1	Effects of shoulder position during static stretching
2	on shear elastic modulus of biceps brachii muscle
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19 Abstract

20 Biceps brachii muscle consists of a long head (BBL) and a short head (BBS). Shortening the BBL and 21 BBS causes tendinopathy of the intertubercular groove and coracoid process. Therefore, it is necessary 22 to stretch the BBL and BBS separately. This study aimed to determine the positions where the BBL 23 and BBS were most stretched, using shear wave elastography (SWE). Fifteen healthy young males 24participated in the study. The shear elastic moduli of the BBL and BBS of the non-dominant arm were 25 measured using SWE. The measurement positions were the resting position (shoulder flexion and 26 abduction 0°) and four stretching positions. The elbow was extended, and the forearm was pronated in 27 all positions. Statistical analysis was performed using Wilcoxon's signed-rank test to compare the shear 28 elastic moduli between the resting and stretched limb positions. In addition, Wilcoxon's signed-rank 29 test was used to compare shear elastic moduli between the stretching positions that were significantly 30 different compared to the resting position. Results show that for BBL and BBS, shear elastic moduli 31 were significantly higher in the shoulder extension + external rotation and shoulder horizontal 32 abduction + internal rotation positions than in the resting position. Moreover, the shear elastic modulus 33 of the BBL was significantly higher in shoulder extension + external rotation than in shoulder 34 horizontal abduction + internal rotation. In contrast, the shear elastic modulus of the BBS was 35 significantly higher in shoulder horizontal abduction + internal rotation than in shoulder extension +

36	external rotation.	The B	BL and	BBS	were	effectively	stretched	by	shoulder	extension	+ (external
37	rotation and horizo	ontal al	bduction	+ inte	ernal r	otation.						

Keywords: Shear elastic modulus, Stretching, Biceps brachii, Rehabilitation, Shear wave elastography
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41 **1. Introduction**

42	The biceps brachii (BB) consists of a long head (BBL) and a short head (BBS). The BBL and BBS,
43	which run differently, tend to have different clinical conditions. Tendinitis of BBL is often observed in
44	the intertubercular groove (Berlemann et al,. 1995), while inflammation of the BBS often occurs at the
45	coracoid process (Karim et al., 2005). Thus, individual stretching positions for BBL and BBS should
46	be considered because of differences in anatomy and clinical symptoms.
47	Several studies have reported stretching positions for BBL and BBS (Houglum et al ,. 2001,
48	Evjenth et al, 1993). Specifically, elbow and shoulder extension and forearm pronation in the sitting
49	or supine position has been reported as stretching positions for BBL and BBS (Houglum et al ,. 2001).
50	The individual stretching position of the BBL has been presented as a position combining shoulder
51	extension, adduction, external rotation, forearm pronation, and elbow extension in the side-lying

52	position (Evjenth et al, 1993). The individual stretching position of the BBS has been presented as a
53	position combining shoulder abduction, external rotation, forearm pronation, and elbow extension
54	(Evjenth et al,. 1993) . However, these are expert opinions, and no experimental studies have
55	quantitatively examined the most effective stretching positions for BBL and BBS in vivo.
56	Ultrasonic shear wave elastography (SWE) has been used to measure the viscoelastic
57	properties of soft tissues. With this method, it is possible to quantitatively calculate the shear elastic
58	modulus of a tissue using the propagation shear wave velocity when the tissue is vibrated using a
59	radiation force (Brandenburg et al., 2014). The shear elastic modulus, which is highly correlated with
60	the passive properties of the muscle (Eby et al., 2013; Koo et al., 2013), has been used as an indicator
61	of muscle elongation to examine the effective stretching positions of individual muscles (Asayama et
62	al., 2021; Ogawa et al., 2020; Umehara et al., 2017; Yanase et al., 2021) . Therefore, SWE can be used
63	to quantitatively examine the most effective stretching positions for the BBL and BBS.
64	BBL and BBS with moment arms for elbow flexion and forearm supination (Hale et al.,
65	2011) are more stretched by elbow extension and forearm pronation. Furthermore, they are affected
66	by the positions of the elbow and shoulder joints because they are bi-articular muscles. Anatomically,
67	a BB with moment arms for shoulder flexion and horizontal adduction is considered to be stretched by
68	shoulder extension or horizontal abduction. In addition, the BBL, which is anatomically located

69	laterally, can be considered more stretched with shoulder extension. Conversely, the BBS, which is
70	anatomically located medially, could be considered more stretched by shoulder horizontal abduction.
71	However, no reports have quantitatively examined stretching methods for BBL and BBS that consider
72	the position of the shoulder joint in addition to the elbow and forearm. Furthermore, studies have not
73	quantitatively examined whether BBL and BBS are stretched by the external or internal rotation of the
74	shoulder, and its effect remains unclear.
75	This study aimed to clarify the stretching position in which the BBL and BBS are most
76	stretched using SWE, focusing on the shoulder joint position. The hypothesis was that the BBL is
77	effectively stretched by elbow extension, forearm pronation, and shoulder extension. In contrast, the
78	BBS is effectively stretched by elbow extension, forearm pronation, and shoulder horizontal abduction.
79	
80	2. Methods
81	2.1 Participants
82	Fifteen healthy young men (age 26.8 ± 5.6 years; height, 172.3 ± 6.4 cm; weight, 65.5 ± 9.8 kg)
83	participated in the study. People with any history of orthopedic or neurological diseases of the upper
84	limb, including BB, were excluded from this study. The sample size was calculated using G*Power
85	software (version 3.1; Heinrich Heine University, Dusseldorf, Germany) for Wilcoxon's signed-rank

86	test (effect size, 0.8; alpha error, 0.05; power, 0.8), which showed that 15 participants were required.
87	The sample size was determined referring to previous studies (Umehara et al,. 2017). All participants
88	were fully informed about the study aims and procedures, and informed consent was obtained before
89	participation. The study was approved by the Ethics Committee of the Kyoto University Graduate
90	School and Faculty of Medicine (R0233).

92 2.2 Stretching procedures

93 The same two investigators with physical therapist licenses performed all procedures. One investigator 94 measured the shear elastic modulus using SWE, whereas the other performed the stretching maneuver 95 for the participant's non-dominant arm. The dominant arm is the arm that rows the ball during sports. 96 All stretching exercises were performed in the prone position, elbow extension, and forearm pronation 97 under the following five conditions (Fig. 1): (A) Resting position: shoulder extension 0° , abduction 0° , 98 internal/external rotation intermediate position, (B) shoulder maximal extension + external rotation, 99 (C) shoulder maximal extension + internal rotation, (D) shoulder maximal horizontal abduction + 100 external rotation, and (E) shoulder maximal horizontal abduction + internal rotation. The order of 101 stretching was as follows: shoulder rotation first, followed by shoulder extension or horizontal 102 abduction, and finally, elbow extension and forearm pronation. The mean and SD values of the range

103	of motion during extension and horizontal abduction for participants were 29.0° \pm 12.8 and 11.5° \pm
104	8.0, respectively. Stretching was performed at the maximum angle at which the participants felt no
105	discomfort or pain. The time from the start of the stretching to the measurement was within 1 min. The
106	order of stretching was randomized using a random number table (Microsoft Excel, Microsoft Corp.,
107	Redmond, WA, USA) (Yanase et al., 2021). The participants rested for at least two minutes after each
108	stretching session to remove the effects of sustained stretching (Asayama et al., 2021). The participants
109	were instructed to relax as much as possible to avoid muscle contractions during stretching.
110	
111	2.3 Measurements of shear elastic modulus
112	The shear elastic moduli of the BBL and BBS were measured using an ultrasonic SWE (Aixplorer;
113	SuperSonic Imagine, Aix-en-Provence, France) with a linear probe (4-15 MHz, SL15-4, Vermon,
114	Tours, France). The presets were set to Penetration and Smoothing 5, and the scale was set to an upper
115	limit of 800 kPa. The measurement site was the distal 70% of brachial length (from the acromion to
116	the lateral epicondyle). The long and short heads were separated by palpation at the site, and the elastic
117	modulus was measured. To ensure that the measurement sites were consistent, body markers were used
118	First, the cross-sectional images of the BBL and BBS were confirmed using the B-mode, and the probe
119	was placed parallel to the muscle fascicle of the BBL and BBS (Fig. 2). A region of interest (ROI) of

120	1.0 cm length and 2.5 cm width was set at the center of the muscle belly. For quantitative analysis, the
121	shear elastic modulus in the ROI was measured using the Q-box TM trace function. The shear elastic
122	moduli were measured twice for BBL and BBS, and the average values were used for the statistical
123	analysis. The shear elastic modulus (G) was calculated from the shear wave speed (V) using the
124	following equation:
125	
126	$G(kPa) = \rho V^2$
127	
128	where ρ is the muscle mass density (1000 kg/m ³); high values of ρ indicate high muscle stiffness.
129	In this study, we calculated the shear modulus by dividing the Young's modulus, which was provided
130	by the device, by three.

132 2.4 Measurements of electromyography

133	Surface electromyography (TeleMyo 2400, Noraxon USA, Scottsdale, AZ, USA) was used to measure
134	muscle activity in the BBL and BBS to confirm no muscle contraction during stretching. Bipolar
135	surface electrodes (Blue Sensor; Medicotest, Olsykke, Denmark) were attached to the muscle belly of
136	the BB within 2 cm of the shear elastic modulus measurement site with a distance of 2 cm between the
137	electrodes. The surface electromyogram (EMG) sampling rate was 1500 Hz, and bandpass filtering
138	was set between 10 and 500 Hz. The root-mean-square (RMS) value was calculated after rectification.
139	To normalize the measured data, the maximum voluntary isometric contraction (MVC) of elbow
140	flexion was measured for 3 seconds in the sitting position at the shoulder joint neutral position and
141	elbow joint flexion at 90°. MVC was measured at the end of the experiment.
142	
143	2.5 Statistical analysis
144	Statistical analysis was performed using the IBM SPSS Statistics software (version 22.0, IBM Corp,
145	Armonk, NY, USA). The reliability of the shear elastic modulus measurements for each position was
146	evaluated using the variation and intraclass correlation coefficients (1,2) (ICC _{1,2}). Normality was

- 147 confirmed using the Shapiro–Wilk test. The shear elastic moduli of BBL and BBS were analyzed using
- 148 Wilcoxon's signed-rank test and Bonferroni correction to compare resting and all stretching positions.

149 In addition, Wilcoxon's signed-rank test was used to compare stretching positions significantly 150 different from the resting position. The significance level was set at p < 0.05. 151 152 3. Results 153 Regarding the reliability of the shear elastic moduli measurements for each position, the coefficient of 154 variation was less than 10%, and the ICC (1,2) was greater than 0.8 for both BBL and BBS, indicating 155 high reliability (Table 1). In addition, the muscle activity during each stretching motion was less than 156 5% MVC in both muscles, confirming that no contraction occurred during the stretching. 157 Tables 2 and 3 show the shear elastic moduli of BBL and BBS, respectively. The shear elastic 158 modulus of the BBL was significantly higher for maximal extension + external rotation and maximal 159 horizontal abduction + internal rotation than for the resting position (p = 0.002 and p = 0.003, 160 respectively). Similarly, the shear elastic moduli of the BBS in maximum extension + external rotation 161 and maximum horizontal abduction + internal rotation were significantly higher than those in the 162 resting position (p = 0.003 and p = 0.002, respectively). 163 Next, Wilcoxon's signed-rank tests were performed between the stretching positions that 164 showed significant differences compared to the resting position, that is, maximal extension + external 165 rotation and maximal horizontal abduction + internal rotation, for both BBL and BBS. The results

166	showed that maximum extension + external rotation was significantly higher than maximum horizontal
167	abduction + internal rotation in BBL ($p = 0.012$) (Fig. 3). In the BBS, maximal horizontal abduction +
168	internal rotation were significantly higher than maximal extension + external rotation ($p = 0.010$) (Fig.
169	4).
170 171	
172	4. Discussion
173	In this study, we investigated the stretching position in which the BBL and BBS were the most
174	stretched using SWE, focusing on the shoulder joint position. For both BBL and BBS, the shear elastic
175	moduli were significantly higher in shoulder extension + external rotation and horizontal abduction +
176	internal rotation than in the resting position (extension 0° and abduction 0°). Furthermore, the shear
177	elastic modulus of BBL was significantly higher in shoulder extension + external rotation than in
178	horizontal abduction + internal rotation, whereas BBS was significantly higher in horizontal abduction
179	+ internal rotation than in extension + external rotation. These results suggest that the elbow and
180	shoulder joint position affect the amount of biceps brachii elongation and that the most stretched
181	position differs between BBL and BBS. This is the first study to quantitatively examine the individual
182	stretching positions for BBL and BBS using SWE.
183	The amount of muscle elongation was affected by the moment arm. In tendon excursion

The amount of muscle elongation was affected by the moment arm. In tendon excursion

184	methods (Maganaris C N et al., 2000), the muscle is stretched by joint motion opposite to the moment
185	arm possessed by the muscle. Considering the muscle attachment sites, the BBL and BBS have
186	moment arms for shoulder joint flexion and horizontal adduction. Thus, the muscles are stretched by
187	shoulder extension and horizontal abduction. The results of this study support our hypothesis.
188	Furthermore, the results of this study suggest that shoulder extension, horizontal abduction,
189	and shoulder rotation affect the extent of BB muscle elongation. Regarding the effect of shoulder
190	rotation, it is possible that the shoulder extension range of motion was greater during external rotation
191	than during internal rotation, which might have resulted in a higher shear elastic modulus with the
192	combination of shoulder extension and external rotation. In contrast, it is possible that the shoulder
193	horizontal abduction range of motion was greater during internal rotation than during external rotation,
194	which might have resulted in a higher shear elastic modulus in the combination of horizontal abduction
195	and internal rotation.
196	The results of this study showed that the shear elastic moduli were highest in shoulder
197	extension + external rotation for BBL and horizontal abduction + internal rotation for BBS.
198	Considering the difference in the most stretched position between BBL and BBS, BBS may have a
199	larger shoulder horizontal adduction moment arm than BBL, and BBL may have a larger shoulder
200	extension moment arm than BBS. This suggests that horizontal shoulder abduction and shoulder

202	Regarding clinical implications, effective stretching positions for muscle lengthening need
203	to be identified separately for BBL and BBS because muscle attachment sites differ between them (van
204	den Bekerom et al., 2016). Based on the results of this study, the recommended stretching position for
205	BBL is elbow extension and forearm pronation combined with shoulder extension and external rotation,
206	and the recommended stretching position for BBS is elbow extension and forearm pronation combined
207	with shoulder horizontal abduction and internal rotation.
208	A limitation of this study is that the participants were healthy people; therefore, it is unclear
209	whether the same effects can be expected for patients with musculoskeletal disorders. Second, the
210	chronic effect of stretching on BB flexibility remains unknown. Further research is required to
211	determine the effects of long-term stretching interventions on BB flexibility using effective stretching
212	positions.
213	
214	5. Conclusion
215	The current study measured the shear elastic moduli obtained by SWE to identify the stretching
216	positions in which BBL and BBS were the most stretched. Our results revealed that the BBL was

extension may have been effective in elongating the BBS and BBL, respectively.

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217 stretched in elbow extension and forearm pronation combined with shoulder extension and external

218	rotation, whereas the BBS was stretched in elbow extension and forearm pronation combined with
219	shoulder horizontal abduction and internal rotation. Shoulder joint position, including rotation, should
220	be considered during BBL and BBS stretching, in addition to the elbow joint position.
221	
222	Declaration of Competing Interest
223	The authors declare no conflicts of interest related to the manuscript.
224	
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228	
229	Author's contributions
230	K.I., K.Y., T.I., and N.I. conceived and designed the research. K.I. and K.Y. performed the
231	experiment and analyzed the data. K.I., K.Y., T.I., and N.I. interpreted the results. K.I. and K.Y.
232	wrote the manuscript. K.I. and K.Y. edited and revised the manuscript. All authors have approved
233	the final version of the manuscript.
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- 279 Maximum shoulder extension + internal rotation. (d) Maximum shoulder horizontal abduction +
- 280 external rotation. (e) Maximum shoulder horizontal abduction + internal rotation.
- 281









- short head (a) B-mode cross-sectional image of the biceps brachii long and short heads. The
- distinction between adjacent long and short heads was performed by palpation. (b) Color mapping in

- the region of interest indicates the muscle shear elastic modulus using shear wave elastography
- 288 (longitudinal image of the biceps brachii long head).
- 289

290 Table 1 Reliability of shear elastic modulus measurements

Massurement position		CV (%)		ICC (1,2)			
Measurement position	BBL	BBS	BBL	95% CI	BBS	95% CI	
Rest (Extension 0° and abduction 0°)	4.7	7.5	0.96	0.90–0.98	0.82	0.48–0.94	
Extension + external rotation	3.9	4.4	0.98	0.95–0.99	0.98	0.96–0.99	
Extension + internal rotation	4.2	2.8	0.98	0.95–0.99	0.98	0.96–0.99	
Horizontal abduction + external rotation	3.0	4.4	0.99	0.98–0.99	0.97	0.93–0.99	
Horizontal abduction + internal rotation	3.6	5.0	0.98	0.98–0.99	0.96	0.90-0.98	

291 CV, coefficient of variation; ICC, intraclass correlation coefficient; BBL, biceps brachii long head;

BBS, biceps brachii short head; CI, confidence interval

293

294 Table 2 Shear elastic modulus of biceps brachii long head

Massurement position	Shear elastic modulus	Comparison with rest		
Measurement position	(Means \pm SD, kPa)	(<i>p</i> value)		
Rest (extension 0° and abduction 0°)	22.9 ± 7.0			
Extension + external rotation	56.4 ± 25.1	0.002		
Extension + internal rotation	22.8 ± 8.1	3.64		
Horizontal abduction + external rotation	34.4 ± 14.4	0.124		
Horizontal abduction + internal rotation	38.5 ± 13.7	0.003		

295 In bold: statistically significant difference compared with rest

297 Table 3 Shear elastic modulus of biceps brachii short head

Macourrement position	Shear elastic modulus	Comparison with rest		
Measurement position	(Means \pm SD, kPa)	(p value)		
Rest (extension 0° and abduction 0°)	20.5 ± 6.0			
Extension + external rotation	36.0 ± 9.2	0.003		
	10			

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Extension + internal rotation	26.9 ± 19.6	1.22
Horizontal abduction + external rotation	41.2 ± 23.5	0.063
Horizontal abduction + internal rotation	48.3 ± 17.9	0.002

298 In bold: statistically significant difference compared with rest

299

300







303 higher elastic moduli compared to those of the rest. Ext+ER, shoulder extension + external rotation;

304 Ha+IR, shoulder horizontal abduction + internal rotation * Ext+ER was significantly greater than

305 Ha+IR (p < 0.05).

306



310 Figure 4 Comparison of shear elastic modulus of BBS between stretching positions with significantly

311 higher elastic moduli compared to those of the rest. Ext+ER, shoulder extension + external rotation;

312 Ha+IR, shoulder horizontal abduction + internal rotation * Ha+IR was significantly greater than

313 Ext+ER (P < 0.05).