

1 **Title:**

2 Architectural changes of the biceps femoris after concentric or eccentric training.

3

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25 **ABSTRACT**

26 **Purpose:** To determine i) the architectural adaptations of the biceps femoris long head (BFIf)
27 following concentric or eccentric strength training interventions; ii) the time course of
28 adaptation during training and detraining. **Methods:** Participants in this randomized
29 controlled trial (control [n=28], concentric training group [n=14], eccentric training group
30 [n=14], males) completed a 4-week control period, followed by 6 weeks of either concentric-
31 or eccentric-only knee flexor training on an isokinetic dynamometer and finished with 28
32 days of detraining. Architectural characteristics of BFIf were assessed at rest and during
33 graded isometric contractions utilizing two-dimensional ultrasonography at 28 days pre-
34 baseline, baseline, days 14, 21 and 42 of the intervention and then again following 28 days of
35 detraining. **Results:** BFIf fascicle length was significantly longer in the eccentric training
36 group ($p<0.05$, d range: 2.65 to 2.98) and shorter in the concentric training group ($p<0.05$, d
37 range: -1.62 to -0.96) after 42 days of training compared to baseline at all isometric
38 contraction intensities. Following the 28-day detraining period, BFIf fascicle length was
39 significantly reduced in the eccentric training group at all contraction intensities compared to
40 the end of the intervention ($p<0.05$, d range: -1.73 to -1.55). There was no significant change
41 in fascicle length of the concentric training group following the detraining period.
42 **Conclusions:** These results provide evidence that short term resistance training can lead to
43 architectural alterations in the BFIf. In addition, the eccentric training-induced lengthening of
44 BFIf fascicle length was reversed and returned to baseline values following 28 days of
45 detraining. The contraction mode specific adaptations in this study may have implications for
46 injury prevention and rehabilitation.

47 **Key Words:** fascicle; muscle adaptation; hamstring; ultrasound; randomized controlled trial

48 INTRODUCTION

49 The ability of a muscle to produce force is partly governed by its architectural characteristics,
50 such as muscle thickness, pennation angle and fascicle length (17). Architectural
51 characteristics have been shown, in many different muscles, to change when exposed to
52 mechanical stimuli, such as resistance training (2, 3, 21, 28, 32). Understanding the changes
53 to muscle architecture in response to a given stimulus is important when aiming to alter
54 muscle function and the risk of injury (2, 3, 7, 36).

55 During the terminal swing phase of the gait cycle, the hamstrings are required to actively
56 lengthen to decelerate the extending knee and flexing hip (38). It is during this phase of the
57 gait cycle where the hamstrings are at their longest, with the biceps femoris long head (BF_{lh})
58 reaching approximately 110% of its length during upright stance (35). These high force,
59 lengthening actions of the hamstrings may contribute to the high rate of strain injuries during
60 running (26), the majority of which occur in the BF_{lh} (16, 24). Interestingly, a previously
61 strain injured BF_{lh} possesses shorter fascicle lengths and greater pennation angles when
62 compared to the contralateral uninjured BF_{lh} (36). Furthermore differences in fascicle length
63 can alter function, with muscles that possess longer fascicles having a greater maximal
64 shortening velocity when compared to those with shorter fascicles (6, 17). Therefore it is
65 important to develop an understanding of how muscle architecture can be altered by physical
66 training in order to influence function, as well as guide hamstring strain injury prevention and
67 rehabilitation practices.

68 Despite the large amount of research showing a range of architectural adaptations following
69 eccentric training interventions (2, 3, 31), investigations which outline the time course for
70 adaptation, including a period of detraining, are limited. Furthermore, the previous research
71 into the adaptability of the BF_{lh} following a training intervention only compared eccentric

72 training to a non-training control group (28). It is therefore it is unclear how BFlf
73 architectural adaptations might differ after eccentric and concentric strength training.

74 Given the high incidence of hamstring injury in the BFlf (16, 24), it is of interest to see how
75 its architecture is altered following either concentric or eccentric strength training. Therefore
76 the purposes of this study were to: 1) determine the architectural adaptations of the BFlf
77 following either a concentric or eccentric strength training intervention and; 2) determine the
78 time course of BFlf architectural adaptations during a 6-week training intervention, and
79 following a 28 day period of detraining.

80 **METHODS**

81 **Participants**

82 Twenty-eight recreationally active males (age 22.3 ± 4.2 y; height 1.81 ± 0.07 m; body mass
83 76.9 ± 8.2 kg) with no history of lower limb injury in the past 12 months were recruited to
84 participate in this study. All participants provided written informed consent prior to testing
85 and training which was undertaken at the Australian Catholic University, Fitzroy, Victoria,
86 Australia. Ethical approval for the study was granted by the Australian Catholic University
87 Human Research Ethics Committee.

88 **Study design**

89 Participants undertook a maximal isokinetic dynamometry familiarization session no less
90 than 7 days prior to having their BFlf architecture assessed. The familiarization session and
91 architectural assessment was completed on both limbs. Following this initial testing session
92 (*28 days pre-baseline*), the participants were paired according to passive BFlf fascicle length
93 and randomly assigned to one of two training groups (allocation ratio 1:1) to undertake either
94 concentric- or eccentric-only knee flexor strength training. All participants (n=28) returned to
95 the lab 4 weeks later (*baseline*) and had the maximal knee flexor strength and BFlf

96 architectural characteristics assessed on both limbs. Following this the participants underwent
97 6 weeks of either a concentric- or eccentric–strength training intervention in a randomly
98 selected limb (the contralateral limb served as a within-participant control). BFlf architecture
99 of both limbs was re-assessed at days 14, 21 and 42 of the intervention, as well as 28 days
100 after the completion of the strength training intervention. Knee flexor strength of both limbs
101 was re-tested at the end of the training intervention (*day 42*) and 28 days after the completion
102 of the intervention. All tests were performed at the same time of the day for each participant.

103 **Outcome measures**

104 **Isokinetic dynamometry**

105 All knee flexor strength testing was completed on a Humac Norm® isokinetic dynamometer
106 (CSMI, Massachusetts, U.S.A), on both legs (left or right) in a randomized order. Participants
107 were seated on the dynamometer with their hips flexed at approximately 85° from neutral and
108 were restrained by straps around the tested/exercised thigh, waist and chest to minimise
109 compensatory movements. All seating variables (e.g. seat height, pad position, etc.) were
110 recorded to ensure the replication of the participants' positions. Gravity correction for limb
111 weight was also conducted and range of motion was set between 0° and 90° of knee flexion
112 (full extension = 0°) with the starting position for each contraction during strength testing
113 being 90° of knee flexion. The starting position for all training contractions were dependent
114 on training group, with the concentric training group starting from 0° of knee extension and
115 the eccentric group beginning from 90°. Prior to all testing sessions, participants undertook a
116 warm-up consisting of three sets of three concentric knee extension and flexion contractions
117 at an angular velocity of 240°/s. The intensity of these contractions increased each set (1st set
118 ~75% and 2nd set ~90% of the participants perceived maximum) until the final set at this
119 velocity was performed at a maximal level. The test protocol began one minute following the
120 final warm-up set and consisted of three sets of three repetitions of concentric and eccentric

121 maximal voluntary contractions of knee flexion at 60°/s and 180°/s (30s inter-set rest). For all
122 concentric knee flexion efforts, the participants were instructed to ‘pull down’ against the
123 lever as fast as possible, whereas during eccentric contractions they were told to ‘resist’ the
124 lever arm from extending their knee as hard as they could. All participants were provided
125 visual feedback of their efforts as well as being verbally encouraged by the investigators to
126 ensure maximal effort for all contractions. The testing order of contraction modes was
127 randomized across the participant pool and the testing protocol has been previously reported
128 to not alter concentric- or eccentric-knee flexor strength (37). Dynamometer torque and lever
129 position data were transferred to computer at 1 kHz and stored for later analysis where it was
130 fourth-order low pass Butterworth filtered (5 Hz). Peak torques at 240, 180 and 60°/s for
131 concentric and 180 and 60°/s for eccentric knee flexion were defined as the mean of the six
132 highest torque values for each contraction mode at each velocity.

133 **BFlf architectural assessment**

134 Muscle thickness and pennation angle of the BFlf were determined from ultrasound images
135 taken along the longitudinal axis (Figure 1) of the muscle belly utilizing a two dimensional,
136 B-mode ultrasound (frequency, 12 Mhz; depth, 8 cm; field of view, 14 x 47 mm) (GE
137 Healthcare Vivid-*i*, Wauwatosa, U.S.A). The same images were utilized to estimate BFlf
138 fascicle length. The scanning site was determined as the halfway point between the ischial
139 tuberosity and the popliteal crease, along the line of the BFlf. Once the scanning site was
140 determined, the distances of the site from various anatomical landmarks were recorded to
141 ensure its reproducibility for future testing sessions. These landmarks included the ischial
142 tuberosity, fibula head and the posterior knee joint fold at the mid-point between BF and
143 semitendinosus tendon. On subsequent visits the scanning site was determined and marked on
144 the skin and then confirmed by replicated landmark distance measures. All architectural
145 assessments were performed with participants in a prone position and the hip in a neutral

146 position following at least 5 min of inactivity. Assessments at rest were always performed
147 first followed by the graded isometric contraction protocol. Assessment of BFlf architecture
148 at rest was performed with the knee at 0° of knee flexion. Assessment of BFlf architecture
149 during isometric contractions was always performed with the knee at 0° flexion and preceded
150 by a maximal voluntary isometric contraction, performed in a custom made device (25). The
151 graded isometric contractions of the knee flexors were performed in the same device at 25, 50
152 and 75% of maximum voluntary isometric contraction (MVIC) with the participants shown
153 the real-time visual feedback of the force produced to ensure that target contraction
154 intensities were met. Assessment of the MVIC of the knee flexors was undertaken in a prone
155 position, with both the hip and knee fully extended (0°). Participants were instructed to
156 contract maximally over a 5-s period, from which the peak force was used to determine the
157 MVIC.

158 To gather ultrasound images, the linear array ultrasound probe, with a layer of conductive gel
159 was placed on the skin over the scanning site, aligned longitudinally and perpendicular to the
160 posterior thigh. Care was taken to ensure minimal pressure was placed on the skin by the
161 probe as this may influence measurement accuracy (15). Finally, the probe orientation was
162 manipulated slightly by the sonographer (RGT) if the superficial and intermediate
163 aponeuroses were not parallel.

164 Analysis was completed off-line (MicroDicom, Version 0.7.8, Bulgaria). For each image, six
165 points were digitized as described by Blazeovich and colleagues (5). Following the digitizing
166 process, muscle thickness was defined as the distance between the superficial and
167 intermediate aponeuroses of BFlf. A fascicle of interest was outlined and marked on the
168 image. The angle between this fascicle and the intermediate aponeurosis was measured and
169 given as the pennation angle (Figure 1). The aponeurosis angle for both aponeuroses was

170 determined as the angle between the line marked as the aponeurosis and an intersecting
171 horizontal line across the captured image (5, 14). Fascicle length was estimated from an
172 outlined fascicle between the aponeuroses. As the entire fascicle was not visible in the probe
173 field of view its length was estimated via the following validated equation from Blazeovich
174 and colleagues (5, 14):

$$175 \quad FL = \sin(AA + 90^\circ) \times MT / \sin(180^\circ - (AA + 180^\circ - PA)).$$

176 Where FL=fascicle length, AA=aponeurosis angle, MT=muscle thickness, AA=aponeurosis
177 angle and PA=pennation angle.

178 Fascicle length was reported in absolute terms (cm) and also relative to muscle thickness
179 (fascicle length/muscle thickness). The same assessor (RGT) conducted and analysed all
180 scans and was blinded to participant identifiers during the analysis. The methodology utilized
181 in this study to assess the BFIf architectural characteristics has been previously reported by
182 our laboratory (36).

183 **Intervention**

184 The participants performed 6 weeks of either maximal eccentric- or concentric-knee flexion
185 strength training, with two sessions in the intervention's first week and 3 sessions a week
186 thereafter on an isokinetic dynamometer (Humac Norm, CSMI, Massachusetts, U.S.A) using
187 the same range of motion and seat positions configuration as dynamometry testing sessions.
188 Only one limb received the strength training stimulus, with the contralateral limb acting as a
189 within-participant control limb. Across the training period the volume (number) of
190 contractions was increased following the progression below:

- 191 • Week 1:
 - 192 ○ Frequency (days/week) = 2
 - 193 ○ Sets = 4
 - 194 ○ Repetitions = 6

- 195 ○ Total repetitions = 48
- 196 • Week 2:
- 197 ○ Frequency (days/week) = 3
- 198 ○ Sets = 4
- 199 ○ Repetitions = 6
- 200 ○ Total repetitions = 72
- 201 • Week 3:
- 202 ○ Frequency (days/week) = 3
- 203 ○ Sets = 5
- 204 ○ Repetitions = 6
- 205 ○ Total repetitions = 90
- 206 • Week 4:
- 207 ○ Frequency (days/week) = 3
- 208 ○ Sets = 5
- 209 ○ Repetitions = 8
- 210 ○ Total repetitions = 120
- 211 • Week 5:
- 212 ○ Frequency (days/week) = 3
- 213 ○ Sets = 6
- 214 ○ Repetitions = 6
- 215 ○ Total repetitions = 108
- 216 • Week 6:
- 217 ○ Frequency (days/week) = 3
- 218 ○ Sets = 6
- 219 ○ Repetitions = 8
- 220 ○ Total repetitions = 144

221 Each training session was separated by at least 48 hours. Contractions were distributed evenly
 222 across 60°/s and 180°/s. All participants started with two sets of three warm up efforts at
 223 60°/s, in the contraction mode utilized for their training. For all training repetitions, the
 224 concentric training participants were moved to full knee extension (0°) by the investigator
 225 and were instructed to flex their knee as fast as possible through to 90° of knee flexion. The
 226 investigator then returned the lever arm to full knee extension and the subsequent repetition
 227 was completed. This was undertaken until all repetitions were completed in their respective
 228 set, with a 30-s inter-set rest period. The eccentric training participants began with their knee
 229 at 90° of flexion. They were then instructed to maximally flex against the lever arm until full
 230 knee extension was reached (0°). The participant was then instructed to relax, the lever arm
 231 was repositioned to 90° of knee flexion by the investigators and the subsequent contraction

232 was performed. This was undertaken until all repetitions were completed in each set, with a
233 30-s inter-set rest period. All participants were provided visual and verbal feedback on the
234 consistency of the torque produced during each repetition. These were compared against
235 personal best performances, which were known by the participant, to aid motivation. During
236 the pre-control (*28 days pre-baseline to baseline*), intervention (*baseline to intervention day*
237 *42*) and detraining periods (*intervention day 42 to post-intervention day 28*), participants
238 continued their habitual levels of physical activity. The only restriction was to not perform
239 any other lower limb strength exercises. Finally, training compliance was determined as a
240 percentage of sessions that were completed within 24 hours of the intended time.

241 **Statistical analysis**

242 All statistical analyses were performed using SPSS version 22.0.0.1 (IBM Corporation,
243 Chicago, IL). Where appropriate, data were screened for normal distribution using the
244 Shapiro-Wilk test and homoscedasticity using Levene's test. Greenhouse-Geisser adjustment
245 was applied when the assumption of sphericity was violated ($p < 0.05$ for Mauchly's test of
246 sphericity). At each contraction intensity, a split-plot design ANOVA, with the within-
247 participant variables being limb (trained or untrained) and time point (*28 days pre-baseline,*
248 *baseline, intervention day 14, intervention day 21, intervention day 42, post-intervention day*
249 *28*) and the between-subject variable being group (eccentric or concentric), was used to
250 compare changes in BFlf architecture throughout the training study. Architectural changes
251 across the 28 day control period (*28 days pre-baseline to baseline*) were not significant
252 ($p > 0.05$). Therefore when determining the alterations in BFlf architectural characteristics
253 following a 6-week intervention, all comparisons were made to *baseline*. Knee flexor peak
254 torque comparisons, at each contraction velocity, used a similar split-plot design ANOVA,
255 however, with different time point variables (*baseline, intervention day 42, and post-*
256 *intervention day 28*). Where significant limb x time x group interactions for architecture and

257 limb x time for knee flexor peak torque were detected, post-hoc t-tests with Bonferroni
258 adjustments were used to identify which comparisons differed. Significance was set at a
259 $p < 0.05$ and appropriate Cohen's d (8) was reported for the comparison effect sizes, with the
260 levels of effect being deemed small ($d = 0.20$), medium ($d = 0.50$) or large ($d = 0.80$) as
261 recommended by Cohen (1988).

262 **Sample Size**

263 Sample size analysis was completed *a-priori* using G-Power (9). The analysis was based on
264 the anticipated differences in fascicle length following the strength training intervention. The
265 effect size was estimated based on the only intervention study to date that has reported
266 changes in the BFIf architecture (28). That study reported a 33% increase in fascicle length
267 following the intervention with an approximate effect size of 1.9. Therefore an effect size of
268 1.2 was deemed as a reasonable starting point. Power was set at 80% with an alpha level of
269 0.05 returning a calculated sample size of 12 per group. As a cross-reference to confirm the
270 effect size, fascicle length differences in individuals with a unilateral BFIf strain injury
271 displayed an effect size of 1.34 when comparing between the previously injured and
272 contralateral uninjured limb (36).

273 **RESULTS**

274 **Participants**

275 The two training groups were similar with respect to age, height and body mass (eccentric
276 training group: age 21.2 ± 2.7 y, height 1.81 ± 0.06 m, body mass 77.9 ± 9.3 kg; concentric
277 training group: age 23.4 ± 5.1 y; height 1.81 ± 0.07 m; body mass 76.2 ± 7.1 kg). Overall,
278 compliance rates were acceptable for all participants ($92\%\pm 2$; min=85%; max=100%), with
279 no differences when comparing the two groups (eccentric training group: $91\%\pm 2$; concentric
280 training group: $93\%\pm 1$).

281 **BFlf architectural comparisons**

282 **Control period, control limb changes and baseline comparisons**

283 A significant limb x time x group interaction effect was found for fascicle length, fascicle
284 length relative to muscle thickness and pennation angle ($p<0.001$). Post-hoc analyses showed
285 no BFlf architectural variables changed during the 4-week pre-intervention control period
286 ($p>0.05$, d range = 0.03 to 0.17). Similarly, there were no significant differences at any time
287 point, in the non-training control limbs for any BFlf architectural variables ($p>0.05$, d range =
288 0.03 to 0.27). Comparisons of all the BFlf architectural variables at baseline displayed no
289 significant differences between the concentric and eccentric training group in legs that were
290 to be trained (i.e. the training leg) ($p>0.05$, d range = 0.22 to 0.43).

291 **Fascicle length and fascicle length relative to muscle thickness changes**

292 A significant limb x time x group interaction effect was found for fascicle length at all
293 contraction intensities ($p<0.001$). Post-hoc analysis showed that fascicle length was
294 significantly longer in the training limb of the eccentric training group ($p<0.05$, d range: 2.65
295 to 2.98, Table 1, Figure 2) and significantly shorter in the training limb of the concentric
296 training group ($p<0.05$, d range: -1.62 to -0.96, Table 1, Figure 2) after 42 days of the

297 intervention compared to baseline, at all contraction intensities. Additionally there was a
298 significant limb x time x group interaction effect for fascicle length relative to muscle
299 thickness ($p < 0.001$). All post-hoc comparisons for the training limbs of each group are
300 presented in Table 1.

301 Following the 28 day detraining period, fascicle length was significantly reduced in the
302 training limb of the eccentric training group in comparison to the end of the intervention, at
303 all contraction intensities ($p < 0.05$, d range: -1.73 to -1.55, Table 1, Figure 2). Post-hoc
304 analysis showed that fascicle length in the concentric training group following 28 days of
305 detraining was no different to that observed end of the intervention, at any contraction
306 intensity ($p > 0.05$, d range: 0.15 to 0.67, Table 1, Figure 2). All other post-hoc comparisons of
307 fascicle length and fascicle length relative to muscle thickness, 28 days following the
308 intervention period, in the training limbs of both groups are presented in Table 1 and Figures
309 1 to 4.

310 **Muscle thickness and pennation angle changes**

311 No significant limb x time x group interaction effect was found for muscle thickness at any
312 contraction intensity ($p > 0.162$). However, a significant limb x time x group interaction effect
313 was detected for pennation angle at all contraction intensities ($p < 0.001$). Post-hoc analysis
314 showed that pennation angle was significantly reduced in the training limb of the eccentric
315 training group ($p < 0.05$, d range: -1.30 to -0.85, Table 1, Figure 2) and significantly increased
316 in the training limb of the concentric training group ($p < 0.05$, d range: 1.60 to 2.50, Table 1,
317 Figure 1 to 4) after 14 days of the intervention compared to baseline, at all contraction
318 intensities. All other comparisons of pennation angle changes in the training limb of both
319 groups are presented in Table 1.

320 Pennation angle was not significantly different in the training limb of the eccentric training
321 group in comparison to the end of the intervention, at any contraction intensity following the
322 28 day detraining period ($p>0.05$, d range: -0.55 to 0.02, Table 1, Figure 2). Post-hoc analysis
323 showed that following the 28 days of detraining, pennation angle of the concentric training
324 group was no different compared to the end of the intervention, at any contraction intensity
325 ($p>0.05$, d range: -0.63 to -0.27, Table 1, Figure 2). All other comparisons of pennation angle
326 changes following the 28 day detraining period are presented in Table 1.

327 **Strength changes**

328 A significant limb x time interaction effect for knee flexor peak torque was found at all
329 contraction velocities for each group ($p<0.001$). Comparisons at all contraction velocities, at
330 baseline, displayed no significant differences between the concentric and eccentric training
331 group ($p>0.05$). Post-hoc analysis also revealed that knee flexor peak torque increased in both
332 the training limb of the eccentric ($p<0.05$, d range: 0.63 to 0.78, Table 2) and the concentric
333 training group ($p<0.05$, d range: 0.53 to 0.72, Table 2) after 42 days of the intervention, at all
334 contraction velocities, when compared to baseline. There were no significant differences in
335 knee flexor peak torque for the untrained limbs of either group after 42 days of the
336 intervention when compared to baseline, at any contraction velocity ($p>0.05$, d range = 0.11
337 to 0.27).

338 There were no significant differences in knee flexor peak torque at any contraction velocity,
339 in either group when comparing their strength following the 28 day detraining period to the
340 values after 42 days of the intervention ($p>0.05$, d range: -0.30 to -0.16, Table 2).
341 Additionally, knee flexor peak torques at all contraction velocities following the 28-day
342 detraining period were significantly greater in the training limb of both training groups when
343 compared to baseline ($p>0.05$, d range: 0.34 to 0.75, Table 2).

344 **DISCUSSION**

345 To the authors' knowledge, this is the first study reporting divergent BFIf architectural
346 adaptations in response to concentric- or eccentric-strength training. Moreover, it is the first
347 to provide evidence that eccentric training-induced increases in BFIf fascicle length are
348 reversed following 28 days of detraining. The main findings were that eccentric strength
349 training resulted in an increase in estimated BFIf fascicle length and a reduction in pennation
350 angle, whereas concentric strength training caused reductions in estimated fascicle length and
351 increases in pennation angle. Additionally, in those who trained eccentrically, a significant
352 reduction in BFIf fascicle length and a non-significant increase in pennation angle were found
353 following a 28 day detraining period when compared to the end of the strength training
354 intervention. In contrast, the concentrically trained group maintained their BFIf architectural
355 characteristics following 28 days of detraining. Finally, improvements in knee flexor strength
356 were not specific to training contraction mode, with significant improvements in concentric
357 and eccentric strength found in both training groups that persisted through the detraining
358 period.

359 Observations of increases in BFIf fascicle length and a reduction in pennation angle
360 (measured at rest) following eccentric strength training in the current study (Figure 1) aligns
361 somewhat with previous literature (28). Potier and colleagues (2009) found a 33% increase in
362 resting BFIf fascicle length with a non-significant 3.1% reduction in resting pennation angle
363 following 8 weeks of eccentric strength training. In comparison, the current study saw a
364 significant 16% increase in resting BFIf fascicle length (the majority of which occurred
365 within 14 days), with a non-significant 7.5% reduction in resting pennation angle.
366 Differences in the training modalities employed (leg curl vs isokinetic dynamometry),
367 intervention length (8 weeks vs 6 weeks) and the site of assessment may explain the different
368 magnitudes of change reported in these studies. Additionally, no previous literature has

369 examined BFlf architectural alterations during graded isometric contractions, following an
370 intervention. In the present study, increases in BFlf fascicle length were observed at the end
371 of the intervention when assessed during all graded isometric contractions in the eccentrically
372 trained individuals. These increases in fascicle length may occur as a result of the addition of
373 in-series sarcomeres, as has been shown in rat vastus intermedius muscles after five days of
374 downhill and presumably eccentric running exercise (18). However, the architectural
375 alterations seen in this study may not be uniform along the BFlf length. Changes in fascicle
376 length (4), muscle thickness and anatomical cross sectional area, after strength training
377 interventions (3), are variable within a muscle. It is possible that the assessment of BFlf
378 architecture in the current study may have occurred at a point on the muscle where the
379 changes were less prominent in comparison to other studies (28). Alternatively, changes in
380 tendon stiffness could theoretically result in altered fascicle lengths, with stiffer tendons
381 causing an increased tension within the muscle which could then result in the elongation of
382 resting BFlf fascicle length. Further research is needed to clarify the mechanism responsible
383 for fascicle length alterations in humans.

384 No previous studies have compared the architectural alterations in the BFlf, following
385 concentric and eccentric training. However, interventions which have employed concentric-
386 or eccentric-knee extensor training have reported inconsistent architectural adaptations. Some
387 have shown a contraction mode specific adaptation similar to that observed in the current
388 study (10, 29) whilst others have not (3). Additionally knee extensor isometric strength
389 training at short and long muscle lengths has also been shown to increase fascicle length (22).

390 A range of factors such as the relative maximum load (3, 10), the participant's age and
391 physical capacity (29) as well as the training stimulus velocity (33) might explain some of the
392 variance between these results. However it is not known why these alterations in the vastus
393 lateralis differ to those reported in the current study. It is possible that differences in the

394 structural and functional characteristics of the muscles may account for this variability.
395 However future research is needed to assist in determining the BFIf adaptive responses to
396 these and many other variables.

397 The increases in BFIf fascicle length and reductions in pennation angle found in the current
398 study following an eccentric strength training may have implications for hamstring strain
399 injury prevention and rehabilitation. Elite athletes with a unilateral history of BFIf strain
400 injury have shorter fascicles and greater pennation angles on their previously injured limb
401 when compared to the contralateral uninjured limb (36). Individuals with a history of
402 hamstring strain injury are at an increased risk of future injury in comparison to those without
403 a history (24, 26). Therefore if shorter fascicles and greater pennation angles in a previously
404 injured athlete are partial contributors to the elevated risk of re-injury, then understanding the
405 most effective methods for altering these architectural characteristics will be of great value.
406 The current data indicates that the continual application of high-intensity, eccentric-only
407 strength training should be considered in hamstring rehabilitation and prevention programs in
408 order to increase BFIf fascicle length and reduce pennation angle. Additionally the current
409 study results suggest that muscle length in training is possibly not the major factor, as
410 previously suggested (12), in determining fascicle length changes as long length, concentric
411 exercise resulted in shortening of fascicle length. Further research is needed to determine how
412 the combination of both concentric and eccentric contractions during conventional strength
413 training methods may alter BFIf architecture.

414 The very rapid response of BFIf architectural adaptations supports previous literature which
415 has found significant increases in fascicle length and pennation angle in the vastus lateralis
416 within 14 days of the commencement of an eccentrically biased strength training intervention
417 (31). Furthermore, rat vastus intermedius in-series sarcomere numbers have been shown to
418 increase within a week of commencing a downhill running protocol (18). In the current study,

419 the majority of fascicle length and pennation angle changes in the eccentric strength training
420 group occurred within the first 14 days of training, with non-significant changes for the rest
421 of the intervention (Figure 1 to 4). A similar, but inverse response was found in the
422 concentric training group after 14 days of training, with non-significant changes for the
423 remainder of the strength training intervention. These results, along with those from other
424 studies (3, 31) suggest that early adaptations to strength training are not only from a neural
425 mechanism (30), but may also be as a result of architectural adaptations.

426 The reported alterations in muscle architecture following periods of detraining are variable,
427 with most conclusions being drawn from observations of prolonged periods of limb
428 unloading, some of which show significant reductions in fascicle length, pennation angle and
429 muscle volume (20, 32), whereas some display no alterations (1). In regards to the detraining
430 responses following high-intensity eccentric- or concentric-strength training, only one study
431 has investigated this, 3-months after a 10 week intervention in the vastus lateralis (3).
432 Blazeovich and colleagues (2007) found no significant alterations in knee extensor strength or
433 vastus lateralis architectural characteristics following a 3-month detraining period. These
434 results are inconsistent with the findings from the eccentric training group in the current study
435 who displayed a significant reduction in BFlf fascicle length and an increase in pennation
436 angle following 28 days of detraining. In comparison, the concentric group displayed similar
437 findings to Blazeovich and colleagues (2007), with architectural variables remaining
438 unchanged following 28 days of detraining (3). The eccentric training group response to the
439 intervention and then to detraining may be of interest for hamstring strain injury prevention
440 and rehabilitation interventions as it has been argued that shorter fascicles (i.e. with fewer in-
441 series sarcomeres) are more prone to muscle damage during high-intensity, eccentric
442 contractions compared with longer fascicles (11, 19, 36). It remains to be seen what effect
443 conventional strength training exercises, which possess both concentric and eccentric actions,

444 have on hamstring muscle architecture. In addition, the apparent rapid decrease in fascicle
445 lengths when the eccentric stimulus is removed would indicate that constant exposure to
446 eccentric exercise may be important to maintain changes in BFlf architecture following an
447 intervention period.

448 The strength training interventions in the current study induced significant increases in
449 concentric and eccentric strength in the training limb of both the concentric and eccentric
450 training groups (Table 2). Previous research investigating knee flexor strength alterations
451 following eccentric- or concentric-strength training interventions are variable (13, 28). To the
452 authors' knowledge, this is the first study to show improvements in both isokinetically
453 derived concentric and eccentric knee flexor strength independent of training modality.
454 However, improvements in concentric strength following an eccentric strength training
455 intervention have been previously reported in the knee flexors, as well as within other muscle
456 groups (27, 34). There is still some contradictory evidence as to whether a contraction mode-
457 specific strength adaptation occurs following either concentric- or eccentric-training (3, 10,
458 29). The current study shows that increases in eccentric strength can be achieved through
459 long length, concentric strength training in the knee flexors. It is unclear if there might be a
460 contraction-mode specific adaptation in longer training programs. However the current
461 findings must be considered in line with the divergent architectural alterations seen between
462 the two strength training interventions.

463 The authors acknowledge that there are limitations in the current study. Firstly, there are
464 methodological limitations with the use of two-dimensional ultrasound for the estimation of
465 BFlf fascicle length. As the field of view utilised in this study does not capture the entire BFlf
466 fascicle, estimation is required. The equation utilised in this study has been validated against
467 cadaveric samples (14), however it must be recognized that there is still a level of error
468 associated with estimations of BFlf fascicle length. Future studies should consider extended

469 field of view ultrasound methods (23) to reduce the level of error when estimating muscle
470 fascicle length. Secondly, the assessment of muscle architecture was only performed on the
471 BFlf and did not include the other knee flexors. Therefore it is unknown what adaptations
472 these other muscles displayed following the intervention and detraining period. However, as
473 the BFlf is the most commonly strain injured hamstring muscle (16), the alterations following
474 concentric and eccentric strength training interventions were of interest from a hamstring
475 strain injury risk and rehabilitation perspective. Finally, the training stimulus was provided
476 with an even distribution of the number of contractions across both slow and fast isokinetic
477 velocities. As vastus lateralis architectural adaptations have been shown to be velocity
478 dependent (33), it is not possible to determine if the changes in this cohort and muscle are due
479 to the velocities utilised. The aim of this study was to investigate the effect of contraction
480 mode, not velocity, on BFlf architectural changes as this may have greater implications for
481 hamstring strain injury prevention and rehabilitation. Further research is needed to determine
482 if there is a contraction velocity-specific adaptation in the knee flexors following a
483 concentric- or eccentric-strength training intervention.

484 In conclusion, the current study reports rapid, contraction-mode specific alterations in BFlf
485 architecture following 6 weeks of either eccentric or concentric strength training
486 interventions. Further, 28 days of detraining resulted in BFlf architectural characteristics
487 returning to baseline levels in individuals who had completed eccentric training, whilst
488 detraining had no influence on the BFlf architectural characteristics in those who completed
489 concentric strength training. The findings of the current study provide insight into BFlf
490 architectural alterations following concentric and eccentric strength training interventions.
491 These results may have implications for hamstring injury prevention and rehabilitation
492 programs which might consider architectural alterations to training interventions as a factor
493 that might mitigate risk of future injury.

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495 N/A

496 **CONFLICT OF INTEREST**

497 The authors report that this study was not funded at that no conflict of interest exists. Results
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501 **REFERENCES**

- 502 1. Abe T, Kawakami Y, Suzuki Y, Gunji A, Fukunaga T. Effects of 20 days bed rest on
503 muscle morphology. *J Gravit Physiol.* 1997;4(1):S10-4.
- 504 2. Blazevich AJ. Effects of physical training and detraining, immobilisation, growth and
505 aging on human fascicle geometry. *Sports Med.* 2006;36(12):1003-17.
- 506 3. Blazevich AJ, Cannavan D, Coleman DR, Horne S. Influence of concentric and
507 eccentric resistance training on architectural adaptation in human quadriceps muscles. *J Appl*
508 *Physiol (1985).* 2007;103(5):1565-75.
- 509 4. Blazevich AJ, Gill ND, Bronks R, Newton RU. Training-specific muscle architecture
510 adaptation after 5-wk training in athletes. *Med Sci Sports Exerc.* 2003;35(12):2013-22. Epub
511 2003/12/04. doi: 10.1249/01.MSS.0000099092.83611.20.
- 512 5. Blazevich AJ, Gill ND, Zhou S. Intra- and intermuscular variation in human
513 quadriceps femoris architecture assessed in vivo. *J Anat.* 2006;209(3):289-310.
- 514 6. Bodine SC, Roy RR, Meadows DA, et al. Architectural, histochemical, and
515 contractile characteristics of a unique biarticular muscle: the cat semitendinosus. *J*
516 *Neurophysiol.* 1982;48(1):192-201.

- 517 7. Brockett CL, Morgan DL, Proske U. Predicting hamstring strain injury in elite
518 athletes. *Med Sci Sports Exerc.* 2004;36(3):379-87.
- 519 8. Cohen D. *Statistical power analysis for the behavioral sciences.* Hillsdale (NJ):
520 Erlbaum; 1988. p 75.
- 521 9. Faul F, Erdfelder E, Lang AG, Buchner A. G*Power 3: a flexible statistical power
522 analysis program for the social, behavioral, and biomedical sciences. *Behav Res Methods.*
523 2007;39(2):175-91.
- 524 10. Franchi MV, Atherton PJ, Reeves ND, et al. Architectural, functional and molecular
525 responses to concentric and eccentric loading in human skeletal muscle. *Acta Physiol (Oxf).*
526 2014;210(3):642-54.
- 527 11. Fyfe JJ, Opar DA, Williams MD, Shield AJ. The role of neuromuscular inhibition in
528 hamstring strain injury recurrence. *J Electromyogr Kinesiol.* 2013;23(3):523-30.
- 529 12. Guex K, Millet GP. Conceptual framework for strengthening exercises to prevent
530 hamstring strains. *Sports Med.* 2013;43(12):1207-15.
- 531 13. Kaminski TW, Wabbersen CV, Murphy RM. Concentric versus enhanced eccentric
532 hamstring strength training: clinical implications. *J Athl Train.* 1998;33(3):216-21.
- 533 14. Kellis E, Galanis N, Natsis K, Kapetanios G. Validity of architectural properties of the
534 hamstring muscles: correlation of ultrasound findings with cadaveric dissection. *J Biomech.*
535 2009;42(15):2549-54.
- 536 15. Klimstra M, Dowling J, Durkin JL, MacDonald M. The effect of ultrasound probe
537 orientation on muscle architecture measurement. *J Electromyogr Kinesiol.* 2007;17(4):504-
538 14.
- 539 16. Koulouris G, Connell DA, Brukner P, Schneider-Kolsky M. Magnetic resonance
540 imaging parameters for assessing risk of recurrent hamstring injuries in elite athletes. *Am J*
541 *Sports Med.* 2007;35(9):1500-6.

- 542 17. Lieber RL, Ward SR. Skeletal muscle design to meet functional demands. *Philos*
543 *Trans R Soc Lond B Biol Sci.* 2011;366(1570):1466-76.
- 544 18. Lynn R, Morgan DL. Decline running produces more sarcomeres in rat vastus
545 intermedius muscle fibers than does incline running. *J Appl Physiol (1985).* 1994;77(3):1439-
546 44.
- 547 19. Morgan DL. New insights into the behavior of muscle during active lengthening.
548 *Biophys J.* 1990;57(2):209-21.
- 549 20. Narici M, Cerretelli P. Changes in human muscle architecture in disuse-atrophy
550 evaluated by ultrasound imaging. *J Gravit Physiol.* 1998;5(1):P73-4.
- 551 21. Narici MV, Flueck M, Koesters A, et al. Skeletal muscle remodeling in response to
552 alpine skiing training in older individuals. *Scand J Med Sci Sports.* 2011;21 Suppl 1:23-8.
- 553 22. Noorkoiv M, Nosaka K, Blazevich AJ. Neuromuscular adaptations associated with
554 knee joint angle-specific force change. *Med Sci Sports Exerc.* 2014;46(8):1525-37.
- 555 23. Noorkoiv M, Stavnsbo A, Aagaard P, Blazevich AJ. In vivo assessment of muscle
556 fascicle length by extended field-of-view ultrasonography. *J Appl Physiol (1985).*
557 2010;109(6):1974-9.
- 558 24. Opar D, Williams M, Timmins R, Hickey J, Duhig S, Shield A. Eccentric hamstring
559 strength and hamstring injury risk in Australian Footballers. *Med Sci Sports Exerc.*
560 2015;47(4):857-65.
- 561 25. Opar DA, Piatkowski T, Williams MD, Shield AJ. A novel device using the nordic
562 hamstring exercise to assess eccentric knee flexor strength: a reliability and retrospective
563 injury study. *J Orthop Sports Phys Ther.* 2013;43(9):636-40.
- 564 26. Orchard JW, Seward H, Orchard JJ. Results of 2 decades of injury surveillance and
565 public release of data in the Australian Football League. *Am J Sports Med.* 2013;41(4):734-
566 41.

- 567 27. Paddon-Jones D, Leveritt M, Lonergan A, Abernethy P. Adaptation to chronic
568 eccentric exercise in humans: the influence of contraction velocity. *Eur J Appl Physiol.*
569 2001;85(5):466-71.
- 570 28. Potier TG, Alexander CM, Seynnes OR. Effects of eccentric strength training on
571 biceps femoris muscle architecture and knee joint range of movement. *Eur J Appl Physiol.*
572 2009;105(6):939-44.
- 573 29. Reeves ND, Maganaris CN, Longo S, Narici MV. Differential adaptations to eccentric
574 versus conventional resistance training in older humans. *Exp Physiol.* 2009;94(7):825-33.
- 575 30. Selvanayagam VS, Riek S, Carroll TJ. Early neural responses to strength training. *J*
576 *Appl Physiol (1985).* 2011;111(2):367-75.
- 577 31. Seynnes OR, de Boer M, Narici MV. Early skeletal muscle hypertrophy and
578 architectural changes in response to high-intensity resistance training. *J Appl Physiol (1985).*
579 2007;102(1):368-73.
- 580 32. Seynnes OR, Maganaris CN, de Boer MD, di Prampero PE, Narici MV. Early
581 structural adaptations to unloading in the human calf muscles. *Acta Physiol (Oxf).*
582 2008;193(3):265-74.
- 583 33. Sharifnezhad A, Marzilger R, Arampatzis A. Effects of load magnitude, muscle
584 length and velocity during eccentric chronic loading on the longitudinal growth of the vastus
585 lateralis muscle. *J Exp Biol.* 2014;217(Pt 15):2726-33.
- 586 34. Shepstone TN, Tang JE, Dallaire S, Schuenke MD, Staron RS, Phillips SM. Short-
587 term high- vs. low-velocity isokinetic lengthening training results in greater hypertrophy of
588 the elbow flexors in young men. *J Appl Physiol (1985).* 2005;98(5):1768-76.
- 589 35. Thelen DG, Chumanov ES, Hoerth DM, et al. Hamstring muscle kinematics during
590 treadmill sprinting. *Med Sci Sports Exerc.* 2005;37(1):108-14.

- 591 36. Timmins R, Shield A, Williams M, Lorenzen C, Opar D. Biceps femoris long head
592 architecture: a reliability and retrospective injury study. *Med Sci Sports Exerc.*
593 2015;47(5):905-13.
- 594 37. Timmins RG, Opar DA, Williams MD, Schache AG, Dear NM, Shield AJ. Reduced
595 biceps femoris myoelectrical activity influences eccentric knee flexor weakness after repeat
596 sprint running. *Scand J Med Sci Sports.* 2014;24(4):e299-e305.
- 597 38. Yu B, Queen RM, Abbey AN, Liu Y, Moorman CT, Garrett WE. Hamstring muscle
598 kinematics and activation during overground sprinting. *J Biomech.* 2008;41(15):3121-6.
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600 Figure 1: A two dimensional ultrasound image of the biceps femoris long head. This image of
601 the biceps femoris long head was taken along the longitudinal axis of the posterior thigh.
602 From these images it is possible to determine the superficial and intermediate aponeuroses,
603 muscle thickness, angle of the fascicle in relation to the aponeurosis. Estimates of fascicle
604 length can then be made via trigonometry using muscle thickness and pennation angle.

605 Figure 2: Changes in the architectural characteristics of the BFlf when assessed at rest in the
606 trained limb and the contralateral untrained limb of both groups following 14, 21 and 42 days
607 of the training intervention and following the detraining period (*day 70*). A) fascicle length
608 B) pennation angle C) muscle thickness D) fascicle length relative to muscle thickness. Error
609 bars illustrate the standard deviation. *=p<0.05 vs Day 0, ** = p<0.001 vs Day 0, ## =
610 p<0.001 vs Day 42.

611

612 Table 1: Changes in the BFlf architectural characteristics in the training limb of each group at
613 the start (day 0), after 14, 21 and 42 days of the training intervention as well as following the
614 detraining period (day 70). All data represented as mean±SD unless otherwise stated. SD =
615 standard deviation, MT = muscle thickness, cm = centimetres, PA = pennation angle, RFL =
616 fascicle length relative to muscle thickness, FL = fascicle length, MVIC = maximum
617 voluntary isometric contraction. *=p<0.05 vs Day 0, ** = p<0.001 vs Day 0, # = p<0.05 vs
618 Day 42, ### = p<0.001 vs Day 42.

619 Table 2: Changes in concentric and eccentric knee flexor peak torque at various contraction
620 velocities in the training limb of each group before (day 0) and after the training intervention
621 (day 42) as well as following the detraining period (day 70). All data represented as
622 mean±SD unless otherwise stated. SD = standard deviation, °/s = degrees per second.
623 *=p<0.05 vs Day 0, ** = p<0.001 vs Day 0.

