



# Effects of knee and hip flexion angle on region-specific and longitudinal compartmental muscle stiffness of the rectus femoris using shear wave elastography in male bodybuilders

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## Abstract

**Purpose** To investigate the effects of different knee and hip flexion angles (KFA and HFA) on region-specific and longitudinal compartmental muscle stiffness of the rectus femoris (RF) using shear wave elastography (SWE). As muscle mechanical tension is a key indicator of activation, measurements of muscle stiffness offer new insights into muscle contractile dynamics.

**Methods** Isometric knee extension (KE) was performed by nine male bodybuilders at maximum voluntary contraction (MVC) using a dynamometer under combined conditions of KFA at 30°, 60°, and 90° and HFA at 0°, 40°, and 80°. Muscle stiffness during KE was measured in the RF, vastus lateralis (VL), and vastus medialis (VM) using SWE. The RF and VL were divided into proximal, middle, and distal regions, and the VL, VM, and RF were compared across joint angle conditions and regions. Additionally, muscle activity during the KE was assessed in the proximal, middle, and distal RF using multichannel electromyography (EMG).

**Results** Muscle stiffness in the proximal RF was higher at HFA 0° and 40° compared to HFA 80° under KFA 30° conditions ( $p < 0.05$ ). The stiffness in the middle RF was higher than that in the middle VL at HFA 0° and 40° under a KFA 30° ( $p < 0.01$ ). EMG revealed no differences across regions or joint angles.

**Conclusion** Performing KE with the hip in an extended position enhanced the stiffness of the RF, particularly in the proximal region, compared to the VL. While region-specific muscle activity was observed using SWE, these differences were not detected using EMG.

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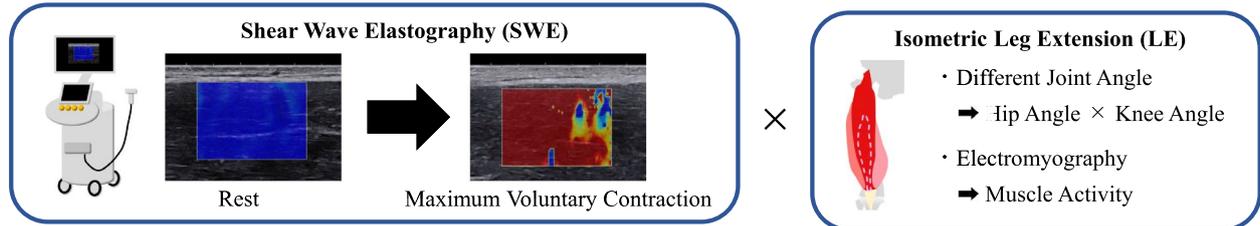
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## Graphical abstract

## Purpose of Study

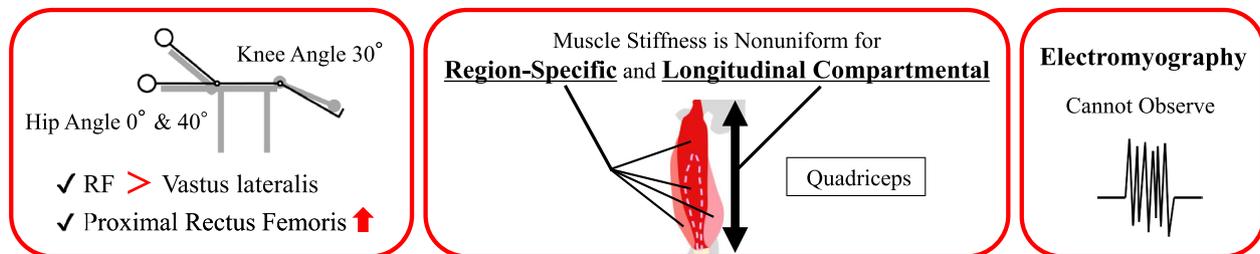
To Measure Region-Specific muscle stiffness in the Rectus Femoris (RF) during LE with Changing Joint Angle

## Research Design Muscle Stiffness × LE with Changing Joint Angle



## Key Finding

LE with Hip Extension Increases the Muscle Stiffness of the Proximal RF



**Keywords** Resistance training · Bodybuilding · Rectus femoris · Shear wave elastography

## Abbreviations

ANOVA	Analysis of variance
ARV	Average rectified value
EMG	Electromyography
HFA	Hip flexion angle
KFA	Knee flexion angle
KE	Knee extension
MVC	Maximum voluntary contraction
RF	Rectus femoris
SWE	Shear wave elastography
VL	Vastus lateralis
VI	Vastus intermedius
VM	Vastus medialis

## Introduction

The anatomy of the rectus femoris (RF) can be compartmentalized longitudinally because of the separation of the proximal and distal innervating motor nerve branches (Yang and Morris 1999). Therefore, there are longitudinally compartmentalized muscle activity dynamics in the RF, in which muscle activity differs within the same muscle (Watanabe et al. 2012). It has been reported that the proximal

RF is particularly active in the longitudinal compartment within the same muscle during walking (Ema et al. 2018; Watanabe et al. 2014b), which is believed to be influenced by anatomical features, such as motor nerve branching (Tubbs et al. 2006; Yang and Morris 1999). Thus, strengthening the proximal RF may contribute to healthier life expectancy in older individuals (Watanabe et al. 2018). Ema et al. (2016) reported higher myoelectrical activity in the vastus muscle group (a collective term for the vastus lateralis [VL], vastus intermedius [VI], and vastus medialis [VM]) during multi-joint exercises, whereas RF activity was more pronounced during knee extension (KE), a single-joint exercise. Similarly, Watanabe et al. (2012) observed high distal RF myoelectrical activity during the KE, whereas proximal RF activity remained low, indicating that conventional KE may not effectively strengthen the proximal RF.

In previous studies examining the quadriceps, the hip flexion angles (HFA) during the KE were often restricted to approximately 80° (Narici MV 1996; Wakahara et al. 2017). The HFA may influence the alignment of the proximal tendon of the RF and alter its contractile dynamics, as the proximal tendon is divided into two parts with different angles of attachment to the pelvis (Gyftopoulos et al. 2008; Hasselman et al. 1995). Watanabe et al. (2014a) found no

effect of varying the HFA on RF myoelectrical activity during the KE. In contrast, we found that the KE with altered HFA, particularly with hip extension, activated the proximal RF (Mitsuya et al. 2023). Further research on the effects of HFA on proximal RF activation is required.

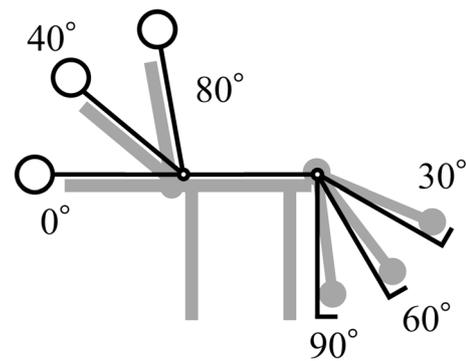
One method of observing muscle contraction dynamics is to measure muscle stiffness using shear wave elastography (SWE). The propagation velocity measured by SWE increases in proportion to the torque and myoelectrical activity (Nordez and Hug 2010; Yoshitake et al. 2014). The validity of muscle stiffness measurements for assessing muscle activation has been proven, as previous studies have used SWE to assess thigh muscle activity during maximum voluntary contraction (MVC) (Avrillon et al. 2018). Because muscle mechanical tension during MVC is a key indicator of muscle activation (Eby et al. 2013), measurements using SWE offer new insights into region-specific muscle contractile dynamics in the quadriceps femoris.

Therefore, the purpose of this study was to measure region-specific muscle stiffness in the quadriceps femoris using SWE during MVC in isometric KE with varying HFA and knee flexion angles (KFA). We hypothesized that extending the HFA beyond the conventional 80° would increase the muscle stiffness in the proximal RF and contribute to overall RF strengthening. Furthermore, SWE was expected to reveal changes in regional RF activation patterns. To further verify muscle activity, the myoelectrical activity of the RF was measured during isometric KE using electromyography (EMG). To reduce the measurement variability caused by inconsistencies in movement, this study was conducted with bodybuilders skilled in movement control during resistance training.

## Methods

### Participants

This study was approved by the Ethics Review Committee of the Nippon Sport Science University (Approval No. 018-H113) and complied with the Declaration of Helsinki (Rickham 1964). All participants provided written informed consent before participation. The purpose and potential risks of the study were fully explained to all participants before enrollment. Nine male bodybuilders (age:  $21 \pm 2$  years, height:  $170 \pm 3$  cm, weight:  $71 \pm 6$  kg, training history:  $4 \pm 1$  years) participated in the study. None of the participants had a history of thigh or knee injuries. They had been training for muscle hypertrophy and engaged in high-intensity resistance training following a muscle-group split regimen for approximately one to three hours per day, at least four times per week.



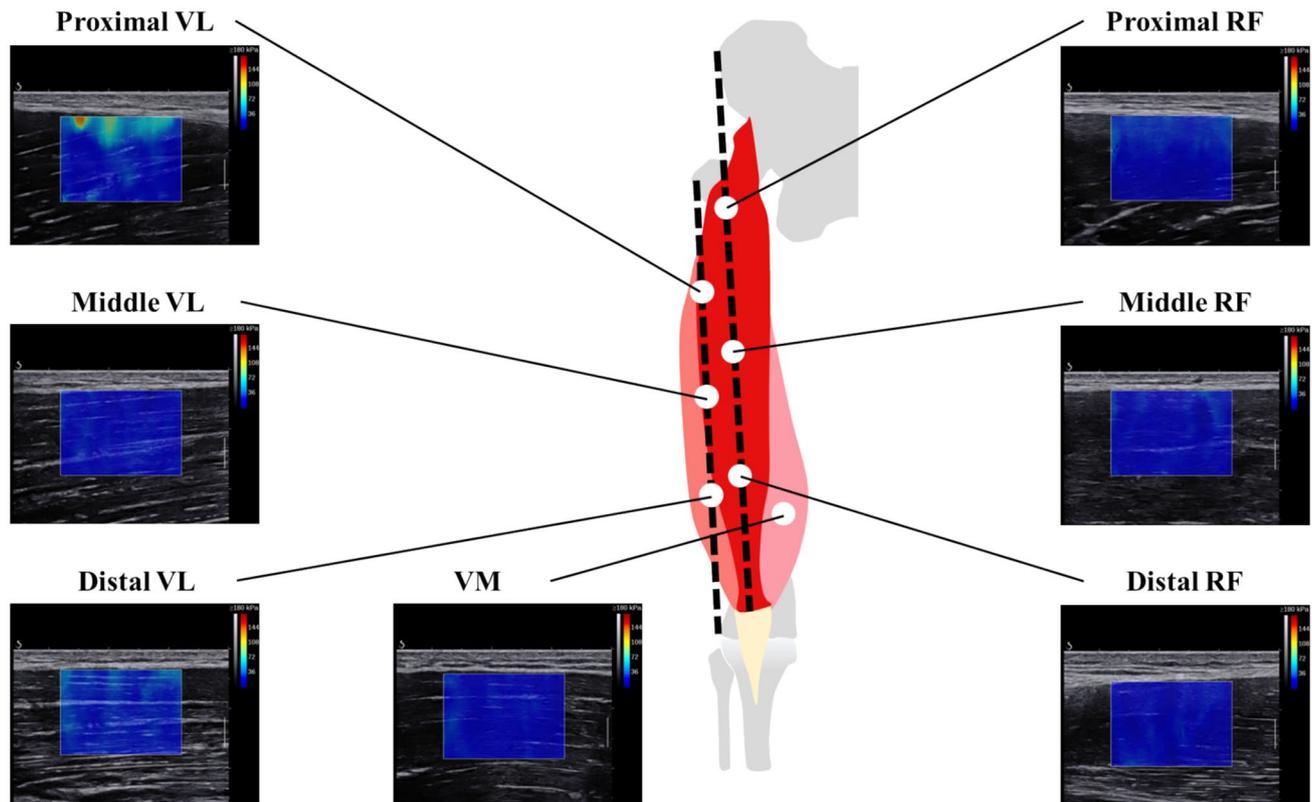
**Fig. 1** HFA and KFA settings. The HFA was set to 0°, 40°, and 80°, and the KFA was set to 30°, 60°, and 90°. Nine isometric KE conditions were applied for each angle combination. *HFA* hip flexion angle, *KFA* knee flexion angle

### MVC torque measurement of isometric KE

The isometric KE was performed using a dynamometer (Biodex System 4, Biodex Medical Systems Inc., USA) under varying HFA of 0°, 40°, and 80° and KFA of 30°, 60°, and 90°, resulting in a total of nine testing conditions. HFA 80° represented the conventional KE, HFA 0° corresponded to the maximum hip extension achievable with the machine settings, and HFA 40° was the intermediate position (Fig. 1). For each joint angle condition, the MVC torque during the isometric KE was measured with a minimum rest interval of two minutes between trials. Force exertion was sustained for five seconds, and the highest recorded value was defined as the maximum isometric KE torque. The trial order was randomized to mitigate the effects of fatigue. The EMG and SWE measurements, described below, were recorded concurrently with the KE torque assessments.

### Measurement of SWE

SWE was measured using an ultrasound imaging system (Aixplorer; SuperSonic Imagine, France). Muscle stiffness was quantified based on the propagation velocity of shear waves within a  $15 \times 20$  mm region of interest (ROI). A linear probe (SuperLinear™ SL10-2, SuperSonic Imagine, France) coated with ultrasound gel was placed parallel to the muscle fibers, with the upper edge of the ROI aligned with the border of subcutaneous fat. The RF was divided into three regions: the proximal RF at 30%, middle RF at 50%, and distal RF at 70% of the line connecting the superior anterior iliac spine to the superior border of the patella in the seated position. For the VL, the proximal, middle, and distal regions were defined as points located at 30%, 50%, and 70%, respectively, along the line connecting the greater trochanter to the lateral epicondyle where the ultrasound image was the most stable after avoiding



**Fig. 2** Quadriceps muscle regions assessed by SWE. *RF* rectus femoris, *VL* vastus Lateralis, *VM* vastus medialis

the iliotibial ligament. The VM was evaluated at a single point where the image was the most stable, ranging from the border between the RF and sartorius muscles to the suprapatellar border. The VI was excluded from the analysis owing to its deep position beneath the RF, which made defining the ROI challenging. Ultrasound images were recorded during the isometric KE, and the image with the most stable waveform was selected for analysis. Owing to the extensive number of measurement points, data collection was divided into two sessions conducted on separate days with a minimum interval of 72 h between sessions (Fig. 2). Considering the effects of fatigue, measurements were performed on two separate days.

The RF was divided into the proximal (30%), middle (50%), and distal RF (70%) along the line connecting the superior anterior iliac spine to the superior border of the patella. For the VL, the proximal (30%), middle (50%), and distal (70%) regions were defined along the line connecting the greater trochanter to the lateral epicondyle, selecting the point where the image was the most stable while avoiding the iliotibial ligament. The VM was evaluated only at a single point where the image was the most stable, ranging from the border between the RF and sartorius muscles to the suprapatellar border.

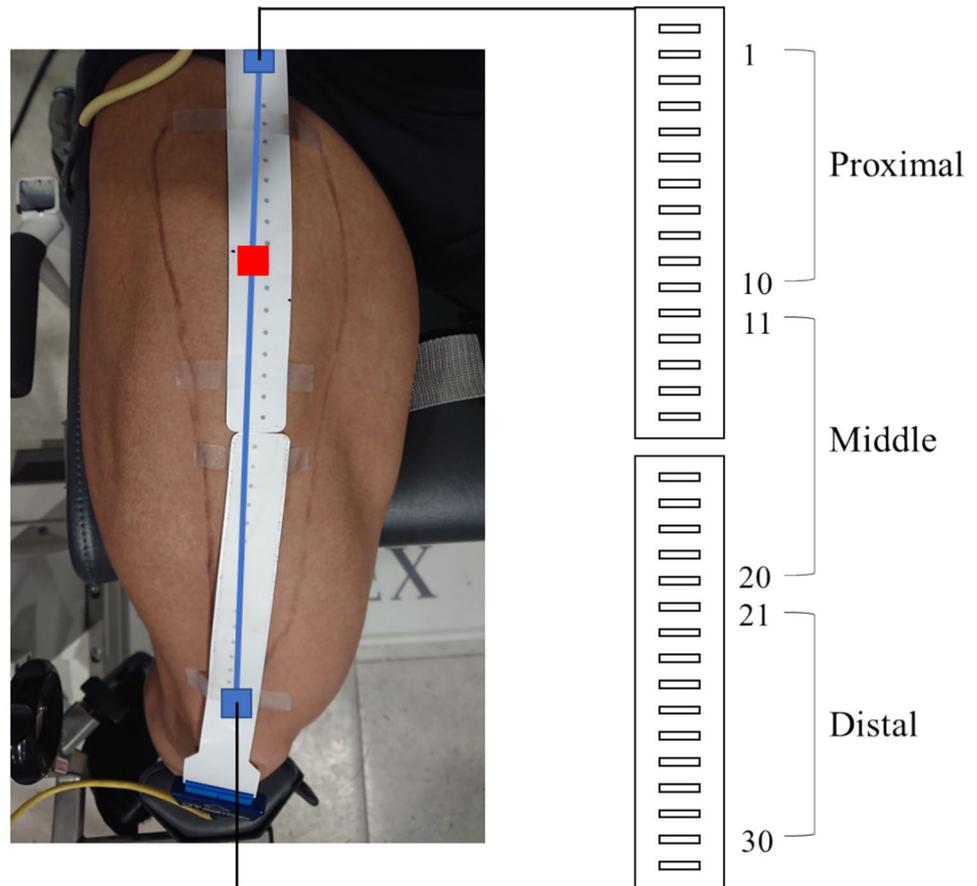
### Analysis method of SWE

If a tendon was present within the region of interest (ROI), the ROI was redefined to exclude the tendon. If no tendon was present, the shear wave propagation velocity within the conventional ROI was used to calculate the absolute value of muscle stiffness during MVC based on previous studies (Ates et al. 2015). To compare muscle stiffness among the quadriceps muscle heads, the middle VL and middle RF were defined as the middle thigh region, whereas the distal VL, distal RF, and VM were collectively defined as the distal thigh region. Proximal VL and proximal RF were not included in the direct comparisons because of their anatomical separation. The coefficient of variation of the measurements was  $0.30 \pm 0.11\%$ . To investigate the region-specific and longitudinally compartmentalized muscle contractile dynamics, muscle stiffness was compared (1) between each HFA within the same region and (2) between regions within the same HFA.

### Measurement of EMG

EMG was measured using a multichannel surface EMG system (EMG-USB2, OT Bioelectronica, Italy) following the protocol described in previous studies (Watanabe et al.

**Fig. 3** Installation of the multi-channel surface EMG



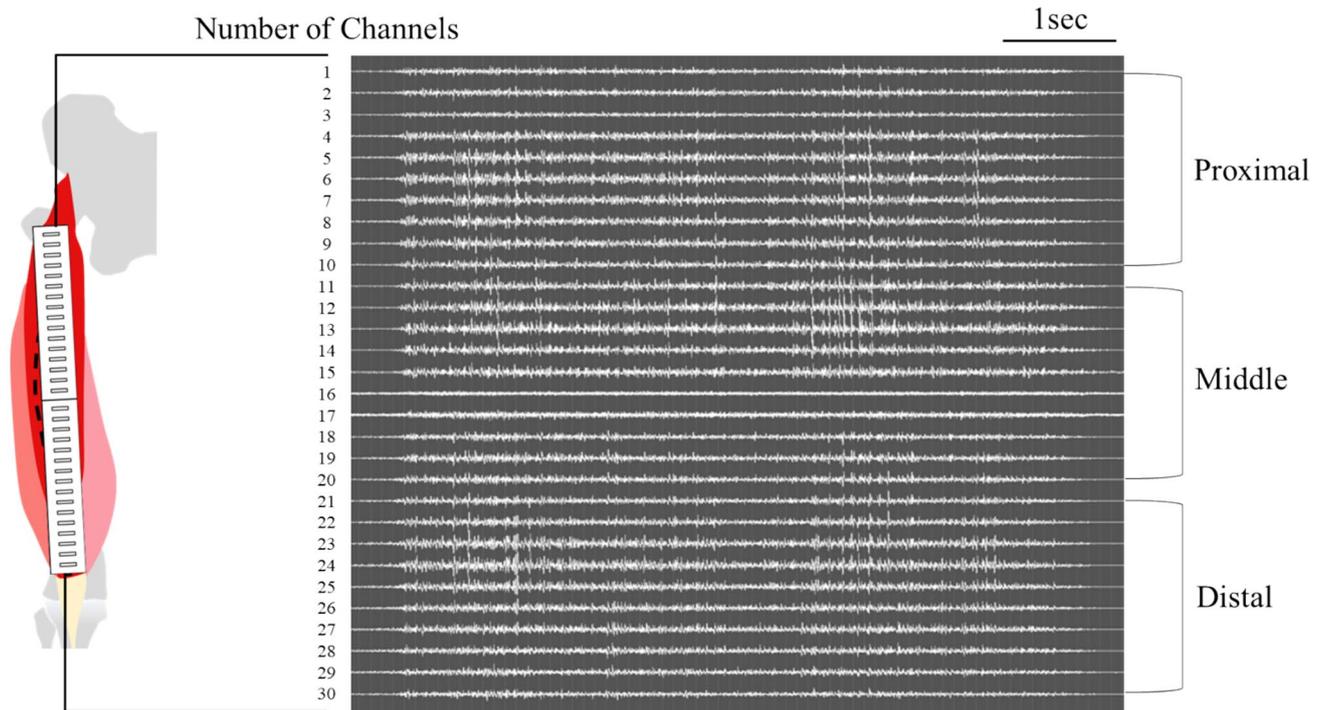
2012). Two electrode plates, each containing 16 electrodes in a straight line, were placed on the proximal and distal regions of the RF, with a total of 30 channels measured after excluding the electrodes at the terminal ends. The 30 electrodes were divided into the proximal, middle, and distal regions, with each region comprising 10 electrodes (Fig. 3). Before electrode placement, the participants shaved their thighs to ensure proper electrode contact. The borders of the VL, VM, sartorius, and tensor fascia latae were identified using an ultrasound imaging system (PRO SOUND SSD-3500; Hitachi, Japan) and these borders were marked with a skin-safe marker for accurate RF localization. The line connecting the superior anterior iliac spine to the superior border of the patella was defined as the centerline. The center of the proximal electrode plate was aligned with the proximal one-third of the centerline. The skin was disinfected with alcohol prior to electrode placement. Figure 3 shows the installed electrodes. Considering the effects of fatigue, measurements were taken on two separate days.

The 30-channel electrode configuration was divided into the proximal, middle, and distal regions, with 10 electrodes assigned to each region. The center of the proximal electrode plate was aligned with the proximal one-third of

the centerline, defined as the line connecting the superior anterior iliac spine and superior border of the patella.

### Analysis method of EMG

Electrical signals acquired from the electrodes were transmitted to a computer via a multichannel surface EMG system and analyzed using a dedicated software (OT BioLab, OT Bioelectronica, Italy). The visualized waveforms were filtered using a bandpass filter between 10 and 450 Hz, and processed as the average rectified value (ARV) for evaluation. The dynamometer and the multichannel surface EMG system were synchronized using a transducer. The ARV was calculated by averaging the ARV for one second before and one second after the point of maximum isometric KE torque exertion, thus totaling two seconds. The ARVs obtained from the 30 electrode channels were categorized into three regions: channels 1 to 10 as proximal, channels 11 to 20 as middle, and channels 21 to 30 as distal. The ARV for each region was calculated by averaging the ARVs of the corresponding channels. The coefficient of variation of the measurements was  $0.39 \pm 0.10\%$ . Based on previous studies (Watanabe et al. 2012), absolute ARVs during MVC were used for comparisons both (1) between HFA within the same



**Fig. 4** Visualized waveforms acquired from multichannel surface EMG

region and (2) between RF regions within the same HFA (Fig. 4).

Waveforms acquired from a total of 30 electrode channels attached to the RF were filtered between 10 and 450 Hz using a bandpass filter and processed as ARVs for evaluation. Channels 1 to 10 were categorized as proximal, channels 11 to 20 as middle, and channels 21 to 30 as distal.

### Statistical analysis

A one-way repeated-measures analysis of variance (ANOVA) was used to compare the maximum isometric KE torque between each HFA within the same KFA and between each KFA within the same HFA. Paired t-tests or two-way repeated-measures ANOVA was used to compare the muscle stiffness among the quadriceps muscle heads with factors for HFA and KFA. Two-way repeated-measures ANOVA was used to compare the muscle stiffness and ARV between each HFA within the same region with factors for HFA and KFA and between regions within the same HFA with factors for region and KFA. If a main effect was detected, one-way and two-way repeated-measures ANOVA was followed by Bonferroni's post hoc test for pairwise comparisons to identify significant differences. Effect sizes were reported as  $r$  for t-tests and partial  $\eta^2$  for one-way and two-way repeated measures ANOVA. All statistical analyses were performed using SPSS Statistics

version 27.0.0.1 for Windows; IBM, Armonk, NY, USA). The significance level was set at  $p < 0.05$ .

## Results

### Maximum isometric KE torque

The maximum isometric KE torque at each joint angle was compared (Table 1). No differences were observed in maximum KE torque between HFA within the same KFA (KFA 30°:  $p = 0.356$ , partial  $\eta^2 = 0.158$ ; KFA 60°:  $p = 0.701$ , partial  $\eta^2 = 0.058$ ; KFA 90°:  $p = 0.131$ , partial  $\eta^2 = 0.287$ ). However, a comparison of the maximum isometric KE torque between each KFA within the same HFA revealed a main effect for all HFAs (HFA 0°:  $p = 0.001$ , partial  $\eta^2 = 0.752$ ; HFA 40°:  $p = 0.002$ , partial  $\eta^2 = 0.780$ ; HFA 80°:  $p = 0.001$ , partial  $\eta^2 = 0.789$ ). Post hoc comparisons indicated that at HFA 0°, the maximum isometric KE torque at KFA 60° and KFA 90° was higher than that at KFA 30° (KFA 60°:  $p = 0.035$ ,  $r = 0.825$ , KFA 90°:  $p = 0.003$ ,  $r = 0.774$ ). Similarly, at HFA 40°, the maximum isometric KE torque at KFA 90° was higher than at KFA 30° ( $p = 0.007$ ,  $r = 0.751$ ) and KFA 60° ( $p = 0.001$ ,  $r = 0.901$ ). At HFA 80°, the maximum isometric KE torque was higher at KFA 90° ( $p = 0.002$ ,  $r = 0.935$ ) and KFA 60° ( $p = 0.004$ ,  $r = 0.916$ ) compared to KFA 30°. These results suggest that the KE force was greater

**Table 1** Maximum isometric KE torque for each joint angle condition

HFA	0°			40°			80°		
	30°	60°	90°	30°	60°	90°	30°	60°	90°
Maximum isometric KE torque (N•m)	157.9±31.4	234.5±76.6 *	255.4±59.0 **	155.9±53.6	219.8±81.3	300.3±93.6 ** ††	138.3±36.5	235.2±43.7 **	270.5±83.6 **

KFA knee flexion angle, HFA hip flexion angle, KE knee extension, \* = vs. KFA 30°:  $p < 0.05$ , \*\* = vs. KFA 30°:  $p < 0.01$ , †† = vs. KFA 60°:  $p < 0.01$ . Values are presented as mean ± standard deviation

**Table 2** Muscle stiffness at rest for each joint angle condition

HFA	0°			40°			80°		
	30°	60°	90°	30°	60°	90°	30°	60°	90°
proximal RF	28.3±11.7	29.4±8.9	36.7±15.4	36.7±10.9	35.2±12.3	36.1±8.2	32.5±11.0	40.4±15.9	35.0±12.0
middle RF	26.8±7.4	32.2±10.4	29.7±10.2	32.7±9.3	32.8±9.7	36.1±15.3	26.5±5.2	38.1±12.2	37.1±7.2
distal RF	30.2±13.1	33.8±7.5	31.6±9.2	30.6±5.9	36.1±7.6	33.7±8.6	31.1±6.0	38.2±5.4	33.1±7.7
proximal VL	30.8±15.8	28.6±8.9	24.5±6.6	21.9±5.7	25.9±10.4	32.0±11.7	25.8±6.1	22.4±9.2	30.4±9.6
middle VL	16.1±5.3	20.8±9.5	20.8±7.8	14.1±5.3	17.7±5.8	21.1±7.4	20.6±9.9	19.6±5.7	19.6±7.3
distal VL	17.6±6.6	24.0±7.9	24.5±5.7	16.0±7.8	20.1±8.4	25.3±10.4	15.8±4.0	21.8±9.6	20.3±6.7
VM	18.1±6.1	21.4±6.4	20.2±5.5	14.2±3.7	15.5±4.1	18.0±6.0	16.1±5.9	17.0±6.1	23.0±5.4

HFA hip flexion angle, KFA knee flexion angle, RF rectus femoris, VL vastus lateralis, VM vastus medialis. Values are presented as mean ± standard deviation

during flexion than during extension, regardless of the HFA condition.

each muscle region. No differences in muscle stiffness were observed across any region or joint angle at rest.

**SWE**

Muscle stiffness at rest and during isometric KE for each joint angle condition are presented in Tables 2 and 3 for

**Comparison of muscle stiffness within each quadriceps head**

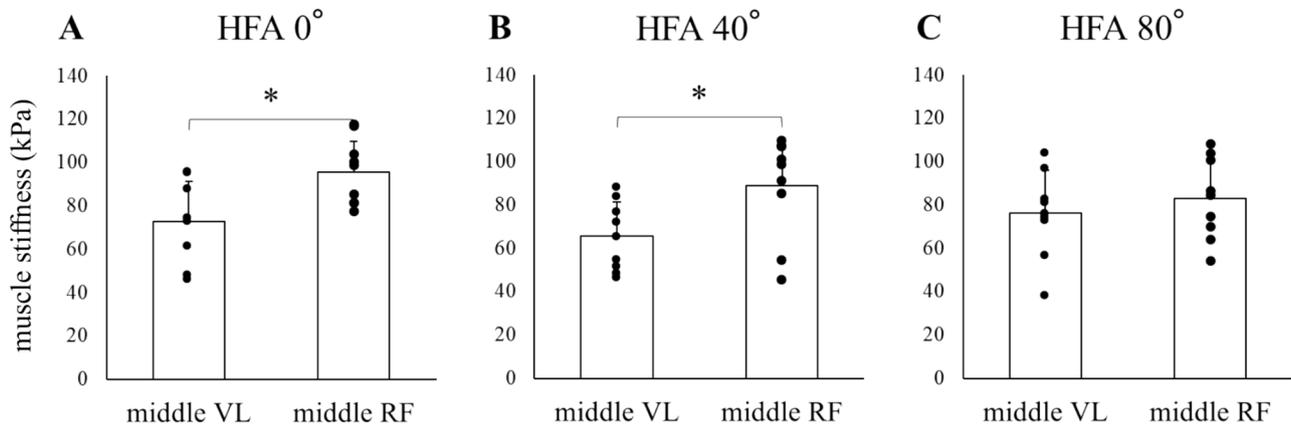
In the middle thigh region, the muscle stiffness of the middle RF was higher than that of the middle VL at HFA of 0° and 40° under the KFA 30° condition (HFA 0°:  $p = 0.035$ ,

**Table 3** Muscle stiffness during isometric KE for each joint angle condition

HFA	0°			40°			80°		
	30°	60°	90°	30°	60°	90°	30°	60°	90°
proximal RF	68.0±17.7	82.0±31.8	88.6±32.5	96.1±23.7	88.1±17.4	85.3±13.4	96.5±12.2	101.4±30.6	89.2±16.8
middle RF	82.9±17.7	99.9±36.3	104.5±27.3	88.9±22.1	94.8±28.0	98.7±21.4	95.5±14.4	88.6±19.4	107.0±16.7
distal RF	83.8±22.8	99.2±38.8	91.9±19.8	91.5±18.0	95.7±20.9	92.7±15.8	92.±18.3	104.8±10.1	98.3±15.2
proximal VL	69.1±26.3	79.9±27.9	107.4±49.8	98.8±45.4	110.8±26.9	154.3±65.7	111.3±36.7	111.6±40.2	127.1±52.0
middle VL	76.1±19.7	70.5±23.3	93.4±46.4	65.7±15.7	97.3±26.4	94.9±34.1	72.7±18.4	84.8±33.7	99.7±43.3
distal VL	88.6±27.8	103.0±31.6	117.3±38.4	69.0±26.0	82.7±18.5	92.6±38.5	62.5±15.4	101.7±41.7	108.2±28.2
VM	71.1±22.0	98.6±33.4	104.8±5.2	90.7±59.0	89.8±21.3	98.3±19.9	81.1±41.7	76.9±29.7	91.2±39.5

HFA hip flexion angle, KFA knee flexion angle, RF rectus femoris, VL vastus lateralis, VM vastus medialis. Values are presented as mean ± standard deviation

## Middle thigh region at KFA30°

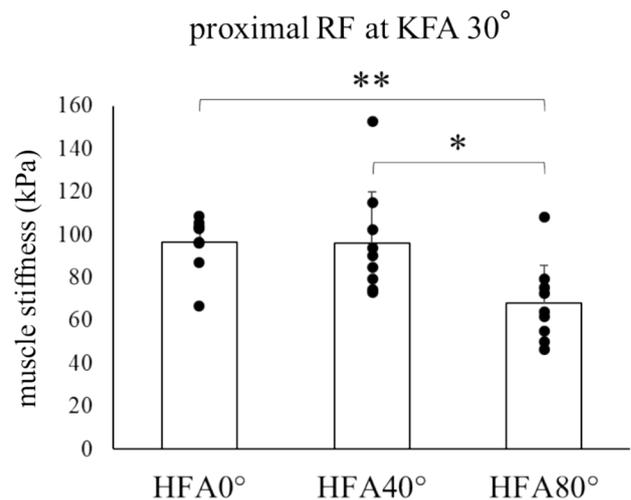


**Fig. 5** Comparison of muscle stiffness within each quadriceps head. KFA: knee flexion angle, HFA: hip flexion angle, VL: vastus lateral, RF: rectus femoris, \*  $p < 0.05$

$r = 0.667$ , Fig. 5A; HFA 40°:  $p = 0.046$ ,  $r = 0.655$ , Fig. 5B). However, no difference was observed at HFA 80° ( $p = 0.565$ ,  $r = 0.223$ ). Additionally, no differences were observed in muscle stiffness at KFA 60° (HFA 0°:  $p = 0.765$ ,  $r = 0.109$ ; HFA 40°:  $p = 0.957$ ,  $r = 0.020$ ; HFA 80°:  $p = 0.107$ ,  $r = 0.540$ ) or KFA 90° (HFA 0°:  $p = 0.699$ ,  $r = 0.155$ ; HFA 40°:  $p = 0.845$ ,  $r = 0.071$ ; HFA 80°:  $p = 0.572$ ,  $r = 0.204$ ). No differences were observed in the distal thigh region. For KFA 30°, the results were HFA 0°:  $p = 0.341$ , partial  $\eta^2 = 0.126$ ; HFA 40°:  $p = 0.667$ , partial  $\eta^2 = 0.048$ ; HFA 80°:  $p = 0.260$ , partial  $\eta^2 = 0.175$ . For KFA 60°, the results were HFA 0°:  $p = 0.242$ , partial  $\eta^2 = 0.183$ ; HFA 40°:  $p = 0.393$ , partial  $\eta^2 = 0.125$ ; HFA 80°:  $p = 0.966$ , partial  $\eta^2 = 0.004$ . For KFA 90°, the results were HFA 0°:  $p = 0.586$ , partial  $\eta^2 = 0.065$ ; HFA 40°:  $p = 0.897$ , partial  $\eta^2 = 0.013$ ; HFA 80°:  $p = 0.149$ , partial  $\eta^2 = 0.212$ . These results suggest that during the isometric KE, the muscle stiffness of the bi-articular RF was greater than that of the mono-articular VL.

### Comparison of muscle stiffness between each HFA within the same region

A main effect was observed in the proximal RF under the KFA 30° condition ( $p = 0.002$ , partial  $\eta^2 = 0.556$ ). Post hoc analysis indicated that muscle stiffness was higher at HFA 80° compared to HFA 0° ( $p = 0.009$ ,  $r = 0.747$ ) and HFA 40° ( $p = 0.039$ ,  $r = 0.829$ ) (Fig. 6). No differences were observed in the middle RF ( $p = 0.411$ , partial  $\eta^2 = 0.105$ ) or distal RF ( $p = 0.605$ , partial  $\eta^2 = 0.061$ ). Similarly, no differences were noted for KFA 60° (proximal RF:  $p = 0.424$ , partial  $\eta^2 = 0.102$ ; middle RF:  $p = 0.680$ , partial  $\eta^2 = 0.047$ ; distal RF:  $p = 0.645$ , partial  $\eta^2 = 0.053$ ) or KFA 90° (proximal RF:  $p = 0.939$ , partial  $\eta^2 = 0.008$ ; middle RF:  $p = 0.616$ , partial



**Fig. 6** Comparison of muscle stiffness between each HFA within the same region. KFA knee flexion angle, HFA hip flexion angle, RF rectus femoris, \*  $p < 0.05$ , \*\*  $p < 0.01$

$\eta^2 = 0.059$ ; distal RF:  $p = 0.619$ , partial  $\eta^2 = 0.058$ ). These results suggest that an increased HFA enhances the muscle stiffness of the proximal RF during isometric KE.

### Comparison of muscle stiffness between each region within the same HFA

Two-way ANOVA showed no differences in muscle stiffness across all joint angle conditions. For KFA 30°, the results were HFA 0°:  $p = 0.854$ , partial  $\eta^2 = 0.020$ ; HFA 40°:  $p = 0.836$ , partial  $\eta^2 = 0.022$ ; HFA 80°:  $p = 0.173$ , partial  $\eta^2 = 0.197$ . For KFA 60°, the results

**Table 4** ARV during isometric KE for each joint angle condition

ARV (mA)	0°			40°			80°		
	HFA	KFA		HFA	KFA		HFA	KFA	
	30°	60°	90°	30°	60°	90°	30°	60°	90°
proximal RF	0.11±0.03	0.13±0.03	0.11±0.03	0.13±0.04	0.10±0.04	0.10±0.05	0.11±0.04	0.12±0.05	0.11±0.05
middle RF	0.10±0.04	0.08±0.05	0.09±0.03	0.10±0.03	0.08±0.04	0.09±0.04	0.08±0.03	0.08±0.03	0.09±0.03
distal RF	0.09±0.06	0.12±0.04	0.11±0.03	0.09±0.04	0.09±0.05	0.10±0.03	0.08±0.03	0.09±0.03	0.12±0.05

ARV Average rectified value, HFA hip flexion angle, KFA knee flexion angle, RF rectus femoris. Values are presented as mean ± standard deviation

were HFA 0°:  $p = 0.299$ , partial  $\eta^2 = 0.140$ ; HFA 40°:  $p = 0.699$ , partial  $\eta^2 = 0.044$ ; HFA 80°:  $p = 0.551$ , partial  $\eta^2 = 0.072$ . For KFA 90°, the results were HFA 0°:  $p = 0.075$ , partial  $\eta^2 = 0.276$ ; HFA 40°:  $p = 0.222$ , partial  $\eta^2 = 0.171$ ; HFA 80°:  $p = 0.543$ , partial  $\eta^2 = 0.074$ .

## EMG

The ARV during the isometric KE for each joint angle condition are listed in Table 4 for each muscle region.

### Comparison of ARV between each HFA within the same region

A two-way ANOVA revealed no differences in ARV across all regions. For the proximal RF, the results were KFA 30°:  $p = 0.586$ , partial  $\eta^2 = 0.074$ ; KFA 60°:  $p = 0.358$ , partial  $\eta^2 = 0.137$ ; KFA 90°:  $p = 0.998$ , partial  $\eta^2 = 0.000$ . For the middle RF, the results were KFA 30°:  $p = 0.301$ , partial  $\eta^2 = 0.181$ ; KFA 60°:  $p = 0.340$ , partial  $\eta^2 = 0.143$ ; KFA 90°:  $p = 0.961$ , partial  $\eta^2 = 0.008$ . For the distal RF, the results were KFA 30°:  $p = 0.832$ , partial  $\eta^2 = 0.030$ ; KFA 60°:  $p = 0.180$ , partial  $\eta^2 = 0.217$ ; KFA 90°:  $p = 0.346$ , partial  $\eta^2 = 0.162$ .

### Comparison of ARV between each region within the same HFA

A two-way ANOVA revealed no differences in ARV across all joint angle conditions: for KFA 30°, HFA 0°:  $p = 0.224$ , partial  $\eta^2 = 0.221$ ; HFA 40°:  $p = 0.128$ , partial  $\eta^2 = 0.337$ ; HFA 80°:  $p = 0.061$ , partial  $\eta^2 = 0.330$ . For KFA 60°, HFA 0°:  $p = 0.096$ , partial  $\eta^2 = 0.374$ ; HFA 40°:  $p = 0.326$ , partial  $\eta^2 = 0.148$ ; HFA 80°:  $p = 0.060$ , partial  $\eta^2 = 0.430$ . For KFA 90°, HFA 0°:  $p = 0.147$ , partial  $\eta^2 = 0.380$ ; HFA 40°:  $p = 0.627$ , partial  $\eta^2 = 0.208$ ; HFA 80°:  $p = 0.117$ , partial  $\eta^2 = 0.301$ .

## Discussion

The RF was analyzed in three regions: proximal, middle, and distal, and muscle stiffness was compared across these regions. Muscle stiffness was higher in the RF compared to the VL. Specifically, in the proximal RF under the KFA 30° condition, muscle stiffness was higher at HFA 0° and 40° compared to HFA 80°. Conversely, the comparison of myoelectrical activity revealed no differences across all joint angles and regions.

The SWE measurements demonstrated region-specific differences in muscle stiffness across the quadriceps, with higher stiffness observed in the RF compared to the VL. This finding aligns with prior research by Wakahara et al. (2017) and Narici et al. (1996) who reported greater muscle hypertrophy in the RF compared to the vastus muscle group during the KE. In contrast, studies examining hypertrophy during squats have shown lower hypertrophy in the RF and greater in the VL (Earp et al. 2015; Enocson et al. 2005; Mangine et al. 2018). The differences in RF activation between single- and multi-joint exercises may be explained by the functional role of the muscles. During multi-joint movements, such as squats, the need to generate hip extension torque may reduce RF activity because the RF also functions as a hip flexor (Ema et al. 2016). This distinction highlights how muscle contraction dynamics vary between single- and multi-joint exercises.

Next, we discuss the longitudinal compartmentalized muscle stiffness of the RF observed using SWE, as the RF is a biarticular muscle, altering the HFA modifies its muscle length. We hypothesized that performing KE with greater hip extension, rather than the conventional KE with HFA 80°, would provide new insights into muscle contractile dynamics across the entire longitudinal region of the RF. Kodesho et al. (2021) used SWE to measure RF muscle stiffness during passive knee joint flexion at an HFA 0° and reported significantly higher muscle stiffness in the proximal RF compared to the distal RF. These findings suggest that altering the HFA influences the RF tension, particularly in the proximal region, leading to increased muscle stiffness. Consistent with this, the present study showed that muscle

stiffness in the proximal RF was higher at HFA 0° and 40°, where the hip angle was closer to extension, compared to HFA 80°. This result indicates that changes in the HFA affect the length of the RF, subsequently influencing proximal RF muscle stiffness. In support of this, our laboratory previously confirmed that performing the KE with hip extension increased proximal RF activity (Mitsuya et al. 2023). Therefore, the longitudinal section of the RF could be strengthened based on the hip joint angle. The KE should be performed at an HFA 0° to 40° to strengthen the RF. Moreover, Maeo et al. (2023) reported that shoulder joint angle variations influenced triceps brachii hypertrophy during elbow extension exercises and concluded that differences in muscle length affected the hypertrophic response. Similar findings have been reported for hamstrings (Maeo et al. 2021). These consistent results indicate that the RF shares similar mechanical characteristics with the triceps brachii and hamstrings, and alterations in muscle length influence contractile dynamics and stiffness.

Myoelectrical activity measurements revealed no differences across the joint angles and regions. The only previous study investigating the longitudinal compartmental myoelectrical activity of the RF during the KE with varying joint angles was conducted by Watanabe et al. (2014a), who reported no significant differences between joint angles or regions. Previous studies have primarily focused on non-trained participants, whereas it has been shown that myoelectrical activity dynamics can differ among trained individuals (Watanabe et al. 2015). To address this gap, the present study was conducted with trained participants. However, no longitudinal differences in myoelectrical activity were observed. This finding suggests that changes in joint angles may not significantly affect RF activity or that surface EMG may not be suitable for assessing longitudinal muscle contractile dynamics. Consistent with this, Zabaleta-Korta et al. used surface EMG to measure the longitudinal myoelectrical activity in the biceps brachii during inclined arm curls (performed during shoulder extension) and preacher arm curls (performed during shoulder flexion). Their findings also showed no significant longitudinal myoelectrical activity patterns (Zabaleta-Korta et al. 2024). In a related study, Zabaleta et al. (2023) examined muscle thickness changes in the biceps brachii after nine weeks of inclined and preacher arm curl training, measured via ultrasonography. The results indicated no significant changes in proximal bicep thickness, whereas distal bicep thickness increased more in the preacher arm curl than in the inclined arm curl. These findings suggest that observing longitudinal compartmental myoelectrical activity using surface EMG alone may be particularly challenging. Alternative imaging techniques, such as transverse relaxation time measurements using magnetic resonance imaging (MRI), may be better suited for assessing region-specific and longitudinal muscle

contractile dynamics. In summary, SWE may be a more effective method than EMG for detecting the effects of muscle length variations owing to joint angle adjustments on muscle contractile dynamics during resistance training. Future studies should consider the complementary use of SWE and EMG, and leverage the unique advantages of each technique for a comprehensive analysis.

The movements commonly performed in resistance training primarily involve isotonic contractions, which differ from the isometric contractions examined in this study. While isometric contractions are well-suited for investigating region-specific and longitudinally compartmentalized muscle contractile dynamics in bi-articular muscles using SWE and EMG, this distinction also represents a limitation of the study. To gain a more comprehensive understanding of the muscle contractile behavior during actual resistance training, it is necessary to investigate isotonic contractions. Additionally, we recommend estimating muscle transverse relaxation times using magnetic resonance imaging (MRI), both pre- and post-exercise, because this method can capture the physiological changes associated with isotonic contractions more effectively. Another limitation of this study is that only the MVCs were measured. However, the validity of comparing EMG and SWE data under MVC conditions has been established in a previous study (Nordez and Hug 2010), and we do not believe that this factor significantly affected the results. Further studies should aim to explore region-specific and longitudinally compartmentalized muscle contractile dynamics using various measurement techniques to expand on the findings of this study. Another limitation of this study was the exclusion of female participants. In the future, we will explore whether the results can be generalized by expanding the target population to include both females and untrained individuals.

## Conclusion

During KE, the RF contracts more strongly than the VL, and performing KE with hip extension increases the muscle stiffness of the proximal RF. Furthermore, region-specific and longitudinal muscle contractile dynamics, which were not detectable using EMG, were successfully observed using SWE. These findings highlight the importance of selecting appropriate measurement techniques to examine region-specific and longitudinal muscle contractile dynamics. Therefore, each region of the quadriceps and the longitudinal section of the RF can be strengthened according to the hip joint angle. We conclude that KE should be performed at an HFA of 0–40° to strengthen the RF.

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**Author contributions** All authors contributed to the study conception and design. Material preparation, data collection and analysis were performed by Hiroku Mitsuya, Koichi Nakazato and Takashi Okada. The first draft of the manuscript was written by Hiroku Mitsuya and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

**Data availability** The data generated during the current study are available from the corresponding author on reasonable request.

## Declarations

**Conflict of interest** This study did not receive any specific grants from funding agencies in the public, commercial, or non-profit sectors. The results of the current study do not constitute an endorsement of the product by the authors.

**Ethical approval** Examples of statements to be used when ethics approval has been obtained: All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki Declaration and its later amendments or comparable ethical standards. The study was approved by the Bioethics Committee of the Nippon Sport Science University (No. 018-H113). This study was performed in line with the principles of the Declaration of Helsinki. Approval was granted by the Ethics Committee of Nippon Sport Science University (November, 2018/ No. 018-H113). Approval was obtained from the ethics committee of Nippon Sport Science University. The procedures used in this study adhere to the tenets of the Declaration of Helsinki. The questionnaire and methodology for this study was approved by the Human Research Ethics committee of the Nippon Sport Science University (Ethics approval number: 018-H113).

**Contest to practice** Informed consent was obtained from all individual participants included in the study.

**Consent to participate** Patients signed informed consent regarding publishing their data and photographs.

## References

- Ateş F, Hug F, Bouillard K, Jubeau M, Frappart T, Couade M, Bercoff J, Nordez A (2015) Muscle shear elastic modulus is linearly related to muscle torque over the entire range of isometric contraction intensity. *J Electromyogr Kinesiol* 25(4):703–708. <https://doi.org/10.1016/j.jelekin.2015.02.005>
- Avrillon S, Hug F, Guilhem G (2018) Between-muscle differences in coactivation assessed using elastography. *J Electromyogr Kinesiol* 43:88–94
- Earp JE, Newton RU, Cormie P, Blazevich AJ (2015) Inhomogeneous quadriceps femoris hypertrophy in response to strength and power training. *Med Sci Sports Exerc* 47(11):2389–2397. <https://doi.org/10.1249/MSS.0000000000000669>
- Eby SF, Song P, Chen S, Chen Q, Greenleaf JF, An KN (2013) Validation of shear wave elastography in skeletal muscle. *J Biomech* 46(14):2381–2387. <https://doi.org/10.1016/j.jbiomech.2013.07.033>
- Enocson AG, Berg HE, Vargas R, Jenner G, Tesch PA (2005) Signal intensity of MR-images of thigh muscles following acute open- and closed chain kinetic knee extensor exercise – Index of muscle use. *Eur J Appl Physiol* 94(4):357–363. <https://doi.org/10.1007/s00421-005-1339-y>
- Gyftopoulos S, Rosenberg ZS, Schweitzer ME, Bordalo-Rodrigues M (2008) Normal anatomy and strains of the deep musculotendinous junction of the proximal rectus femoris: MRI features. *AJR Am J Roentgenol* 190(3):W182–W186. <https://doi.org/10.2214/AJR.07.2947>
- Hasselman CT, Best TM, Ct H, Martinez S, Garrett WE Jr (1995) An explanation for various rectus femoris strain injuries using previously undescribed muscle architecture. *Am J Sports Med* 23(4):493–499. <https://doi.org/10.1177/036354659502300421>
- Kodesho T, Taniguchi K, Kato T, Katayose M (2021) Intramuscular differences in shear modulus of the rectus femoris muscle during passive knee flexion. *Eur J Appl Physiol* 121(5):1441–1449. <https://doi.org/10.1007/s00421-021-04644-1>
- Maeo S, Huang M, Wu Y, Sakurai H, Kusagawa Y, Sugiyama T, Kanehisa H, Isaka T (2021) Greater hamstrings muscle hypertrophy but similar damage protection after training at long versus short muscle lengths. *Med Sci Sports Exerc* 53(4):825–837. <https://doi.org/10.1249/MSS.0000000000002523>
- Maeo S, Wu Y, Huang M, Sakurai H, Kusagawa Y, Sugiyama T, Kanehisa H, Isaka T (2023) Triceps brachii hypertrophy is substantially greater after elbow extension training performed in the overhead versus neutral arm position. *Eur J Sport Sci* 23(7):1240–1250. <https://doi.org/10.1080/17461391.2022.2100279>
- Mangine GT, Redd MJ, Gonzalez AM, Townsend JR, Wells AJ, Jajtner AR, Beyer KS, Boone CH, La Monica MB, Stout JR, Fukuda DH, Ratamess NA, Hoffman JR (2018) Resistance training does not induce uniform adaptations to quadriceps. *PLoS ONE* 13(8):e0198304. <https://doi.org/10.1371/journal.pone.0198304>
- Mitsuya H, Nakazato K, Hakkaku T, Okada T (2023) Hip flexion angle affects longitudinal muscle activity of the rectus femoris in leg extension exercise. *Eur J Appl Physiol* 123(6):1299–1309. <https://doi.org/10.1007/s00421-023-05156-w>
- Narici MV, Hoppeler H, Kayser B, Landoni L, Claassen H, Gavardi C, Conti M, Cerretelli P (1996) Human quadriceps cross-sectional area, torque and neural activation during 6 months strength training. *Acta Physiol Scand* 157(2):175–186. <https://doi.org/10.1046/j.1365-201X.1996.483230000.x>
- Nordez A and Hug F (2010) Muscle shear elastic modulus measured using supersonic shear imaging is highly related to muscle activity level. *J Appl Physiol* (1985) 108(5):1389–1394. <https://doi.org/10.1152/jappphysiol.01323.2009>
- Rickham PP (1964) Human experimentation. Code of ethics of the World Medical Association. Declaration of Helsinki. *Br Med J* 2(5402):177
- Tubbs RS, Stetler W Jr, Savage AJ, Shoja MM, Shakeri AB, Loukas M, Salter EG, Oakes WJ (2006) Does a third head of the rectus femoris muscle exist? *Folia Morphol (Warsz)* 65(4):377–380
- Wakahara T, Ema R, Miyamoto N, Kawakami Y (2017) Inter- and intramuscular differences in training-induced hypertrophy of the quadriceps femoris: Association with muscle activation during the first training session. *Clin Physiol Funct Imaging* 37(4):405–412. <https://doi.org/10.1111/cpf.12318>
- Ema R, Sakaguchi M, Akagi R, Kawakami Y (2016) Unique activation of the quadriceps femoris during single- and multi-joint exercises. *Eur J Appl Physiol* 116(5):1031–1041. <https://doi.org/10.1007/s00421-016-3363-5>
- Ema R, Sakaguchi M, Kawakami Y (2018) Thigh and psoas major muscularity and its relation to running mechanics in sprinters. *Med Sci Sports Exerc* 50(10):2085–2091. <https://doi.org/10.1249/MSS.0000000000001678>

- Watanabe K, Kouzaki M, Moritani T (2012) Task-dependent spatial distribution of neural activation pattern in human rectus femoris muscle. *J Electromyogr Kinesiol* 22(2):251–258. <https://doi.org/10.1016/j.jelekin.2011.11.004>
- Watanabe K, Kouzaki M, Moritani T (2014a) Non-uniform surface electromyographic responses to change in joint angle within rectus femoris muscle. *Muscle Nerve* 50(5):794–802. <https://doi.org/10.1002/mus.24232>
- Watanabe K, Kouzaki M, Moritani T (2014b) Regional neuromuscular regulation within human rectus femoris muscle during gait. *J Biomech* 47(14):3502–3508. <https://doi.org/10.1016/j.jbiomech.2014.09.001>
- Watanabe K, Kouzaki M, Moritani T (2015) Spatial EMG potential distribution of biceps brachii muscle during resistance training and detraining. *Eur J Appl Physiol* 115(12):2661–2670. <https://doi.org/10.1007/s00421-015-3237-2>
- Watanabe K, Kouzaki M, Moritani T (2018) Relationship between regional neuromuscular regulation within human rectus femoris muscle and lower extremity kinematics during gait in elderly men. *J Electromyogr Kinesiol* 41:103–108. <https://doi.org/10.1016/j.jelekin.2018.05.011>
- Yang D, Morris SF (1999) Neurovascular anatomy of the rectus femoris muscle related to functioning muscle transfer. *Plast Reconstr Surg* 104(1):102–106. <https://doi.org/10.1097/00006534-199907000-00015>
- Yoshitake Y, Takai Y, Kanehisa H, Shinohara M (2014) Muscle shear modulus measured with ultrasound shear-wave elastography across a wide range of contraction intensity. *Muscle Nerve* 50(1):103–113. <https://doi.org/10.1002/mus.24104>
- Zabaleta-Korta A, Fernández-Peña E, Torres-Unda J, Francés M, Zubillaga A, Santos-Concejero J (2023) Regional hypertrophy: The effect of exercises at long and short muscle lengths in recreationally trained women. *J Hum Kinet* 87:259–270. <https://doi.org/10.5114/jhk/163561>
- Zabaleta-Korta A, Latorre-Erezuma U, Fernández-Peña E, Torres-Unda J, Santos-Concejero J (2024) Regional hypertrophy of muscle cannot be predicted by surface electromyography. *Isokinet Exer Sci* 32(2):155–161. <https://doi.org/10.3233/IES-230079>

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