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Changes in Passive Properties of the Gastrocnemius Muscle-Tendon Unit During a 4-Week Routine Static Stretching Program.

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Changes in passive properties of the gastrocnemius muscle–tendon unit during a 4-week routine static stretching program

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Abstract

Context: Static Stretching (SS) is commonly performed within a warm-up routine to increase the range of motion (ROM) of a joint and to decrease muscle stiffness. However, the time course of changes in ankle dorsiflexion (DF) ROM and muscle stiffness during a routine SS program is unclear.

Objective: The present study investigated changes in ankle DF ROM, passive torque at DF ROM, and muscle stiffness during a routine SS program performed three times weekly for 4 weeks.

Design: A quasi-randomized controlled trial design.

Participants: The subjects comprised 24 male volunteers (age 23.8 ± 2.3 years; height 172.0 ± 4.3 cm; body mass 63.1 ± 4.5 kg) randomly assigned to either a group performing a 4-week stretching intervention program (SS group) or a control group.

Main Outcome Measures: The DF ROM, passive torque, and muscle stiffness were measured during passive ankle dorsiflexion in both groups using a dynamometer and ultrasonography once weekly during the 4-week intervention period.

Results: In the SS group, DF ROM and passive torque at DF ROM significantly increased after 2, 3, and 4 weeks compared with the initial measurements. Muscle stiffness also decreased significantly after 3 and 4 weeks in the SS group. However,

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there were no significant changes in the control group.

Conclusions: Based on these results, the SS program effectively increased DF ROM and decreased muscle stiffness. Furthermore, an SS program greater than 2 weeks duration effectively increased DF ROM and changed the stretch tolerance, and an SS program greater than 3 weeks in duration effectively decreased muscle stiffness.

Key words: time course, muscle stiffness, stretch tolerance, ultrasound

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Stretching is commonly performed within a warm-up routine to increase joint flexibility, improve performance, and reduce injury risk. Numerous previous studies reported that static stretching (SS) increased the joint range-of-motion (ROM) both acutely[1, 2] and following routine SS[3-5]. Hamstring and plantar flexor muscle stretching increased knee extension and ankle dorsiflexion (DF) ROM both acutely and chronically, according to systematic literature reviews[6, 7]. Potentially, the joint ROM increase following SS may be caused by: decreased passive torque, muscle-tendon unit (MTU), and muscle stiffness; and changes in psychological factors such as pain and stretch tolerance[8, 9].

Previous studies evaluating acute effect of SS reported that a 3- to 5-min duration decreased MTU and muscle stiffness[9-13]. In a study examining MTU stiffness over time following SS (constant-torque stretching) at 2, 4, and 8 min, the initial decrease in MTU stiffness dissipated in less than 10 min following a 2 min SS, but after 4- and 8-min SS, the effect was maintained for 10 min[14]. We recently reported that decreased MTU and muscle stiffness were maintained for 10 min following a 5-min constant-angle SS session[12], which is consistent with a prior study[14]. However, Mizuno et al. (2013) reported that the MTU and muscle stiffness decreases following a 5-min constant-angle SS disappeared within 10–15 min, whereas the increased ROM persisted for 30 min. These results suggested that the increased ROM immediately following SS

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may be attributed to changes in both MTU viscoelasticity and stretch tolerance, and the ROM increase at 15–30 min after SS could be attributed only to a stretch tolerance change. These studies concluded that the retention time of the acute effects of SS was shorter for MTU viscoelasticity than for stretch tolerance.

Other studies have similarly examined the chronic effect of SS. For example, previous studies reported that passive torque and MTU stiffness decreased after a 3- to 6-week routine SS program[15-18]. In addition, stretch tolerance changed after a 2- to 6-week routine SS program[19-21]. We reported that muscle stiffness decreased after 4 weeks of routine SS[22]. However, the time course of changes in muscle stiffness and stretch tolerance immediately following SS were discordant[9]; thus, a discrepancy in the time course of muscle stiffness and stretch tolerance changes may also occur during a routine SS program. Furthermore, the ideal SS program duration required to change the ROM, muscle stiffness, and stretch tolerance is unclear.

This study investigated changes in the gastrocnemius MTU passive properties over time, including DF ROM, muscle stiffness, and stretch tolerance during a 4-week SS program. A previous study showed that the acute effects of SS on muscle stiffness dissipated faster than the stretch tolerance[9]. Therefore, we hypothesized that muscle stiffness changes caused by the routine SS program would occur later during the

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program than the stretch tolerance changes.

Methods

Study Design

A quasi-randomized controlled trial design was used to investigate changes in ankle DF ROM, passive torque at DF ROM, and muscle stiffness during a routine SS program performed three times weekly for 4 weeks. The gastrocnemius MTU passive properties (DF ROM, passive torque at DF ROM, and muscle stiffness) were measured at the initial evaluation and once weekly over 4 weeks in both groups. As an *a priori* sample size calculation, we calculated the sample size that was needed for split-plot analysis of variance (ANOVA) [alpha error = 0.05, power = 0.80, effect size = 0.25 (middle)] using G*Power 3.1 software (Heinrich Heine University, Düsseldorf, Germany). The results showed that the requisite number of subjects for this study was 11 for each group. Considering a possible dropout, 12 participants were recruited for each group. After an initial evaluation of MTU passive properties, participants were randomly allocated in a 1:1 ratio to either the SS group (N = 12) or the control group (N = 12) using the alternation method. To control for immediate SS impacts, all procedures in the SS group were performed at least 24 h after the last SS session[22]. The subjects were instructed not

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to initiate any other stretching or strength training program during the experimental period.

Participants

Twenty-four healthy male volunteers who were non-athletes participated in this study (age 23.8 ± 2.3 years; height 172.0 ± 4.3 cm; body mass 63.1 ± 4.5 kg). Subjects with a history of neuromuscular disease or lower extremity musculoskeletal injury were excluded. All subjects participated in sports at a recreational level and had not been involved in any regular resistance or flexibility training. Written informed consent was obtained from all subjects. Subject demographics of each group are summarized in Table 1. There were no significant demographic differences between the two groups based on an unpaired *t*-test. In addition, this study was approved by the ethics committee.

Procedures

Assessment of DF ROM and passive torque at the DF ROM

The subjects laid in a prone position on a dynamometer table (MYORET RZ-450, Kawasaki Heavy Industries, Kobe, Japan) secured at the hips with adjustable lap belts.

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The dominant knee was maintained in full extension, and the ipsilateral foot was securely attached to the dynamometer footplate with adjustable lap belts to prevent the heel that moving away from the footplate. The ankle was passively dorsiflexed at a constant 5°/s velocity beginning at a 30° plantar flexion until reaching the DF ROM. In this study, DF ROM was defined as the angle where subjects experienced discomfort without pain[9, 11, 12, 14]. The passive torque at ankle angles of 0°, 30° dorsiflexion, and DF ROM were measured during the procedure using a dynamometer. Passive torque at DF ROM served as the index of stretch tolerance; a passive torque increase at the DF ROM indicated modified stretch tolerance[9].

Muscle stiffness assessment

Myotendinous junction (MTJ) displacement at the gastrocnemius muscle medial head during passive ankle dorsiflexion was determined using B-mode ultrasonography (Famio Cube SSA-520A; Toshiba Medical Systems Corporation, Tochigi, Japan). MTJ was visualized on a continuous sagittal plane ultrasound image using an 8-MHz linear-array probe. An acoustically reflective marker was placed on the skin under the ultrasound probe to confirm that the probe remained stable during measurement. The MTJ displacement was defined as the distance between the MTJ and the reflective

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marker. A customized fixation device secured the probe to the skin. Ultrasound MTJ images were quantified using open-source digital measurement software (Image J, National Institutes of Health, Bethesda, MD, USA). To ensure accuracy, the MTJ was identified at the inner fascial edge surrounding the muscle at its fusion to the tendon; displacement was measured during 0° and 30° ankle dorsiflexion. Muscle stiffness was calculated by dividing the passive torque change during 0–30° ankle dorsiflexion by the MTJ displacement[12].

Surface electromyography (EMG)

Electromyography (EMG) (TeleMyo2400; Noraxon USA Inc., Scottsdale, AZ, USA) confirmed that the subjects were relaxed and muscles were inactive during passive ankle dorsiflexion. Surface electrodes (Blue Sensor M, Ambu, Denmark) at a 2.0-cm interelectrode distance were placed on the medial and lateral gastrocnemius muscle bellies[13].

An EMG was recorded from the muscle bellies while the subjects performed an isometric maximum voluntary contraction (MVC), obtained during maximal isometric plantar flexion with the ankle at 0°. Strong verbal encouragement was provided during the contraction to promote maximal effort. EMG activity was calculated from the root

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mean square (RMS), and a full wave rectification was performed using an RMS smoothing algorithm at a 50-ms window interval. EMG activity recorded during passive ankle dorsiflexion was expressed as a percentage of MVC. The EMG sampling rate was 1500 Hz.

Static stretching (SS) program

Subjects in the SS group were placed in a prone position with the knee extended, similar to conditions during the DF ROM and passive torque measurements. During SS, the ankle was passively dorsiflexed, starting from 30° of plantar flexion to the DF ROM, and was held at the DF ROM for 30 s, e,g, constant-angle stretching method. We previously confirmed that an SS greater than 2 min significantly decreased muscle stiffness [23]. Therefore, the 30-s maneuver was repeated four times, 2 min in total. A previous study reported that stretching exercises performed three times weekly were sufficient to improve ROM compared to stretching once weekly[5]. Therefore, the SS maneuver was performed three times weekly over a 4-week period. The sessions were conducted every 2 or 3 days. Subjects in the control group did not receive any intervention.

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Measurement reliability

All measurements were performed by the same experienced examiner. We selected seven subjects (age, 23.8 ± 1.1 years; height, 172.7 ± 4.9 cm; body mass, 65.2 ± 2.8 kg) from the control group and adopted the initial and 1 week data for the reliability analysis.

Statistical analysis

SPSS (version 17.0; SPSS Japan Inc., Tokyo, Japan) was used for statistical analyses. Measurement reliability was assessed using the intraclass correlation coefficient (ICC [1, 1]). The Shapiro–Wilk test was performed to evaluate the normality of the data, and the assumption was met for almost all variables, suggesting the use of a parametric analysis. Differences between the SS and control groups for all variables relative to the initial evaluation were assessed with an unpaired *t*-test. Split-plot ANOVA and one-way repeated ANOVA compared the SS and control groups over time and the initial evaluation vs. data at 1, 2, 3, and 4 weeks. When one-way repeated ANOVA indicated a significant effect associated with time, the Dunnett's multiple comparison test was employed to determine the change time course compared with the initial evaluation.

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Differences were considered statistically significant at an alpha level of $p < 0.05$. Descriptive data are shown as mean ± standard deviation.

Results

Reliability assessment

Measurement reliability assessments are summarized in Table 2. The ICC (1, 1) was 0.836 (95% confidence interval [CI]; 0.464–0.960) for DF ROM, 0.942 (95% CI; 0.782–0.986) for passive torque at DF ROM, and 0.941 (95% CI; 0.779–0.986) for muscle stiffness.

DF ROM, passive torque at DF ROM, and muscle stiffness changes over time

There were no significant differences between the two experimental groups in all variables relative to the initial evaluation. The DF ROM, passive torque at DF ROM, and muscle stiffness changes over time in both groups are shown in Table 3. The split-plot ANOVA indicated that there were significant group × time interaction effects for DF ROM, passive torque at DF ROM, and muscle stiffness ($F = 20.6$, $p < 0.01$, $\eta_p^2 = 0.483$; $F = 5.88$, $p < 0.01$, $\eta_p^2 = 0.211$; and $F = 11.0$, $p < 0.01$, $\eta_p^2 = 0.334$, respectively). There was also a significant time effect on DF ROM, passive torque at DF ROM, and

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muscle stiffness in the SS group, but there was no significant time effect on the variables in the control group.

In the SS group, the DF ROM significantly increased after 2 weeks ($p < 0.05$), 3 weeks ($p < 0.01$), and 4 weeks ($p < 0.01$). Similarly, the passive torque at DF ROM significantly increased after 2 weeks ($p < 0.05$), 3 weeks ($p < 0.01$), and 4 weeks ($p < 0.01$). In addition, muscle stiffness significantly decreased after 3 ($p < 0.05$) and 4 weeks ($p < 0.05$).

EMG activity

The GM and LG EMG activities were <2% MVC, which confirmed a lack of contractile contribution to the DF ROM, passive torque, and muscle stiffness.

Discussion

We investigated the gastrocnemius MTU passive property changes during a 4-week routine SS program. The major study finding was that the DF ROM and passive torque at DF ROM changes occurred earlier than the muscle stiffness change during the routine SS program. Although previous studies investigated the acute impact of SS on passive properties[9, 10, 12, 14], this is the first known study demonstrating the time course of MTU

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passive property changes during a 4-week routine SS program in vivo.

Our study revealed two-way ANOVA (group × time) interactions in the DF ROM and passive torque at DF ROM. In addition, the multiple comparison test indicated that DF ROM and passive torque at DF ROM in the SS group significantly increased after 2–4 weeks compared with the initial evaluation, with no significant changes in the control group. These results suggest that a 2-week or longer SS program effectively increases the DF ROM, which is consistent with previous studies[3-5]. The passive torque at DF ROM, which indicated stretch tolerance, also increased after 2 weeks of the SS program. These results suggest that a 2-week or longer SS program may be required to change DF ROM and stretch tolerance. Although the mechanism of stretch tolerance change after routine SS program is unknown, afferent input from muscles and joints during stretching inhibits signals from nociceptive fibers, which may increase pain thresholds[24-26].

In the evaluation of effects of routine SS program on muscle stiffness, there was a two-way ANOVA (group × time) interaction observed. Furthermore, a multiple comparison test revealed that muscle stiffness significantly decreased after 3 and 4 weeks in the SS group. These results suggest that a SS program greater than 3 weeks effectively decreases muscle stiffness. The underlying mechanism of this change is

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unknown, but previous studies reported that the decreased muscle stiffness acutely and chronically following an SS program might be associated with alterations in the properties of intramuscular connective tissue properties rather than muscle fiber lengthening[11, 12, 22]. Therefore, the muscle stiffness decrease after 3 weeks of routine SS may also reflect a change in intramuscular connective tissue flexibility. Our results showed that muscle stiffness significantly decreased after 3 and 4 weeks, with no change observed during the initial 1–2 weeks. There may be a dose-response relationship between the SS duration and the MTU stiffness response[14]. Therefore, a 1- to 2-week SS program may be insufficient to decrease muscle stiffness, and an SS program lasting at least 3 weeks may be necessary using the current study protocol.

Our results showed a discrepancy the between muscle stiffness and stretch tolerance changes during the 4-week routine SS program. In particular, more than 2 weeks of routine SS increased passive torque at DF ROM, which indexes stretch tolerance, but more than 3 weeks of routine SS was required to decrease muscle stiffness. These results show that the stretch tolerance changed earlier than muscle stiffness during a routine SS program, which confirms our hypothesis, though the underlying mechanism is unclear. As for the acute effect of SS, Mizuno et al. (2013) reported that the acute benefits of 5-min SS on muscle stiffness persisted for a shorter

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time than the stretch tolerance benefits. Potentially, the stretch tolerance change occurred earlier than the muscle stiffness decrease during the routine SS program. Decreased muscle stiffness can be beneficial in improving athletic performance or preventing injury[27, 28]; further study is needed to clarify the long-term effects of routine SS not only on passive properties, such as DF ROM and muscle stiffness, but also on improving performance and preventing injury. Notably, under the current study protocol of 2-min SS, three times weekly over a 4-week period, it is unclear whether the same decreased muscle stiffness could be realized under an SS program with longer single sessions combined with a shorter program duration. Additional study determining the ideal SS program duration and intervention frequency maximizing the muscle stiffness decrease is needed.

Our results showed that there was a discrepancy in the time course of muscle stiffness and stretch tolerance changes during a routine SS program, i.e., the stretch tolerance changed earlier than muscle stiffness during a routine SS program. In addition, decreased muscle stiffness can be beneficial in improving athletic performance or preventing injury[27, 28]. Therefore, taken together, it was suggested that it is necessary to perform the routine SS program to cause a decrease in muscle stiffness in order to improve the athletic performance or prevent injury.

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This study had some limitations. First, the examiner taking measurements was not blinded to the group. Therefore, a bias in the results cannot be completely discounted. Second, we have not investigated the time course of changes in passive properties during a detraining period after 4 weeks of static stretching program. Therefore, further research is required to determine the prolonged effect of SS program on passive properties.

Conclusion

This study investigated the change in the gastrocnemius MTU passive properties, the DF ROM, muscle stiffness, and stretch tolerance, during a 4-week routine SS program. Our results showed that the changes in muscle stiffness and stretch tolerance occur at different speeds during the 4-week routine SS program. In particular, these results suggest that a SS program greater than 2 weeks effectively increases DF ROM and changes stretch tolerance, and a SS program greater than 3 weeks is needed to decrease muscle stiffness.

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Table 1. Subject demographics.

[a]SS group: performed a routine static stretching (SS) maneuver.

Table 2. Reliability assessment for DF ROM, passive torque at DF ROM, and muscle stiffness.

Data presented as mean ± standard deviation

DF, dorsiflexion; ROM, range-of-motion; ICC, intraclass correlation coefficient; CI, confidence interval

Table 3. Passive property changes of the gastrocnemius muscle–tendon unit over time.

* $p < 0.05$, ** $p < 0.01$; significantly different from the initial measurement.

SS, static stretching; DF, dorsiflexion; ROM, range-of-motion.

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	SS group[a] (N = 12)	control group (N = 12)	p-value
Age (years)	23.9 ± 3.0 (21–33)	23.6 ± 1.0 (22–26)	p = 0.23
Height (cm)	171.4 ± 4.4 (163–183)	172.7 ± 4.0 (163–180)	p = 0.89
Body mass (kg)	61.9 ± 5.1 (50–70)	64.3 ± 3.3 (57–70)	p = 0.33

	Test 1	Test 2	ICC (1, 1)	95% CI
DF ROM	34.9 ± 2.7	35.0 ± 2.6	0.836	0.464–0.960
Passive torque at DF ROM	40.7 ± 8.5	40.1± 8.2	0.942	0.782–0.986
Muscle stiffness	37.8 ± 6.7	38.3 ± 6.5	0.941	0.779–0.986

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	DF ROM (°)		Passive Torque at DF ROM (Nm)		Muscle Stiffness (Nm/cm)	
	SS group	Control group	SS group	Control group	SS group	Control group
Initial	34.8 ± 3.8	37.6 ± 5.6	39.3 ± 5.0	40.6 ± 10.5	38.7 ± 9.2	39.9 ± 8.9
1 week	37.3 ± 4.2	36.8 ± 5.1	48.0 ± 10.0	41.6 ± 9.8	37.1 ± 8.6	41.1 ± 6.8
2 weeks	39.6 ± 3.1*	37.4 ± 5.2	51.8 ± 10.5*	42.1 ± 10.8	37.2 ± 8.4	39.0 ± 8.0
3 weeks	40.7 ± 3.9**	37.3 ± 4.9	53.8 ± 10.9**	41.1 ± 7.2	30.4 ± 7.0*	41.0 ± 7.1
4 weeks	43.9 ± 4.5**	36.8 ± 4.7	58.3 ± 10.7**	41.9 ± 9.2	29.6 ± 6.8*	41.5 ± 7.5
Effect size	$\eta_p^2 = 0.483$		$\eta_p^2 = 0.211$		$\eta_p^2 = 0.334$	