Title page

Title : The effects of a 4-week static stretching program on the individual muscles comprising the hamstrings

Running title: Effect of 4-week stretching on muscle hardness

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Key words: shear wave elastography, semitendinosus, semimembranosus, biceps femoris

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ABSTRACT

The aims of this study were to investigate the effects of a 4-week intervention of static stretching (SS) on muscle hardness of the semitendinosus (ST), semimembranosus (SM), and biceps femoris (BF) muscles. Shear elastic modulus was measured by using ultrasound shear wave elastography as the index of muscle hardness. Thirty healthy men (age, 22.7 ± 2.2 years) volunteered for this study and were randomly assigned to the SS-intervention group (n = 15) or the control group (n = 15). Subjects in the SS-intervention group received a 4-week stretch intervention for the hamstrings of their dominant leg. Shear elastic moduli of the hamstrings were measured at initial evaluation and after four weeks in both groups at a determined angle. In all muscles, the shear elastic modulus decreased significantly after SS intervention. The percentage change in the shear elastic modulus from the value at initial evaluation to after four weeks intervention was greatest in the SM. These results suggest that SS-intervention has chronic effects on reducing hardness of the hamstring muscle components, especially the SM muscle.

Key words: shear wave elastography, semitendinosus, semimembranosus, biceps femoris

INTRODUCTION

Static stretching (SS) is often used to increase flexibility of the hamstrings. There are many studies investigating the acute (Magnusson SP, Aagard P, Simonsen E, Bojsen-Moller F, 1998; Magnusson SP, Aagaard P, Nielson JJ, 2000; Matsuo et al. 2013) or chronic (Ben & Harvey 2010；Marshall PW, Cashman A, Cheema BS, 2011) effects of SS on the flexibility of the hamstrings. Most previous studies (Magnusson SP, Simonsen EB, Aagaard P, Sorensen H, Kjaer M, 1996; Folpp H, Deall S, Harvey LA, Gwinn T, 2006; Ylinen et al. 2009; Ben & Harvey 2010) reported that the joint range of motion increased, but the stiffness of the muscle–tendon unit MTU and passive torque did not decrease after a few weeks of SS intervention on the hamstrings. On the other hand, a recent study (Marshall et al. 2011) showed that stiffness of the hamstrings decreased after a 4-week SS intervention, which is inconsistent with previous studies.

Marshall et al. reported that the reason for the change in passive torque after their stretch intervention may be due to high volume of the exercise (30 sec×4 sets、4 exercises/session, self-stretching, 12 to 15 min/session). However, the interventions in studies reporting no change in passive torque are not necessarily low in volume, for instance, Ben & Harvey (2010) adopted interventions of 30 min/day, and Folpp et al. (2006) of 20 min/day. Since previous studies using passive torque as an index had shown conflicting results, we attempted to evaluate the effect of stretching from another viewpoint, focusing solely on muscles by measuring shear elastic modulus.

One of the major concerns is that the effect of SS on individual muscles of the hamstrings cannot be determined by measuring the effect of the stiffness of MTU and passive torque during stretching. The passive torque measured during knee extension or hip flexion is a sum of the passive resistance of all the properties including the whole hamstring muscles and the surrounding structures. With the development of the method evaluating the mechanical properties of the muscle using ultrasound shear wave elastography, shear elastic modulus, which is an index of muscle hardness of each muscle composing the hamstrings can be evaluated separately(Nakamura M, Ikezoe T, Tokugawa T, Ichihashi N, 2015). While shear wave elastograpy measures the muscle hardness in the direction perpendicular to the line of force during contractile loading, this value is found to be correlated to muscle stiffness along the line of the muscle-tendon unit. (Nakamura et al. 2014). Therefore, in this paper, shear elastic modulus is used as a value to estimate the muscle hardness or extensibility.

High injury rate of the hamstring muscle strain during sports activity is found

(Brooks JH, Fuller CW, Kemp SP, Reddin DB, 2006; Feeley et al. 2008; Ekstrand J, Hagglund M, Walden M, 2011), and there is a relevance between the injury cite and the characteristics of the sport. For example, sprinters have a high risk of straining the biceps femoris(BF) muscle (Verrall GM, Slavotinek JP, Barnes PG, Fon GT, 2003; Koulouris G, Connell DA, Brukner P, Schneider-Kolsky M, 2007). On the other hand, dancers and ballerinas have a high risk of straining for the semimembranosus(SM) muscle (Askling CM, Tengvar M, Saartok T, Thorstensson A, 2008). Investigating the effects of SS on individual muscles of the hamstrings would be valuable for the understanding and preventing of muscle strain in the components of the hamstrings. Our previous study using ultrasound shear wave elastography (Umegaki et al. 2015) reported that the hardness of individual muscles comprising the hamstrings is decreased immediately after 5 min of SS with hip flexion and knee extension, and that the SS maneuver may be most effective for the SM muscle. There is also a study which measured the immediate change in shear modulus of each hamstring muscles before and after 5 times of 90 seconds static stretching (Miyamoto, N., Hirata, K.,Kanehisa, H.,2015). This study found that hardness is reduced in SM and semitendinosus (ST) muscles, but not in the BF from hamstring stretching by passive knee extension maneuver. There are, however, no studies investigating the effects of a few-week SS program on the individual muscles of the hamstrings. The originality of this study is that the effect of stretching intervention was evaluated using shear elastic modulus as an index of muscle hardness and that we measured each component of the hamstrings separately, which was not found in previous studies.

The aims of this study were to investigate the effects of a 4-week intervention of SS on muscle hardness of the ST, SM, and BF muscles, and to examine the differences in the chronic effects between these muscles using shear elastic modulus measured by ultrasound shear wave elastography.

METHODS

Subjects

Thirty healthy men (age 22.7 ± 2.2 years; height 171.4 ± 4.6 cm; weight 63.7 ± 8.5 kg) participated in this study. The subjects were non-athletes who were not involved in any kind of regular stretching activity. Subjects with a history of neuromuscular disease, or musculoskeletal injury involving their lower limbs, were excluded from this study. The purpose and procedures were explained to all subjects, following which written informed consent was obtained. This study was approved by the ethics committee of Kyoto University Graduate School and the Faculty of Medicine (E-1524).

Table 1 near here

Experimental protocol

Subjects were randomly assigned to the SS-intervention group (n = 15) or the control group (n = 15). Subjects in the SS-intervention group performed a 4-week SS program for the hamstrings of their dominant leg, while subjects in the control group received no intervention. The dominant leg was defined as the leg with which the subject used to kick a ball. The subject did not participate in any other exercise during the intervention period. Shear elastic modulus of the hamstrings at 90° hip flexion and 45° knee flexion were measured before and after the 4-week intervention in both groups to evaluate muscle hardness. The subjects were instructed to remain relaxed during each of the measurements. The experimental protocol is shown in Fig. 1. To avoid the acute effects of SS, all measurements in the SS-intervention group were performed at least 24 h after the last SS session (Nakamura M, Ikezoe T, Takeno Y, Ichihashi N, 2012).

Figure 1 near here

SS protocol

The SS-intervention group performed the 4-week SS program using a dynamometer (Biodex system 4.0, Biodex Medical Systems Inc., USA). The subjects lay in supine position, with their pelvis inclined anteriorly by placing a wedge between the pelvis and the bed. The dominant lower leg was attached to the dynamometer, with the hip angle fixed at 90° flexion. The knee was then passively extended to the angle that the subjects could tolerate without pain or discomfort, which was determined for each subjects before intervention. This form of stretching was chosen for the reproducibility of the stretch intensity, stretching position with the pelvis stabilised, and for the consistency with the measurement position. In our previous study (Umegaki et al.2015), we reported that the shear elastic modulus of individual muscles of the hamstrings decreased after 5 min of SS. Therefore, in this study, SS of 5 min was undertaken three times per week for four weeks.

Measurement of shear elastic modulus

Shear elastic modulus of the ST, SM, and BF muscle bellies in the dominant leg was measured using ultrasound shear wave elastography (Aixplorer; SuperSonic Imagine, Axi-en-Provence, France). The measurement sites were defined as the midpoint of the femur from the greater trochanter to the medial epicondyle for the ST and SM muscles, and to the lateral epicondyle of the femur for the BF muscle. These anatomical points were confirmed by palpation and B-mode images. An ultrasound transducer (50 mm long SL-15-4 liner ultrasound transducer) was positioned on the measurement sites, parallel to the direction of the muscle fibers, which was confirmed by tracing several fascicles without interruption across the B-mode image. For each muscle, images were taken after the transducer was held at the measurement cite for around 5 s in order to confirm that the shear wave elastography(SWE) in the region of interest(ROI) showed stable color distribution. The measurements were taken twice for each muscle, and the mean values were used for statistical analysis. The region of interest was set near the centre of the muscle belly image, and the mean shear wave propagation speed (m/s) of an 11mmdiameter circle set near the centre of the ROI was automatically calculated. The accuracy of the measurement showed excellent reliability. ICC (1.1) and CV for ST, SM, and BF were 0.985 (1.8%), 0.971 (1.9%), and 0.983 (1.6%), respectively.

In the supine position, shear elastic modulus of the ST, SM, and BF muscles was measured at 90° hip flexion and 45° knee flexion. The shear elastic modulus was measured in <10 s for each measurement to avoid effects on muscle flexibility. Measurement of each component was performed in random order to exclude any effects of the measurement order. Each component was measured twice, and the mean values were used in the statistical analysis. None of the subjects expressed discomfort or pain, and the shear elastic modulus values did not reach the upper limit value during the measurement.

Inter-day reliability of the measurements

To determine inter-day reliability of the measurements of the shear elastic modulus, measurements were taken in seven healthy men (age, 23.6 ± 1.8 years; height, 173.4 ± 3.7 cm; weight, 68.4 ± 7.2 kg) once each on two different days.

A priori sample size calculation

We calculated the sample size needed for split-plot analysis of variance (ANOVA) (alpha error = 0.05, power = 0.80, effect size = 0.4 [large]), and the requisite number of subjects for this study was 14 in each group. We chose a large effect size on the basis of previous studies (Marshall et al. 2011; Akagi & Takahashi 2013).

Statistical analysis

Statistical analyses were performed using SPSS (version 18.0, SPSS Japan INC., Tokyo, Japan). The inter-day reliability of the measurements was assessed using the intraclass correlation coefficient (ICC). Differences between the SS-intervention group and the control group, with regard to subject characteristics and all outcomes at baseline, were assessed using an unpaired t-test. For the shear elastic modulus of each muscle, split-plot ANOVA using two factors (groups [the SS-intervention group and the control group] and test time [before and after four weeks]) was used to analyze interaction effects. When a significant interaction was observed, a paired t-test was used to determine the differences between the value at baseline and after four weeks in both the SS-intervention group and the control group. For shear elastic modulus in the SS-intervention group, two-way repeated measures ANOVA using two factors (test time [before and after four weeks] and muscles [ST, SM, and BF muscles]) was used to analyze interaction effects. When a significant interaction was observed, the Bonferroni's post-hoc test was used to compare the difference in the percentage change of shear elastic modulus between muscles. Percentage change of shear elastic modulus was calculated using the following equation: Percentage change (%) = (at baseline − after four weeks) / at baseline × 100. Differences were considered statistically significant at an alpha level of 0.05.

RESULTS

Characteristics of the subjects

No participants withdrew from the study and all subjects completed the SS program. Therefore, all data of the SS-intervention group (n = 15) and the control group (n = 15) were used for statistical analysis. The characteristics of the subjects are shown in Table 1. There were no statistically significant differences between groups in the measured characteristics.

Inter-day reliability of the measurements

The ICC(1,1)s of inter-day measurements are shown in Table 2. The ICC(1,1) ranged from 0.818 to 0.959 for the shear elastic modulus of each muscle. Since ICC value of <0.40 is generally considered as poor reliability, 0.40-0.75 as moderate to good, and >0.75 as excellent reliability (Leong et al., 2013), the results showed excellent reliability.

Table 2 near here

Shear elastic modulus

The shear elastic modulus values are shown in Table 3. There were no significant group differences in shear elastic modulus of any of the muscles at baseline. Split-plot ANOVA indicated a significant interaction in all muscles (ST: $F(1,28) = 7.2$, $p = 0.01$; SM: $F = 30.2$, $p < 0.01$; BF: $F(1,28) = 10.1$, $p < 0.01$). In the SS-intervention group, shear elastic modulus of all the muscles significantly decreased after the intervention. In the control group, there were no changes in shear elastic modulus for any of the muscles.

Table 3 near here

As for the percentage change in the shear elastic modulus in the SS-intervention group, two-way ANOVA indicated a significant interaction ($F = 17.6$, $p < 0.01$). The Bonferroni's post-hoc test indicated that the percentage change in the shear elastic modulus of the SM muscle was significantly higher than that of the ST and BF muscles (Fig. 2).

Figure 2 near here

DISCUSSION

The results of this study showed that shear elastic modulus of the ST, SM, and BF muscles decreased after the 4-week SS intervention, which suggests that the SS program reduced the hardness of all muscles comprising the hamstrings. Although past studies (Magnusson et al. 1996;Folpp et al. 2006; Ylinen et al. 2009;Ben & Harvey 2010;) have investigated the chronic effects of SS on the MTU of the hamstrings; to the best of our knowledge, this is the first report investigating the chronic effects of SS on the individual muscles comprising the hamstrings using ultrasound shear wave elastography. Our results contradict to the majority of the previous studies on stretching of the hamstring muscles that reported no change in stiffness (Magnusson et al. 1996; Folpp et al. 2006; Ylinen et al. 2009; Ben & Harvey 2010). The intervention used in our study is not greater in volume compared to the studies reporting no change. Therefore, we assume that the difference may be due to the index used: shear elastic modulus or passive torque, the former focusing on the muscle itself, and the latter focusing on the whole muscle-tendon unit and the surrounding structure. The current study indicates that 4 weeks of stretching intervention for the hamstrings increases flexibility in the hamstrings muscle, especially in SM.

In this study, the shear elastic modulus as the index of the individual muscle hardness, reduced in all ST, SM and BF after the SS intervention. Based on the previous study indicating the positive correlation of the shear elastic modulus and passive torque during stretching of the muscle (Nakamura et al, 2014), the current result suggested that the 4 weeks SS intervention improved the flexibility of the hamstrings.

Our results showed that the shear elastic modulus of the ST, SM, and BF muscles decreased after the SS intervention, which was similar to a previous study (Akagi & Takahashi 2013) investigating the chronic effects of SS on the hardness of the medial and lateral gastrocnemius muscle. Therefore, our SS protocol (5 min of SS, three times per week for four weeks) may be effective for reduced hardness of all muscles comprising the hamstrings.

As for differences in the effects of the SS intervention between the muscles, the percentage change in shear elastic modulus of the SM muscle was significantly greater than that of the ST and BF muscles. These results support our previous study (Umegaki et al.2015), which reports that the acute effect of SS is greatest in the SM muscle among the hamstring muscles. In our previous study (Umegaki et al.2015), it was estimated that passive tension applied to the SM muscle was highest during this SS maneuver, therefore; the chronic effect of the SS intervention might also be greatest in the SM muscle.

Poor flexibility of the hamstrings increases the risk of muscle strain (Witvrouw E, Danneels L, Asselman P, D'Have T, Cambier D, 2003; Bradley & Portas 2007). Our results indicated that SS program reduced the hardness of the individual muscles comprising the hamstrings. Therefore, the SS intervention protocol may be useful alter the incidence of hamstrings muscle strain. In particular, considering that the effect of the SS intervention was greatest in the SM muscle, the SS intervention protocol in this study may be more effective for preventing muscle strain in dancers and ballerinas, who are at high risk of straining the SM muscle (Askling CM, Tengvar M, Saartok T, Thorstensson A, 2008). We, however, did not investigate whether this SS intervention protocol is effective for preventing hamstring muscle strain, and therefore further research is still required to clarify the effect of the SS program on injury prevention.

This study also has some limitations that remain to be addressed. First, because the subjects in the present study were healthy young men, it is unknown whether the SS protocol used in this study could be effective for women, the elderly, or subjects with a history of hamstring muscle strain. Second, it could not be demonstrated whether passive tension applied to the SM muscle among the hamstrings was the highest during this SS maneuver, because shear elastic modulus during SS was not measured. Furthermore, the experimenter who performed the measurements and data analysis was not masked to the nature of the study design.

5 CONCLUSIONS

Our results indicate that the SS intervention significantly decreased the shear elastic modulus of all the hamstring muscles. The percentage change in the shear elastic modulus after four weeks was greatest in the SM. Since poor flexibility of the hamstrings is known to increases the risk of muscle strain (Witvrouw et al. 2003; Bradley & Portas 2007), our results indicated that SS program may contribute to alter the risk of injury.

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Table 1. Characteristics of the subjects

	SS-intervention group	Control group
Age (years)	22.5 ± 2.9	22.9 ± 1.2
Height (cm)	171.9 ± 5.3	171.0 ± 3.8
Weight (kg)	65.7 ± 8.4	61.7 ± 8.3

Values are expressed as mean ± SD (standard deviation).

Table 2. Inter-day reliability of the measurements

	ICC(1,1)	CV(%)
Shear elastic modulus of ST	0.938	4.2
Shear elastic modulus of SM	0.818	3.9
Shear elastic modulus of BF	0.959	2.1

ST: semitendinosus, SM: semimembranosus, BF: biceps femoris, ICC: intraclass correlation coefficient, **CV**: coefficient of variation

Table 3. The effects of SS on shear elastic modulus

	SS-intervention group		Control group	
	baseline	after four weeks	baseline	after four weeks
ST (kPa)	54.5 ± 14.4	47.5 ± 12.5**	48.7 ± 17.2	48.2 ± 14.3
SM (kPa)	126.6 ± 25.4	99.4 ± 18.9**	120.7 ± 39.5	118.0 ± 25.4
BF (kPa)	95.9 ± 28.0	82.3 ± 22.5**	85.6 ± 25.5	83.6 ± 20.5

Values are expressed as mean ± SD (standard deviation).

** $p < 0.01$; significant difference compared with baseline.

ST: semitendinosus, SM: semimembranosus, BF: biceps femoris

Figure 1. Flow chart of the experimental protocol

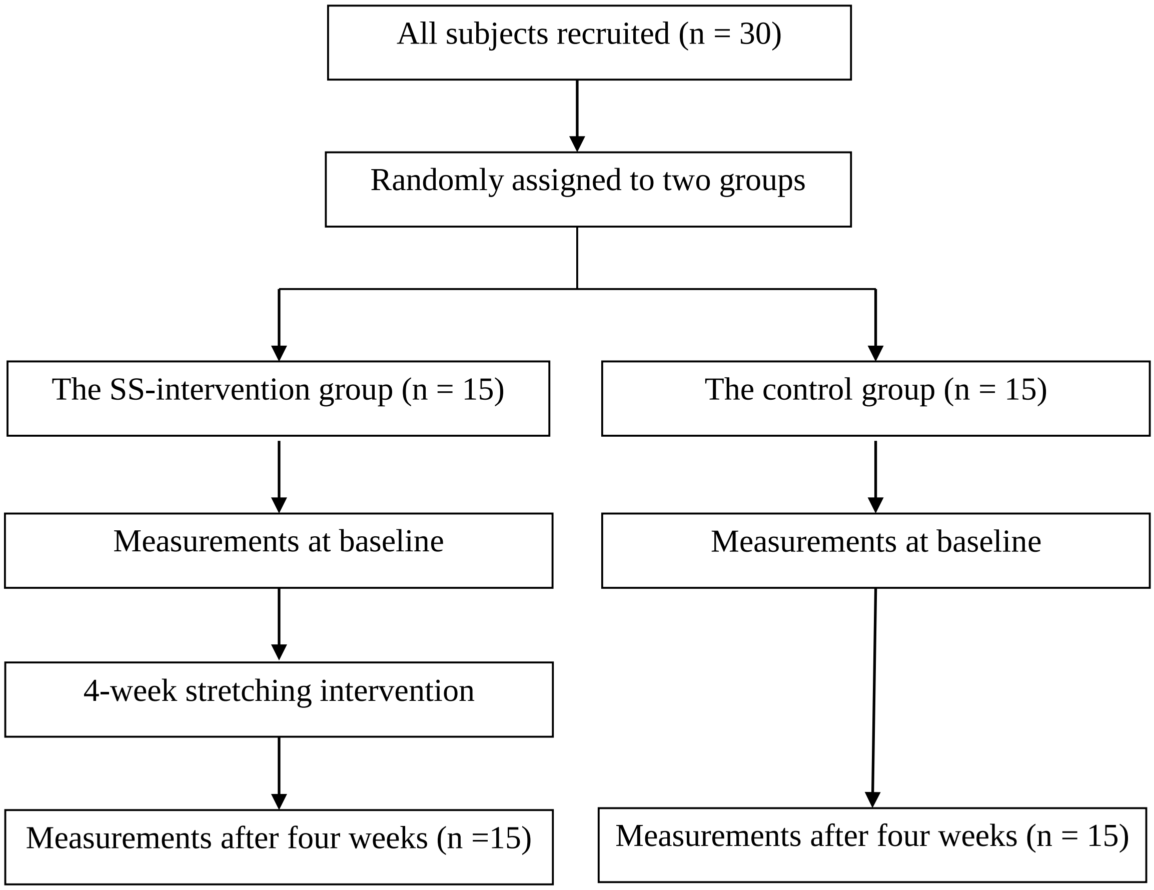
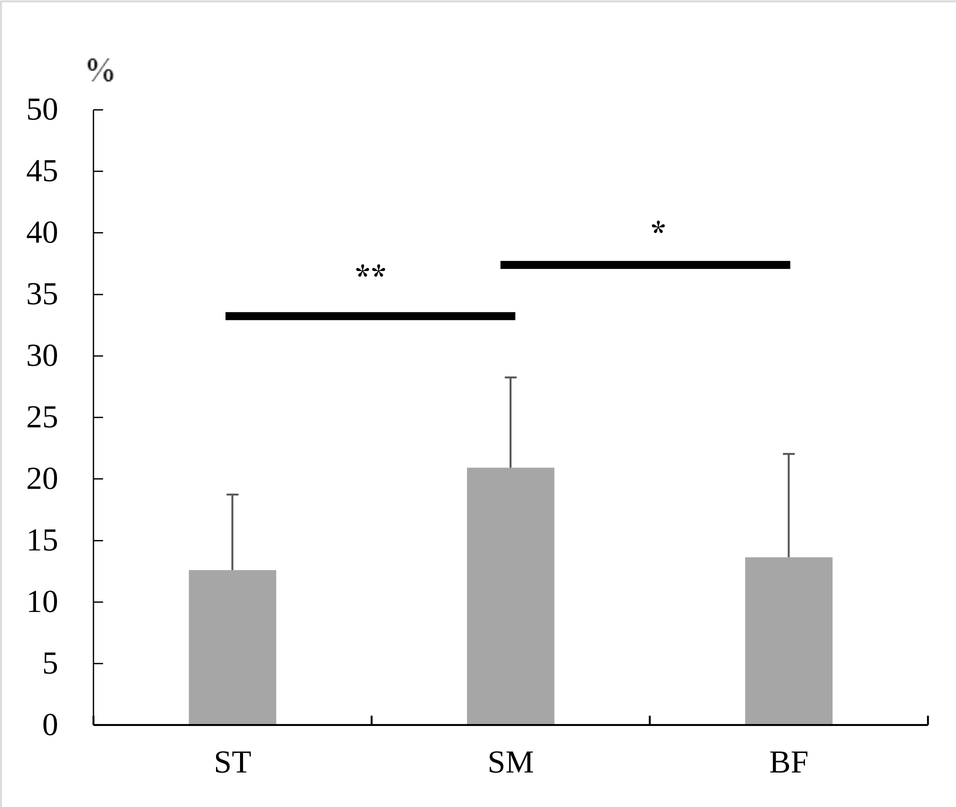


Figure 2. Differences in the percentage change of shear elastic modulus between muscles



Values are expressed as mean ± SD (standard deviation).

* $p < 0.05$

** $p < 0.01$

ST: semitendinosus, SM: semimembranosus, BF: biceps femoris