

1 Patellar tendon in vivo regional strain with varying knee angle

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3 Stephen J. Pearson<sup>1</sup>, Azlan S. A. Mohammed<sup>2</sup>, Syed R Hussain<sup>1</sup>

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5 <sup>1</sup>Centre for Health, Sport and Rehabilitation Sciences Research, University of Salford,

6 Greater Manchester, UK

7 <sup>2</sup>School of Computer Sciences, Universiti Sains Malaysia (USM), 11800, Penang, Malaysia

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9 Address for correspondence: Stephen J. Pearson, Ph.D., Sport, Exercise &

10 Physiotherapy, University of Salford, Manchester M66PU, United Kingdom;

11 E-mail: s.pearson@salford.ac.uk

12 Tel: 01612952673

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20 **ABSTRACT**

21 **Purpose:** Descriptive data on the aspects of site specific in vivo tendon strain with varying  
22 knee joint angle are non-existent. The present study determines and compares surface and  
23 deep layer strain of the patellar tendon during isometric contractions across a range of knee  
24 joint angles.

25 **Methods:** Male participants (age  $22.0 \pm 3.4$ ) performed ramped isometric knee extensions at  
26 knee joint angles of  $90^0$ ,  $70^0$ ,  $50^0$  and  $30^0$  of flexion. Strain patterns of the anterior and  
27 posterior regions of the patellar tendon were determined using real-time B-mode  
28 ultrasonography at each knee joint angle. Regional strain measures were compared using an  
29 automated pixel tracking method.

30 **Results:** Strain was seen to be greatest for both the anterior and posterior regions with the  
31 knee at  $90^0$  ( $7.76 \pm 0.89\%$  and  $5.06 \pm 0.76\%$ ). Anterior strain was seen to be significantly  
32 greater ( $p < 0.05$ ) than posterior strain for all knee angles apart from  $30^0$ ,  $90^0 = (7.76$  vs.  
33  $5.06\%)$ ,  $70^0 = (4.77$  vs.  $3.75\%)$ , and  $50^0 = (3.74$  vs.  $2.90\%)$ . The relative strain (ratio of  
34 anterior to posterior), was greatest with the knee joint angle at  $90^0$ , and decreased as the knee  
35 joint angle reduced.

36 **Conclusions:** The results from this study indicate that not only are there greater absolute  
37 tendon strains with the knee in greater flexion, but that the knee joint angle affects the  
38 regional strain differentially, resulting in greater shear between the tendon layers with force  
39 application when the knee is in greater degrees of flexion. These results have important  
40 implications for rehabilitation and training.

41

42 **Key Words:** Tendon; localised Strain; Patella; Knee extension; Isometric

## 43 INTRODUCTION

44 The tendon structure in humans is described as viscoelastic in nature and is capable of  
45 transferring high loads to the bony structures to enable stabilisation, movement and  
46 locomotion. Although this structure is strong and generally able to withstand high transient  
47 loads, it is at risk of both chronic and acute injury. With this high loading, there is potential  
48 for acute injury to the tendon structure due to unaccustomed high loading rates or  
49 unstable/unequal loading (i.e. landing from a jump) (Richards et al., 1996). Injuries to the  
50 structures around the knee are common in sport, with those sports involving jumping  
51 particularly prone to tendon overload injuries (Van der worp et al., 2011; Zwerver et al.,  
52 2011; Visnes et al., 2014).

53 Repeated chronic overload or under load of the tendon could lead to tendinopathic-type  
54 changes, resulting in inflammatory or degenerative tendon conditions (Leadbetter 1992; Riley  
55 2008; Rufai et al., 1995; Vogel et al., 1993). In particular, athletes show characteristic issues  
56 related to the tendon whereby the structure can either become inflamed or degenerative.

57 These conditions which are thought to be more chronic are termed 'tendinopathies' (Astrom  
58 and Rausing, 1995; Kader et al., 2002; Maffulli and Kader, 2002; Regan et al., 1992). A  
59 number of factors may be responsible for the development of such conditions, including  
60 inappropriate training loads or volumes (Korkia et al., 1994), improper application of  
61 technique or equipment (Ilfeld, 1992; James, 1995; Kibler et al., 1992), and imbalance of the  
62 contractile and supporting soft tissue system (Almekinders, 1998; Herring and Nilsen, 1987).

63 With respect to insertional tendinopathies, a biomechanical mechanism has been put forward  
64 termed 'stress shielding' (Orchard et al., 2004), whereby the posterior patellar tendon is  
65 'shielded' from the loading experienced by the anterior tendon. Here it is suggested that the  
66 'lack' of loading at the posterior region of the tendon may in fact result in cartilaginous or

67 atrophic type changes over time (Rufai et al., 1995; Vogel et al., 1993). It could also be  
68 suggested that as the knee moves through its range of motion, the tendon is differentially  
69 exposed to load throughout its cross-section, thus resulting in shear type stressors, causing  
70 micro damage which could accumulate to a point where tangible injury to the structure is  
71 evident (Riley, 2008). Previous work on both the Achilles and patellar tendons in cadaveric  
72 specimens has shown that differential strain exists in these structures when loaded  
73 (Almekinders et al., 2002; Basso et al., 2002; Lersch et al., 2012), although these studies do  
74 not have consensus on the patterns of differential loading across the tendon. Cadaveric  
75 specimens however, do not always represent valid measures for structures in living  
76 individuals for a number of reasons including changes in the specimens after death,  
77 alterations in the structures due to preparation and testing limitations (slipping of tissues in  
78 holding clamps). More recently, using automated non-invasive tracking via ultrasound  
79 imaging, it has been reported in humans that there are in vivo differences in regional strain  
80 for the patellar tendon during loading (Arndt et al., 2012; Pearson et al., 2014), indicating that  
81 shear is present in the tendon structure when loaded in vivo. However, to date there is no data  
82 on the potential for differential strain in vivo of the patellar tendon over a range of knee  
83 angles, something which occurs normally during movement. As the patellar tendon shows a  
84 change in the moment arm across the range of knee angles (Krevolin et al., 2004; Tsaopoulos  
85 et al., 2006), it may be suggested that the localised strains seen previously at 90<sup>0</sup> of knee  
86 flexion (Pearson et al., 2014) may alter with knee joint angle (See figure 5 for illustration of  
87 moment turning effect).

88 Therefore, the purpose of the current study was to quantify and compare the regional strain  
89 patterns (anterior/posterior) of the patellar tendon using a previously described speckle  
90 tracking method over a range of knee angles during maximal isometric contractions. It was  
91 hypothesised that there would be differences in strain with changing knee angle, and that the

92 ratio of anterior to posterior strain would change with knee angle. The information from this  
93 study will enable a more realistic and complete understanding of tendon strain behaviour  
94 during loading over a range of joint angles, thus informing the development of rehabilitation,  
95 training and injury prevention strategies.

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## 99 **METHODS**

### 100 **Subjects and experimental design:**

101 Ten healthy male participants (aged  $22.0 \pm 3.4$ , height  $1.75 \pm 0.03\text{m}$ , body mass  $80 \pm 4.3$  kg)  
102 took part in the cross-sectional study. The investigation was approved by the local ethics  
103 committee and all participants gave their written informed consent to take part. The study  
104 conformed to the principles of the World Medical Association's Declaration of Helsinki. All  
105 participants had a familiarisation session in the laboratory prior to any testing.

### 106 **Methods**

107 A 7.5 MHz 100mm linear array B mode ultrasound probe (Mylab 70, Esaote Biomedica,  
108 Italy) with a depth resolution of 67mm was used to image the patellar tendon in the sagittal  
109 plane (ensuring both bone ends were in view - patella and tibia), with the knee joint fixed at a  
110 range of flexion angles ( $90^0$ ,  $70^0$ ,  $50^0$  and  $30^0$ ). Ultrasound images were captured at 25Hz for  
111 later processing. All scaling in pixels per millimetre was determined from image J software  
112 (NIH), by using the known depth of field in the ultrasound images ( $1\text{mm} = 11$  pixels in the x

113 and y directions), and used as a calibration factor in the automated tracking programme to  
114 ensure equivalent pixel to mm ratios.

115 Subjects were then required to perform ramped isometric knee extension contractions and  
116 simultaneous torque outputs were recorded using a dynamometer (Type 125 AP; Kin Com,  
117 Chattanooga, TN) with the participant in a seated position. A series of three ramped  
118 contractions (180 seconds rest between each) were carried out at each of the four knee angles  
119 ( $90^{\circ}$ ,  $70^{\circ}$ ,  $50^{\circ}$ ,  $30^{\circ}$ , full extension =  $0^{\circ}$ ), with the hip fixed at  $85^{\circ}$ , (supine =  $0^{\circ}$ ). Torque was  
120 transmitted via a cuff and lever attachment placed at the lower leg ~ 3 cm above the medial  
121 malleolus. Leg mass (gravity correction) at all angles apart from  $90^{\circ}$  was calculated and  
122 added or subtracted from the output extension or flexion forces respectively (horizontal force  
123 (leg mass)  $\times$   $\cos \theta$  knee angle), and converted to torque via multiplication of the level arm.  
124 Prior to any efforts. three maximal isometric conditioning contractions were carried out.  
125 Instructions were given to develop the ramped isometric tension from rest to maximal over a  
126 period of 3-4 seconds. The mean value of strain from each series of three contractions at each  
127 knee angle were used for later analysis. To enable the torque to be synchronised with the  
128 ultrasound output, a square wave signal generator was used.

129 The electrical activity of the long head of the biceps femoris (BF) muscle was also measured  
130 via electromyography (EMG) to ascertain the level of antagonistic muscle co-contraction  
131 during the isometric knee extension efforts (Pearson and Onambele, 2006). This was in turn  
132 used to adjust the net knee extension torque to calculate total torque (see below). The long  
133 head of the BF has previously been shown to be representative of the hamstrings group  
134 (Carolan and Cafarelli, 1985) and the relationship between BF torque and EMG reported to  
135 be generally linear (Lippold, 1952). In brief, the maximal flexion EMG was determined from  
136 a series of three knee flexion efforts. Then the root mean square activity corresponding to the  
137 peak torque period was analysed over 50 ms epochs and averaged for 1 sec during the plateau

138 of the isometric maximal effort, this was carried out at each tested knee angle. This has  
139 previously been suggested to be acceptable in terms of signal-to-noise ratio (Hermens et al.,  
140 1999). Any subsequent EMG of the BF during maximal knee extension efforts was divided  
141 by the maximal flexor torque EMG, and the maximal flexor torque was then multiplied by  
142 this value to determine co-contraction torque. Patellar tendon forces were then determined by  
143 dividing the total torque by the patellar tendon lever arm, determined from the literature  
144 (Krevolin et al., 2004). All grayscale ultrasound images gave regional attributes of  
145 dimension, position, coordinates and pixel grayscale values. In all compared frames the  
146 coordinates of the region of interest (ROI) in the frame were offset along the horizontal and  
147 vertical image planes and shifted by a pixel at a time to determine the degree of match.

148

#### 149 **Tracking algorithm**

150 The algorithm used for tracking the ROI in the grayscale images has been described  
151 previously (Pearson et al., 2014). Briefly, a block matching method with normalised cross  
152 correlation (NCC) was utilised to determine similarity in the ROI between subsequent frames  
153 (images collected from video recorded at 25 Hz from start i.e. resting to maximum  
154 contraction force and split into subsequent frames for analysis). Here the ROI were arranged  
155 in two layers (Fig 1), with ROI 1 at 10mm from the patellar pole and the ROI 2 resting inter  
156 distance at 30mm. These ROI were then utilised to enable relative movement from the initial  
157 start, enabling strain to be determined for each layer region. As previously (Pearson et al.,  
158 2014), the ROI block size used was 15x15 pixels and the search window was 2x ROI (width)  
159 and 1x ROI size (height). For each frame search window comparison, the ROI were displaced  
160 by 1 pixel at a time from the original ROI start in the previous frame and compared using  
161 NCC. These data were then stored in a matrix for determination of best match based on the

162 highest correlation values. The threshold level of correlation was set to  $\geq 0.9$ . If this value  
 163 was not reached or exceeded, the ROI was not shifted in the subsequent frame. If a  
 164 correlation value above the threshold was detected, then the tracking resets the matching  
 165 template and starts with the new updated position of the template ROI block in the next frame  
 166 (Equation 1).

167 [Fig 1 near here]

$$\rho_{nm(k,l)} = \frac{\sum_{i=-K}^K \sum_{j=-L}^L [F_1(n+i, m+j)F_2(n+k+i, m+l+j)]}{\left[ \sum_{i=-K}^K \sum_{j=-L}^L [F_1(n+i, m+j)]^2 \right]^{1/2} \left[ \sum_{i=-K}^K \sum_{j=-L}^L [F_2(n+k+i, m+l+j)]^2 \right]^{1/2}}$$

168

169 [1]

170 Where F1 is the start frame and F2 is the subsequent frame. The value  $(n, m)$  represents the  
 171 image block centre, with the sum being over  $(i, j)$ , with  $(k, l)$  being the lateral and axial  
 172 displacements respectively. The normalised cross correlation value (1 to -1, with 1 being the  
 173 best match) is represented by  $\rho_{nm(k, l)}$ . The algorithm calculates this based on the two  
 174 regions of interest ( $R_1$  and  $R_2$ ), (Fig 1), which are tracked from the first to the last frame, with  
 175 the determined differences between the two ROI recorded up to maximum (100% MVC).

176 Three trials were utilised to determine the means for each knee effort. Subsequent measures  
 177 are anterior excursion and posterior excursion with knee angles  $(90 - 30^0)$ . The total  
 178 displacements of  $x$  and  $y$  over the frames is from determination of  $R_1$  and  $R_2$  measured from  
 179 the first frame ( $f_1$ ) to the last frame ( $f_n$ ), with the resultant displacement as final frame -  
 180 initial frame reference (Equation 2). Strain, therefore, is the displacement divided by the  
 181 initial measure between  $R_1$  and  $R_2$ .

$$\text{Disp} = \left( \sqrt{(x_{k2} - x_{k1})^2 + (y_{k2} - y_{k1})^2} \right)_{f0} - \left( \sqrt{(x_{k2} - x_{k1})^2 + (y_{k2} - y_{k1})^2} \right)_{f1}$$

182  
183 [2]

184

## 185 **Statistics**

186 Intraclass coefficient correlations (ICC's) were utilised to determine the reliability of all  
187 measures. This was carried out by using repeated tracking on the regions during the ramped  
188 isometric contractions. Anterior vs. posterior strain comparisons by knee angle were made  
189 using repeated measures two-way ANOVA and Bonferroni *post hoc* pairwise tests. Paired t  
190 tests were utilised to compare strain ratio values at each knee angle and at fixed levels of  
191 force. The alpha level for all tests was set at 0.05. Statistical power was determined using G  
192 power (3.09, Franz Faul, Universitat, Kiel, Germany). For a power (1-β) of 0.8 and moderate  
193 effect size (0.4) a sample size of ten was determined.

194

## 195 **RESULTS**

### 196 **Reliability**

197 Pooled repeated displacement tracking analyses for the images of both layers gave an ICC of  
198 0.88. The determined maximal mean percentage of hamstrings co-contraction (BF activity  
199 during maximal isometric knee extension), was seen to be  $7.50 \pm 0.30$  %, which indicated a  
200 relatively low hamstring involvement during the maximal isometric agonist efforts.

### 201 **Anterior vs. Posterior Strain**

202 Tendon strain comparisons between the anterior and posterior regions showed significant  
203 differences pooled across the range of knee angles (anterior =  $4.74 \pm 1.95\%$ ; posterior =  $3.57$   
204  $\pm 1.08\%$ ), ( $p < 0.05$ ;  $F(1, 9) = 89.81$ ;  $\eta_p^2 = 0.90$ ; power = 1), with both regions declining as  
205 the knee joint angle decreased towards  $30^\circ$  (Figure 2), in line with the reductions in MVC  
206 force. Pooled strain at each knee angle showed significant differences with change in knee  
207 angle ( $90^\circ = 6.38 \pm 1.58\%$ ;  $70^\circ = 4.19 \pm 0.73\%$ ,  $50^\circ = 3.34 \pm 0.56\%$ ,  $30^\circ = 2.71 \pm 0.47\%$ ), ( $p$   
208  $< 0.05$ ;  $F(1.56, 14.10) = 92.95$ ;  $\eta_p^2 = 0.91$ ; power = 1).

209 The interaction of knee angle with region was also seen to be significant ( $p < 0.05$ ;  $F(1.83,$   
210  $16.50) = 24.40$ ;  $\eta_p^2 = 0.73$ ; power = 1), indicating differential strain by region at different  
211 knee angles. This was reflected in the strain ratios determined below.

212 [Figure 2 near here]

### 213 **Regional strain ratios**

214 The anterior region was found to exhibit significantly higher strain values than the posterior  
215 region at all angles of flexion  $90^\circ$  ( $7.76\% \pm 0.89$  vs.  $5.06\% \pm 0.76$ ,  $t(9) = 10.78$ ,  $p < 0.001$ ,  $r$   
216  $= 0.96$ ),  $70^\circ$  ( $4.77\% \pm 0.66$  vs.  $3.75\% \pm 0.38$   $t(9) = 3.10$ ,  $p = 0.013$ ,  $r = 0.71$ ), and  $50^\circ$   
217 ( $3.74\% \pm 0.37$  vs.  $2.90\% \pm 0.25$ , ( $t(9) = 6.40$ ,  $p < 0.001$ ,  $r = 0.90$ ) apart from  $30^\circ$  ( $2.84\% \pm$   
218  $0.53$  vs.  $2.57\% \pm 0.31$ ,  $p = 0.84$ ,  $r = 0.54$ ) (Figure 2). Moreover, the magnitude of differences  
219 in strain between the anterior and posterior regions (anterior-to-posterior strain ratio) was  
220 greatest at  $90^\circ$  and lowest at  $30^\circ$ , with the regional values being very similar at  $30^\circ$  knee  
221 flexion (Figure 3).

222 To account for differences in MVC force with change in knee angle, a standardised force  
223 level of 2117N (corresponding to the mean maximal force at  $30^\circ$  of knee flexion) was used to  
224 compare the anterior and posterior data sets. Once again, tendon strain was found to be higher

225 in the anterior region compared to the posterior region at all knee joint angles, and the  
226 anterior-to-posterior strain ratio was greatest at 90<sup>0</sup> of flexion and lowest at 30<sup>0</sup> of flexion.  
227 (Figure 4). Comparisons of pooled anterior vs. posterior strain at the fixed level of force  
228 across the knee angles showed a significant difference ( $2.96 \pm 0.43\%$ ;  $2.35 \pm 0.50\%$ ), ( $t(3) =$   
229  $4.27$ ,  $p = 0.02$ ,  $r = 0.92$ ).

230 [Figure 3 near here]

231 [Figure 4 near here]

232

233

234 **DISCUSSION**

235 To the authors knowledge this is the first study to report in vivo differential longitudinal  
236 strain in the patellar tendon with change in knee joint angle. The study examined the regional  
237 strain patterns of the patellar tendon during ramped isometric maximal contractions, over a  
238 range of knee joint angles. It was hypothesised that there would be differential intra-  
239 tendinous strain patterns, as well as variations in the anterior-to-posterior strain ratio with  
240 changes in knee joint angle. The findings of the present study have confirmed these  
241 hypotheses.

242 Tensile strain was found to be significantly greater ( $p < 0.05$ ) in the anterior region than the  
243 corresponding posterior region at the knee joint angles of  $90^0$ ,  $70^0$  and  $50^0$ . In addition, the  
244 anterior-to-posterior strain ratio (magnitude of differences in strain between anterior and  
245 posterior regions) was greatest at  $90^0$  (1.53) of knee flexion and lowest with the knee at  $30^0$  of  
246 flexion (1.10). These data add to previous observations showing intra-tendinous variations in  
247 loading patterns within the patellar tendon, and extend these observations by showing for the  
248 first time that the patellar tendon exhibits differential intra-tendinous strain characteristics  
249 specific to the knee joint angle at which loading is applied.

250 The mean maximal strain values of the patellar tendon reported here with the knee at  $90^0$  of  
251 flexion ( $7.76\% \pm 0.89$ ) are in general agreement with previous work (Hansen et al., 2006;  
252 Malliaras et al., 2013; Pearson et al., 2014), who reported strains of  $6.9 \pm 0.6$ , 9-12% and 7.5-  
253 7.9% respectively . The higher strain reported by Malliaras and co workers may in part be  
254 due to subject characteristics i.e. they may have been initially untrained and so likely to have  
255 more compliant tendons. However strain with the knee at fixed flexion angles less than  $90^0$   
256 as reported, here have not previously been examined in living specimens.

257 Previous research relating to the strain patterns of the anterior and posterior regions of the  
258 patellar tendon under load is diverse. Some authors have found the anterior region to display  
259 a significantly greater level of tensile strain relative to the posterior region (Almekinders et  
260 al., 2002; Pearson et al., 2014), whereas others have reported the posterior region to exhibit a  
261 greater magnitude of strain than the anterior region (Basso et al., 2002; Dillon et al., 2008).  
262 Despite such variability, which is perhaps, in part, attributable to inter-study differences in  
263 methodologies including the knee joint angle utilised , and also living vs. dead tissues (in  
264 vivo, vs. in vitro), the findings of the present study are in general agreement with those of  
265 Almekinders et al. (2002) and Pearson et al. (2014). Using real-time ultrasonography and  
266 dynamometry, Pearson and co workers reported a greater level of tensile strain in the anterior  
267 region compared to the posterior region at a knee joint angle of 90<sup>0</sup>. Similarly, Almekinders  
268 and co-workers utilised cadaver knee specimens instrumented with strain gauges and  
269 reported tendon strain to increase at the anterior region and decrease at the posterior region as  
270 the knee was brought into flexion, although it is noteworthy that a compression of posterior  
271 tensile strain was not observed in the present study, perhaps due to the more complex  
272 interplay in living systems (antagonist/agonist muscle action and tissue characteristic  
273 differences) and the applied forces.

274 More specifically, in the present study, tendon strain values were found to be greater at the  
275 anterior region compared to the posterior region throughout the entire functional flexion  
276 range, with significantly higher values at all level of flexion apart from 30<sup>0</sup>. These findings  
277 suggest that the posterior region of the patellar tendon is relatively stress shielded which  
278 could result in deterioration over time (Rufai et al., 1995; Vogel et al., 1993), leading to  
279 reduced mechanical strength and increased potential for injury. In line with this notion,  
280 previous studies on isolated patellar tendon fascicles have also shown decreased mechanical  
281 strength and stiffness at the posterior region compared to the anterior region (Hansen et al.,

282 2010; Haraldsson et al., 2010), thus highlighting the lack of functional adaptation and  
283 weaker, more injury prone state of the posterior region.

284 Interestingly, the present study also found that the magnitude of differences in strain between  
285 the anterior and posterior regions was dependent on the knee joint angle at which loading was  
286 applied, with the greatest relative difference observed at 90<sup>0</sup> of flexion (1.53) and lowest at  
287 30<sup>0</sup> of flexion (1.10). Such differential intra-tendinous strain patterns with the change in knee  
288 joint angle may be, in part, a factor of the alterations in the patellar lever arm arrangement  
289 with respect to the 'angle of pull' across the bony ends of the proximal and distal connections  
290 of the patellar tendon. Indeed, work by DeFrate et al. (2007) and Aalsberg et al. (2005), using  
291 MRI show changes in the patellar line of pull relative to the tibia with changes in knee  
292 flexion, suggesting a more 'posterior' pull on the tibia with increased knee flexion angle. It  
293 could be reasoned therefore that the alterations in the tendon alignment with respect to the  
294 bony anchors, and the patella with respect to the pull of the quadriceps muscle may result in  
295 changes on the intrinsic elements of the tendon resulting in differential regional strain as seen  
296 here. In addition, the notion that the turning moment of the patella increases with increased  
297 knee flexion is supported by the study of Ward et al. (2012). Here, fluoroscopic images  
298 showed an increase in the patella flexion angle with increased knee flexion such that the pull  
299 of the quadriceps against the patella becomes more angular to the patellar tendon (see figure  
300 5 for illustration). One could thus envisage how this turning moment (action to lift the patella  
301 bone away from the patellar tendon as it glides around the intercondylar fossa of the femur),  
302 would generate more strain at the anterior aspect of the patellar tendon.

303 [Figure 5 near here]

304 The regional strain patterns observed in the present study are indeed indicative of shear stress  
305 within the patellar tendon during applied loading conditions, which is greatly implicated in

306 the development and progression of patellar tendinopathy (Almekinders et al., 2002). The  
307 increased magnitude of tensile strain observed in the anterior region may suggest that it could  
308 ultimately reach a stage where it can no longer bear the functional demand and, as a result,  
309 becomes tendinopathic via 'overuse'. On the other hand, the stress-shielded nature and lack  
310 of functional adaptation of the posterior region could also play an important role in the  
311 development of tendinopathy, simply owing to its weaker state (Almekinders et al., 2002;  
312 Orchard et al., 2004). Both these hypotheses can be seen to have some rationale here, i.e.  
313 unaccustomed loading of a 'relatively' weaker area of the tendon may result in some kind of  
314 insult to the structure, especially where patellar loading is carried out with the knee in  
315 flexion.

316

317 Interestingly, changes in transverse strain with loading observed by Wearing and co workers  
318 (2013) may be reflective of regional differences in longitudinal strain and it would be useful  
319 if measures of transverse strain could be more selective of layer or regions in order to enable  
320 relationships to longitudinal regional strain to be made.

### 321 **Practical applications:**

322 The findings here indicate that shear strain of the patellar tendon increases with increased  
323 knee flexion angle. Hence to enable a progressive return to play in sport where tendon injury  
324 has occurred it is recommended to begin loading with a reduced knee flexion angle and  
325 progress this in conjunction with load over time. Similarly to minimise effects of training  
326 induced damage, a range of angles should be utilised when preparing this structure for play,  
327 limiting maximal knee flexion wherever possible.

### 328 **Conclusions:**

329 In conclusion, we have shown that by varying the knee flexion angle we not only observe  
330 changes in absolute patellar tendon strain, but that the ratio between anterior and posterior  
331 tendon strain is also altered. The understanding that shear inducing difference between the  
332 surface and deep layers of the tendon occur differentially with knee angle may have  
333 implications for practices to help prevent tendon related injury and also aid targeted  
334 rehabilitation strategies.

335 The authors wish to acknowledge potential limitations with the current methodology, such  
336 that ROI identification and hence strain patterns during subsequent loading may be affected  
337 by out of plane movement. However using the current technology it is not possible to assess  
338 this effectively. We also acknowledge that the work presented here is in 2D and that future  
339 work would aim to extend this to a 3D scenario to detail further the characteristics of tendon  
340 strain under loading.

341

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483 Figure legends

484 Figure 1. Showing an example ultrasound image of the patellar tendon. Overlaid are the  
485 regions of interest seen as red squares (ROI), tracked with increasing force production. It can  
486 be seen that the ROI move laterally along the image with subsequent force production up  
487 until maximum for both anterior and posterior layers.

488

489 Figure 2. Mean values ( $\pm$  SD) of maximal strain values for all measured regions at each knee  
490 angle. (■) anterior region and (□) posterior region. \* Significantly different  $p < 0.05$ ,  
491 (anterior vs. posterior). ^ significantly different  $p < 0.05$ , (pooled strain).

492

493 Figure 3. Strain ratio determined at maximal strain for both anterior and posterior regions for  
494 each knee angle.

495

496 Figure 4. Strain ratio determined at a fixed force of 2117N (mean maximal force at knee  
497 angle of  $30^{\circ}$ ), at each knee angle.

498

499 Figure 5. Illustration showing potential turning effect of patella on tendon with knee flexed.