'s (38) z test (two
328 dependent groups [i.e., same group], overlapping [i.e. one variable in common], two-tailed test,
329 alpha level= 0.05, confidence value= 0.95, null value= 0). Pearson's product moment bivariate
330 correlations were also calculated between MVT and location specific QF ACSA measurement
331 at 5% intervals between 10 and 90% of femur length. Between-participant coefficient of
332 variation (CV_B) for all variables was calculated as follows: $[\text{cohort SD} \div \text{cohort mean}] \times 100$.
333 L_F and θ_P were compared between muscles (i.e. once mean values had been derived across two
334 measurement sites within each muscle) using a one-way ANOVA with stepwise multiple
335 comparison corrected least significant difference (LSD) post-hoc testing.

336

337 **Results**

338 *Group level muscle strength, size and architecture*

339 Knee extension MVT was 246 ± 42 Nm (range: 173-396 Nm; CV_B 17.3%) and QF
340 muscle force was 5874 ± 960 N (range: 3886-8681 N; CV_B 16.3%), respectively. Whole QF
341 $EFFPCSA$, VOL, and $ACSA_{MAX}$ were 167 ± 19 cm² (range: 124-206 cm²; CV_B 11.4%), $1838 \pm$
342 263 cm³ (range: 1254-2573 cm³; CV_B 14.3%), and 90 ± 12 cm² (range: 68-125 cm²; CV_B
343 13.8%), respectively. QF muscle thickness was 92 ± 11 mm (range: 74-123 mm; CV_B 11.5%).
344 Constituent QF muscle size variables are reported in Table 1.

345

346 Overall QF L_F was 106.6 ± 8.9 mm (range: 87.8-125.5 mm; CV_B 8.4%), and QF θ_P was
347 $15.5 \pm 1.8^\circ$ (range: 12.2-19.0°; CV_B 11.5%). L_F (i.e. mean of 2 sites) differed between
348 constituent QF muscles (ANOVA $P < 0.001$). Specifically, VI L_F (98.7 ± 9.7 mm) was shorter
349 than that of the VM (105.8 ± 15.2 mm), VL (113.1 ± 11.7 mm) and RF (108.9 ± 14.7 mm; LSD

350 0.001≤P<0.019) and VM L_F was shorter than that of VL (LSD P=0.019). L_F did not differ
351 between any other individual QF muscles (LSD 0.211≤P≤0.221). θ_p differed between
352 constituent QF muscles (One-way ANOVA P<0.001). Post-hoc testing revealed that θ_p
353 differed between all individual QF muscles (LSD [all] P<0.001; VL 16.0 ± 3.2°; VI 13.4 ±
354 3.3°; VM 19.2 ± 3.9°; and RF 13.5 ± 2.6°) except between VI and RF (LSD P=0.814). PT_{MA}
355 was 4.41 ± 0.30 cm (range: 3.65-5.16 mm; CV_B 6.7%).

356

357 *Correlation of muscle size variables with maximum voluntary torque and muscle force.*

358 MVT was correlated with all four QF size variables with the bivariate correlation
359 between MVT and VOL producing the highest r-value (r=0.773, P<0.001), followed by
360 ACSA_{MAX} (r=0.697, P<0.001), EFFPCSA (r=0.685, P<0.001) and muscle thickness (r=0.406,
361 P=0.003; Fig. 2A-D). Statistical comparisons revealed no differences between the bivariate
362 correlation coefficients of MVT and MRI-derived measures of muscle size: MVT with VOL
363 or ACSA_{MAX} (z=1.614, P=0.107), MVT with VOL or EFFPCSA (z=1.555, P=0.120), MVT
364 with ACSA_{MAX} or EFFPCSA (z=0.151, P=0.880). However, correlation coefficients between
365 MVT with VOL (z=3.699, P<0.001), ACSA_{MAX} (z=2.417, P=0.015), or EFFPCSA (z=2.393,
366 P=0.017) were each greater than the correlation coefficient between MVT and muscle
367 thickness.

368 Muscle force was correlated with, or had a tendency to be correlated with, all four QF
369 muscle size variables but with lower correlation coefficients than for MVT (VOL r=0.627,
370 P<0.001; ACSA_{MAX} r=0.598, P<0.001; EFFPCSA r=0.575, P<0.001; and muscle thickness
371 r=0.269, P=0.054; Fig. 3A-D). The correlation coefficients produced for each muscle size
372 variable with muscle force were lower than for with MVT: VOL with muscle force or MVT
373 (z=3.306, P<0.001), ACSA_{MAX} with muscle force or MVT (z=2.069, P=0.039), EFFPCSA with
374 muscle force or MVT (z=2.252, P=0.024), and muscle thickness with muscle force or MVT

375 ($z= 2.275, P=0.023$). In summary, muscle size variables explained 9.2 to 20.4% more of the
376 variance in MVT than in muscle force. For context we also calculated the correlation between
377 PT_{MA} and MVT finding a weak, significant relationship ($r=0.285; P=0.041$).

378

379 *Anatomical cross-sectional area along the length of the femur*

380 The $ACSA_{MAX}$ of each of the four constituent muscles occurred at $28.4 \pm 3.6\%$ (VM),
381 $57.1 \pm 5.1\%$ (VL), $57.9 \pm 4.3\%$ (VI) and $68.2 \pm 5.7\%$ femur length (RF; Fig. 4A). Considering
382 location/femur length specific ACSA of the whole QF (i.e. sum of all 4 constituents at each
383 femur length), the highest ACSA occurred at 55% femur length ($71 \pm 10 \text{ cm}^2$; Fig. 4B). As the
384 association between muscle force and muscle size measures were weaker than that for MVT
385 (see previous paragraph), only bivariate correlations between location specific ACSA measures
386 and MVT were conducted. Significant correlations were observed between MVT and QF
387 ACSA measurements at 25-85% of femur length, with the highest correlation ($r= 0.719,$
388 $P<0.001$) occurring at 65% of femur length (i.e. more proximal than the position of highest
389 location specific ACSA; Fig. 4C). In fact, ACSA measured at 65% femur length and two
390 adjacent locations (60 and 70%) had marginally, but not significantly ($0.0688 \leq z \leq 0.4744,$
391 $0.6352 \leq P \leq 0.9452$), higher correlation coefficients with MVT ($r=0.701$ to 0.719) than
392 $ACSA_{MAX}$ ($r=0.697$). Statistical comparisons between correlation coefficients revealed that the
393 association between MVT and ACSA at 65% of femur length was: greater than the association
394 between MVT and ACSA at 10-45% of femur length ($1.979 \leq z \leq 4.457, 0.001 \leq P \leq 0.048$) and
395 75-90% of femur length ($2.093 \leq z \leq 3.493, 0.001 \leq P \leq 0.036$); and had a tendency to be greater
396 than 50-55% of femur length ($z= 1.711-1.843, P=0.065-0.087$). The association of MVT-
397 ACSA at 65% of femur length did not differ compared to the that between MVT-ACSA at 60%
398 or 70% of femur length ($0.556 \leq z \leq 0.656, 0.512 \leq P \leq 0.579$).

399

400 **Discussion**

401 This study examined the relationship between QF size measures (VOL, $_{EFF}PCSA$,
402 $ACSA_{MAX}$ and muscle thickness) and both knee extensor strength (MVT) and internal muscle
403 force in a large cohort of healthy young men, as well as the influence of ACSA location along
404 the femur on the relationship with strength. Our first two hypotheses were refuted as
405 incorporating thorough muscle architecture measurements when determining $_{EFF}PCSA$ did not
406 result in this index of QF size becoming the pre-eminent muscle size determinant of MVT, and
407 muscle force was not more strongly associated with muscle size variables than joint-level
408 MVT. In agreement with our final hypothesis the MVT-ACSA relationship was influenced by
409 ACSA measurement location with stronger correlations at specific locations of 60-70% of
410 femur length than for other locations.

411

412 Knowledge of the muscle size measurements underpinning strength and strength
413 change are important in order to understand, assess/monitor and potentially modify the most
414 important determinants. Surprisingly, in the current study determination of $_{EFF}PCSA$ involving
415 comprehensive MRI scanning along the length of the constituent QF muscles, and the
416 incorporation of two ultrasonographic muscle architecture measurements per muscle did not
417 result in $_{EFF}PCSA$ being the highest correlate of MVT. In fact, comparisons of correlation
418 coefficients revealed no difference between the association of $_{EFF}PCSA$, $ACSA_{MAX}$, or VOL
419 with MVT, although qualitatively VOL explained 59.8% of the variance in MVT, which was
420 ~11-13% greater than either $_{EFF}PCSA$ (46.9%) or $ACSA_{MAX}$ (48.6%). As far as we are aware
421 the current study involved the most thorough in vivo investigation to date with: (i) a large
422 cohort ($n=52$ vs. $n=19$ (17), $n=26$ (11), $n=39$ (7)); (ii) a relatively homogenous cohort (low-
423 moderate physically active males, as opposed to mixed sex and training status groups that
424 might introduce other variables); (iii) duplicate measurements of knee extension strength on

425 two occasions with a highly reliable dynamometer and protocol (between-session within-
426 participant coefficient of variation of 3.0% in the current experiment). In addition, specific to
427 the $_{EFF}PCSA$ measurements we took multiple ultrasonographic measurements at two sites for
428 each constituent muscle with a long ultrasound array to provide a rigorous assessment of
429 muscle architecture, as opposed to estimation from cadaveric data (7) or muscle length ((11)–
430 elbow flexors), or single site ultrasound measurements ((11)– elbow extensors). In keeping
431 with previous literature (39) this revealed that muscle architecture (θ_p and L_f) differed between
432 the constituents QF muscles indicating that all four muscles should be assessed to accurately
433 determine $_{EFF}PCSA$ of each muscle and thus also of the whole muscle group. Despite these
434 attempts to improve the $_{EFF}PCSA$ measurement, the current study provides robust evidence that
435 with these techniques $_{EFF}PCSA$ is not more strongly associated with strength than VOL or
436 $ACSA_{MAX}$, and qualitatively VOL actually explained ~13% more variance than $_{EFF}PCSA$. Our
437 finding that VOL was qualitatively the greatest correlate of muscular strength over cross-
438 sectional area measurements ($_{EFF}PCSA$ and $ACSA_{MAX}$) was consistent with one previous
439 report ((11)– elbow extensors), but not others that found $_{EFF}PCSA$ ((11)– elbow flexors) or
440 $ACSA_{MAX}$ (7) to be pre-eminent, but these studies did not complete statistical comparisons of
441 these associations. Only one previous report did a statistical comparison, also finding VOL to
442 be a superior, but not statistically greater, determinant of muscular strength than ACSA (17).
443 Overall, the differences between these three indices of muscle size ($_{EFF}PCSA$, $ACSA_{MAX}$,
444 VOL) in predicting strength appear relatively subtle, but with 3 of 5 datasets indicating VOL
445 maybe marginally superior.

446

447 The statistically equivalent but qualitatively weaker association of $_{EFF}PCSA$ with
448 strength, compared to VOL, appears contrary to classic physiological theory that $_{EFF}PCSA$ best
449 represents the number of actin-myosin cross bridges in parallel and able to generate tension

450 between tendons. There seem to be four possible explanations for this finding. Firstly, our
451 ability to assess complex muscle architecture, and thus accurately determine $_{EFF}PCSA$, may be
452 limited with the use of two-dimensional ultrasonography. Employing more sophisticated three-
453 dimensional muscle architecture imaging techniques (i.e. diffusion tensor MRI) for the
454 determination of $_{EFF}PCSA$ seems a logical progression for future research. Secondly, it seems
455 likely that VOL can be measured with less error than $_{EFF}PCSA$, given the calculation of PCSA
456 involves the combination of multiple variables with the potential for accumulated measurement
457 error that might reduce the association with MVT. Thirdly, classic physiology assumes purely
458 longitudinal force transmission. However, evidence for the existence and importance of lateral
459 force transmission has been documented in both amphibian myofibers (19) and mammalian
460 whole muscle (40, 41). Therefore, lateral force transmission, potentially between fibres,
461 fascicles and even constituent muscles (20, 42, 43) could mean that $_{EFF}PCSA$ is not the only
462 geometric determinant of contractile force production. Specifically, due to lateral force
463 transmission the length and/or shape of a muscle/muscle group may also influence force
464 production and explain why VOL qualitatively explained the largest amount of variance in
465 muscular strength in the current study. Finally, whilst PT_{MA} was a weaker predictor of isometric
466 strength in this study ($r=0.285$) than in some previous reports (17, 24, 44–46; $r= 0.400$ to
467 0.790), it is possible that VOL, which incorporates muscle length as well as cross-sectional
468 area, may provide a (better) proxy for body size and PT_{MA} , potentially explaining the higher
469 correlation of VOL with MVT than cross-sectional areas.

470

471 Muscle thickness measured with ultrasound was a statistically weaker correlate of MVT
472 ($r=0.406$) than the MRI measures of muscle size (i.e. VOL, $ACSA_{MAX}$, or $_{EFF}PCSA$; $r= 0.685$ -
473 0.773). Similar ($r= 0.411$ - 0.470 ; (47, 48), and greater ($r= 0.550$ - 0.700 (49, 50)) associations
474 between QF muscle thickness and muscular strength have been reported in the research

475 literature. However, these prior studies measured only one site per muscle and several
476 measured only two of the four constituent QF muscles (47, 48, 50). Overall, the measurement
477 of muscle thickness with ultrasound appears to be a substantially weaker determinant of
478 isometric strength than MRI-based measures of muscle size, in this study explaining >30% less
479 variance in strength (explained variance: 16.5% for MT vs 46.9-59.8% for MRI measures).
480 Thus, while ultrasound measures of muscle thickness have the advantages of convenience and
481 low cost they are significantly less informative about the functional capacity of the quadriceps
482 and thus may have limited utility in comparing or monitoring individuals (i.e. during training,
483 disuse or aging).

484

485 We had anticipated that the joint-level relationship between muscle size and strength
486 may be limited by the influence of other neuromechanical factors (i.e. moment arm and
487 antagonist activation/torque) that might dilute/confound this relationship, and thus removal of
488 these factors would lead to a stronger relationship between muscle force and size variables.
489 However, the relationships between muscle force and muscle size variables were consistently
490 and statistically lower than for joint-level MVT (i.e. explaining 9.2 to 20.4% less variance). In
491 essence contrary to our hypothesis removal of the confounding factors (moment arm and
492 antagonist torque) weakened rather than strengthened the relationship of muscle size with
493 force/torque. Given the convincing evidence for a very high correlation between $_{EFF}PCSA$ and
494 muscle force in isolated muscles ($r=0.99$; (8)), it seems our attempt to remove joint-level
495 neuromechanical factors may have further confounded rather than distilled the relationship
496 between muscle size and force/torque. This finding might question the validity of the moment
497 arm and antagonist measurements used in the current study, or suggest accumulated
498 measurement error when estimating muscle force via a calculation involving multiple
499 variables. Furthermore, the calculation of muscle force has been used as an intermediate step

500 for the assessment of specific tension (which subsequently involves dividing muscle force by
501 cross-sectional area; (51, 52)), and these results also query the validity of estimating specific
502 tension in this way.

503

504 It is possible that the contemporary methods that we employed are not sufficient to
505 accurately assess antagonist torque. Whilst we assessed activation of the largest of the knee
506 flexors (medial and lateral hamstrings), only two/three of nine knee flexor muscles were
507 measured; and antagonist torque was estimated based on an assumed linear relationship with
508 EMG (22), and this relationship may also be confounded by cross-talk from other muscles (53).
509 Improvements in the assessment of antagonist torque during knee extension seem warranted
510 and this may require more comprehensive assessment of antagonist co-activation. For example,
511 with the measurement of surface EMG of additional knee flexor muscles (i.e. the medial and
512 lateral gastrocnemius) and more careful determination of the torque-EMG relationship for
513 these muscles. In practice, however, antagonist torque was estimated as 5.3% of knee extension
514 MVT, and thus appeared to be a relatively modest correction within the calculation of
515 quadriceps muscle force. Given this modest influence of correcting for antagonist EMG on the
516 muscle force calculation, this may place particular concern on the functional validity of the
517 PT_{MA} measurement also used in the calculation of muscle force. Most contemporary studies,
518 including the current investigation, typically measure PT_{MA} in resting conditions to ensure
519 good image quality (i.e. no movement artefact), and at a relatively extended knee joint angle
520 due to space constraints within the bore of an MRI scanner. However, it is known that PT_{MA}
521 changes with contraction vs. rest (54) and joint angle (36). Although we corrected PT_{MA} values
522 according to the differences in knee joint angle between the imaging and the strength
523 measurements (52), it is possible that these confounding factors in the assessment of PT_{MA} may
524 have compromised the precision of this measurement.

525 The relationship between MVT and ACSA in the present investigation was found to be
526 systematically influenced by the location of the ACSA measurement relative to femur length,
527 with r-values progressively increasing from 10% up to a peak at 65% of femur length ($r=0.719$)
528 before gradually declining between 65% and 90% of femur length. Thus the location showing
529 the largest association between MVT and ACSA (65% of femur length) was proximal of the
530 location with the largest location specific ACSA of the whole quadriceps, which had a
531 marginally lower correlation (55% of femur length, $r=0.633$). The correlation coefficient
532 between MVT and ACSA measured at 65% of femur length was found to be greater than, or
533 have a tendency to be greater than, all other ACSA measures along the femur other than those
534 directly adjacent (i.e. ACSA at 60 and 70% of femur length). Additionally, the r-value for the
535 association between MVT and ACSA at 65% femur length was also marginally superior, but
536 statistically equivalent to that of ACSA_{MAX} (i.e. sum of maximum ACSA from each constituent
537 QF muscle, irrespective of location) and MVT ($r=0.697$). Therefore, this rigorous MRI
538 assessment at locations all along the femur supports the earlier suggestion based on ultrasound
539 measurements that proximal QF ACSA may be particularly important for the strength of this
540 muscle group (24). One practical implication of our observation that ACSA at 65% of femur
541 length had a similar association with MVT as ACSA_{MAX} is that a single slice image at this one
542 location, which is a quicker, cheaper and analytically less laborious than the imaging of the
543 whole muscle needed to determine ACSA_{MAX}, may be as effective at predicting function.

544

545 It was notable that the location specific ACSA (65% of femur length) showing the
546 highest correlation with MVT was also proximal to the ACSA_{MAX} of all three vastii muscles
547 (VM 28.4%; VL 57.1%; VI 57.9%) collectively comprising 87% of QF volume, with only the
548 RF having a more proximal ACSA_{MAX} (68.2%). We are not aware of any reason that the RF
549 would have a disproportionate influence on knee extension strength. Alternative explanations

550 for this observation of proximal QF ACSA being most strongly related to strength may include
551 the differences between the imaging and strength measurements, in terms of both the state of
552 muscle contraction (resting for MRI vs. maximum contraction for MVT) and joint
553 configurations (hip and knee close to the anatomical position for MRI scanning vs. flexed hip
554 and knee at MVT for the isometric dynamometry). For example with the hip and knee flexed
555 in the dynamometer it might be expected that all four constituents of the QF would move
556 distally compared to the anatomical position in the MRI scanner (i.e. proximal muscle locations
557 [$\sim 65\%$ of femur length] in the scanner may be closer to mid-thigh when seated on the
558 dynamometer). On the other hand, as the muscle contracts, even during an isometric
559 contraction the fibres and fascicles shorten considerably. We have previously found fascicle
560 length within the QF to shorten by 24% between rest and MVT, and as the muscle remains
561 isovolumetric, cross-sectional area shows a corresponding increase ($_{EFF}PCSA +27\%$ from rest
562 to MVT; (18)). In the QF this shortening of the muscle belly occurs in a non-symmetrical
563 manner primarily due to lengthening of the distal connective tissues, as for example the deep
564 VL aponeurosis at mid-thigh (50% femur length) has been shown to move ~ 17 mm proximally
565 from rest to 80% MVT (55). Therefore, the substantial increase in cross-sectional area, from
566 rest to MVT is likely accompanied by a proximal shift in the location of $ACSA_{MAX}$. In summary
567 the way in which these potentially competing effects of contraction state and joint
568 configuration combine to explain the apparent importance of proximal ACSA for isometric
569 strength measurements is unclear.

570

571 The current study was not without its limitations and it is important to acknowledge
572 them. The highly specific nature of the strength assessment (isometric contraction of the knee
573 extensors at a knee joint angle of 115°) used in the current investigation means that the results
574 presented cannot necessarily be generalized or assumed to be the same for other muscle groups,

575 modes of contraction (i.e. concentric or eccentric) or joint angles. Theoretically, as muscle
576 architecture is known to change substantially with contractile force (18) the most relevant
577 muscle architecture measurements for maximum contractile function would seem be those
578 measured at MVT. In practice, however, obtaining high quality architecture measurements at
579 MVT is highly challenging, and would not have been feasible for two sites of each of the four
580 individual muscles within the current study. Furthermore, based on our prior work the use of
581 architecture measurements made during MVCs, compared to resting measurements, did not
582 enhance the relationship between knee extension MVT and $QF_{EFF}PCSA$ (18). Finally, the aim
583 of this study was to examine human muscle size-strength relationships and the influence of
584 some specific methodological considerations. We are conscious that other factors, beyond the
585 scope of this study, have also been suggested to influence strength (e.g. fibre type composition
586 (56) and agonist neuromuscular activation (57, 58)), and future work could examine
587 multifactorial determinants of strength.

588

589 In conclusion, despite incorporating comprehensive muscle architecture measurements
590 to enhance the determination of $EFFPCSA$, statistical comparisons of correlation coefficients
591 revealed no differences between the association of $EFFPCSA$, ACSA or VOL with MVT; and
592 VOL explained the highest variance in isometric knee extension MVT (~60%). This suggests
593 that with contemporary methods of muscle architecture measurements, $EFFPCSA$ offers no
594 advantage over purely MRI-derived indices of muscle size, and researchers interested in
595 understanding/explaining muscular strength may wish to use muscle volume as it does not
596 require additional architecture measurements and appears to explain marginally more variance
597 in strength. The location of ACSA measurements substantially effected the strength of the
598 association with MVT, with the highest association for ACSA measured at the relatively
599 proximal position of 65% of femur length. ACSA measured at this location was as strongly

600 associated with MVT as ACSA_{MAX} despite requiring only a single slice/image in contrast to
601 scanning the whole muscle for ACSA_{MAX}.

602

603 **Grants**

604 This study was supported financially by the Versus Arthritis Centre for Sport, Exercise, and
605 Osteoarthritis (Grant reference 20194).

606

607

608 **Acknowledgements**

609 The authors thank the participants for their time in taking part in the study.

610

611 The authors declare no conflict of interest (financial or otherwise) and that the results of this
612 study are presented clearly, honestly, and without fabrication, falsification, or inappropriate
613 data manipulation. The results of this study do not constitute endorsement by the American
614 College of Sports Medicine.

615

616 **AUTHOR CONTRIBUTIONS**

617 T.G.B., G.J.M., T.M.M.-W., and J.P.F. conception and design of research;

618 T.G.B., G.J.M., and T.M.M.-W. performed experiments;

619 T.G.B. and T.M.M.-W. analyzed data;

620 T.G.B. and J.P.F. interpreted results of experiments;

621 T.G.B. prepared figures;

622 T.G.B., and J.P.F. drafted manuscript;

623 T.G.B., G.J.M., T.M.M.-W., and J.P.F. edited and revised manuscript;

624 T.G.B., G.J.M., T.M.M.-W., and J.P.F. approved final version of manuscript.

625

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628

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FIGURE CAPTIONS

Fig. 1 Representative: (A) ultrasound images of vastus lateralis (VL; 50% of femur length), vastus intermedius (VI; 50% of femur length), rectus femoris (75% of femur length), and vastus medialis (VM; 40% of femur length; 0% is knee joint space); (B) axial magnetic resonance image of the thigh; and (C) sagittal magnetic resonance image of the knee joint. TFCP, tibio-femoral contact point.

Fig. 2 Scatterplots showing the relationships between knee extension maximum voluntary torque (MVT) and quadriceps femoris size measures: (A) muscle volume [VOL]; (B) maximum anatomical cross-sectional area [$ACSA_{MAX}$]; (C) effective physiological cross-sectional area [$EFFPCSA$]; and (D) muscle thickness. Solid lines indicate the trend of the relationship between variables.

Fig. 3 Scatterplots showing the relationships between quadriceps femoris muscle force and quadriceps femoris size measures: (A) muscle volume [VOL]; (B) maximum anatomical cross-sectional area [$ACSA_{MAX}$]; (C) effective physiological cross-sectional area [$EFFPCSA$]; and (D) muscle thickness. Solid lines indicate the trend of the relationship between variables.

Fig. 4 Location specific anatomical cross-sectional area (ACSA) at 5% intervals along the length of the femur for (A) the constituent quadriceps femoris (QF) muscles and (B) overall QF muscle group. (C) Bivariate correlations between QF ACSA at 5% intervals along femur length and knee extension isometric maximum voluntary torque; 0% = distal, 100% = proximal. Significance of bivariate correlations are indicated as follows: * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$. Data displayed in A and B are mean \pm SD.

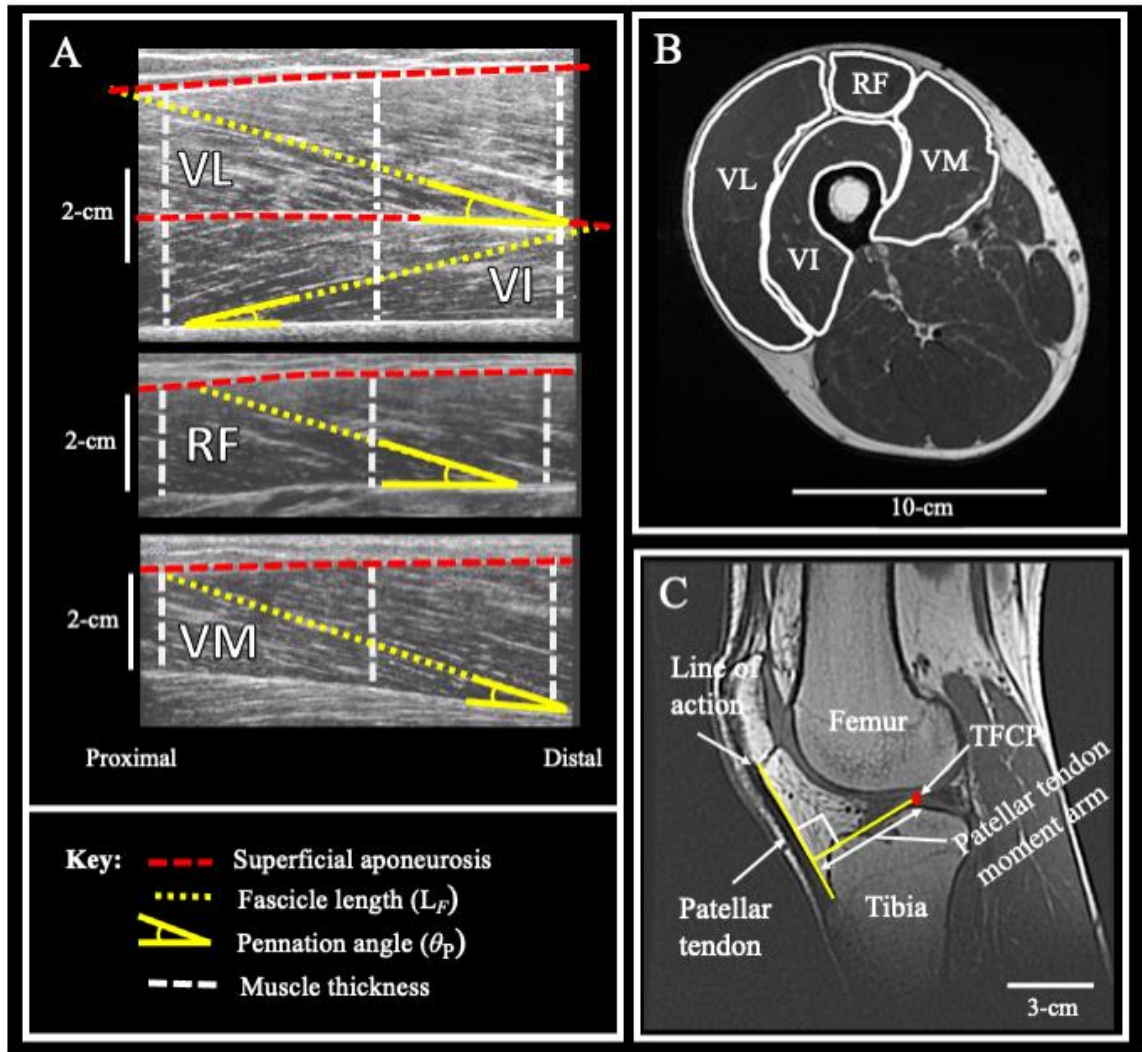


Fig. 1

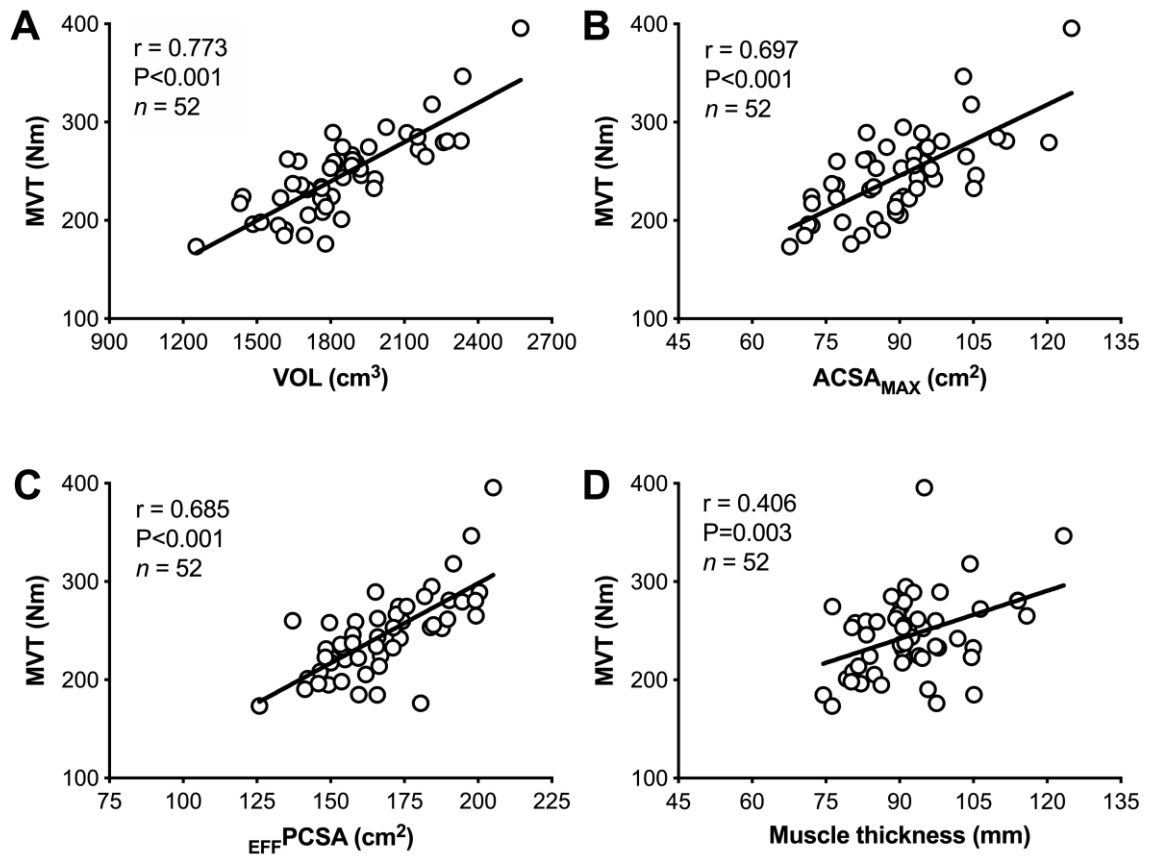


Fig. 2

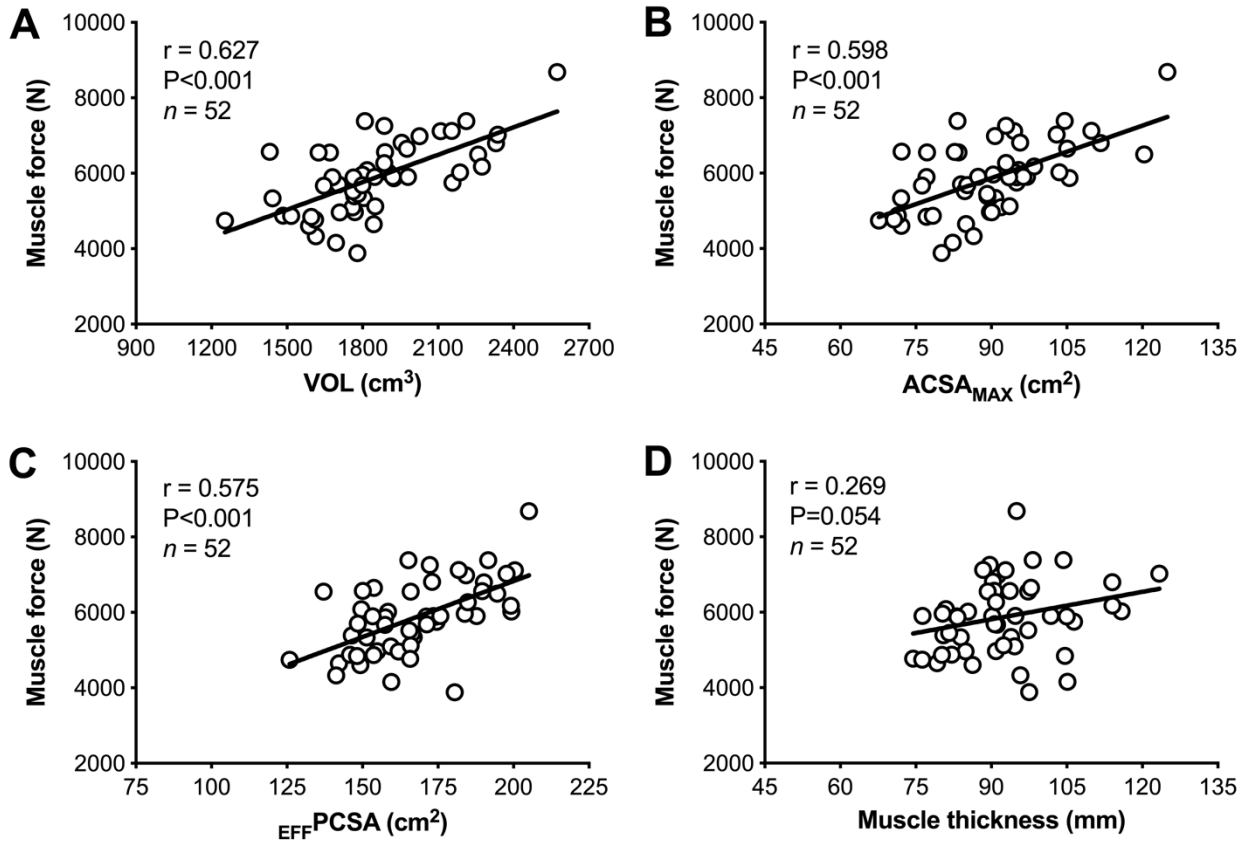


Fig. 3

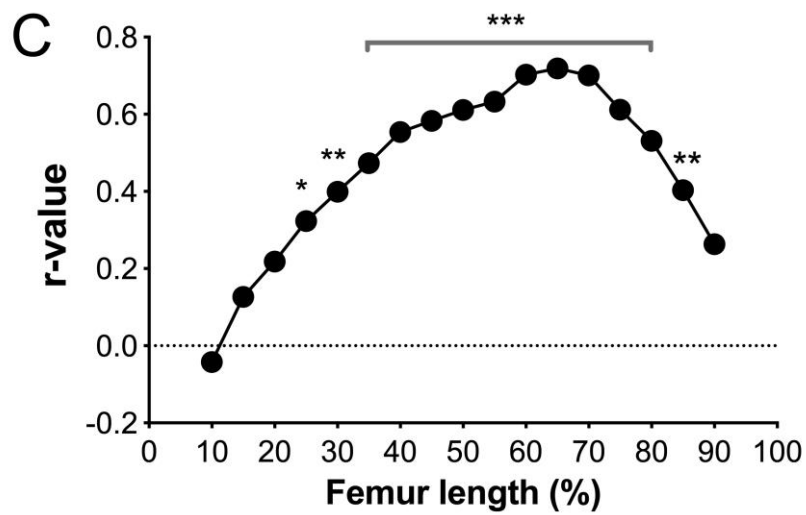
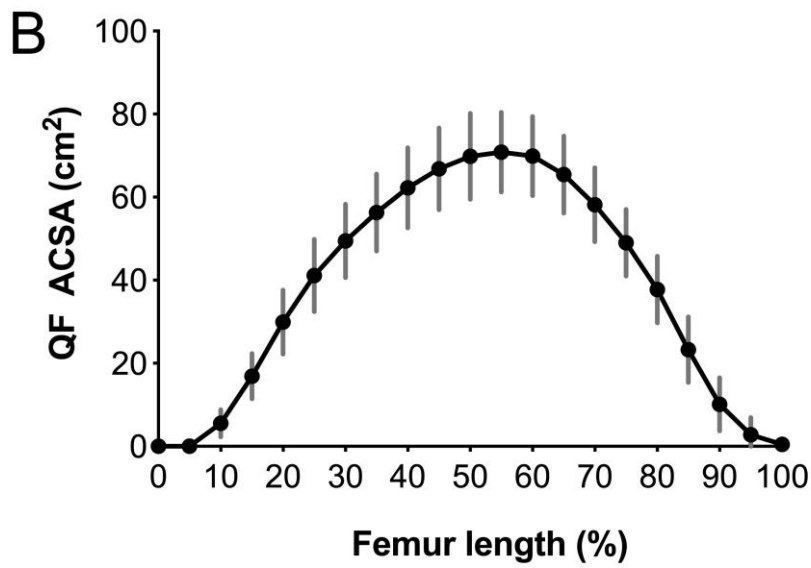
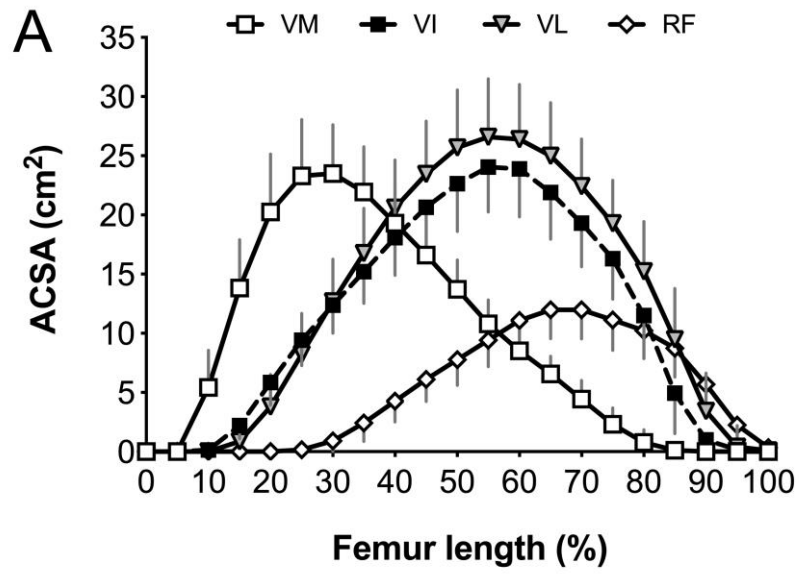


Fig. 4

TABLES

Table 1. Muscle size of overall and constituent quadriceps femoris (QF) muscles

	Muscle volume (cm ³)	ACSA _{MAX} (cm ²)	EFFPCSA (cm ²)	Muscle thickness (mm)
VM	441 ± 68	25 ± 4	40 ± 7	26 ± 5
VI	547 ± 104	25 ± 4	54 ± 10	20 ± 4
VL	610 ± 98	28 ± 5	52 ± 8	25 ± 3
RF	240 ± 47	13 ± 2	21 ± 3	22 ± 3
QF	1838 ± 263	90 ± 12	167 ± 19	92 ± 11

Data are Mean ± SD. VM, vastus medialis. VI, vastus intermedius. VL, vastus lateralis. RF, rectus femoris.