Decrments in Stiffness are Restored within 10 min

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Abstract
The purpose of this study was to clarify the temporal course of stiffness in the muscle-tendon unit after stretching. In 11 male participants, displacement of the myotendinous junction on the gastrocnemius medialis muscle was measured ultrasonographically during the passive-dorsiflexion test, with the ankle was passively dorsiflexed at 1°/s to the end of the range of motion. Passive torque, representing resistance to stretch, was also measured using an isokinetic dynamometer. On 4 different days, passive-dorsiflexion tests were performed before and immediately, 5, 10 or 15 min after stretching, which comprised dorsiflexion to end range of motion and holding that position for 1 min, 5 times. As a result, end range of motion and passive torque at end range of motion were significantly increased after stretching \( P<0.05 \) as compared with each previous value. Although stiffness of the muscle-tendon unit was significantly decreased immediately and 5 min after stretching \( P<0.05 \), this change recovered within 10 min. These results suggest that static stretching for 5 min results in significantly increased range of motion over 30 min, but significant decreases in stiffness of the muscle-tendon unit returned to baseline levels within 5–10 min.

Introduction
Stretching before exercise is a common practice, and the effects reportedly include improved joint range of motion (ROM) and prevention of injury \[34,40\]. However, decreased muscle-tendon unit (MTU) stiffness as a result of static stretching may have major effects on subsequent performance. Various studies have demonstrated that static stretching decreases stiffness of the MTU \[21,22,25,27,35\], representing one factor in increasing joint ROM \[7,27,41,43\]. This increase in joint ROM is important in some dynamic sports in which enhanced static flexibility would be expected to affect performance, such as rhythmic gymnastics. However, the decrease in stiffness of the MTU that results from static stretching represents one factor decreasing maximum muscle strength \[36\]. This phenomenon is known as “stretching-induced force deficit”, and some reports have also described decreases in isometric muscle strength \[3,8\], concentric muscle strength \[5–7\], muscle endurance \[29\], sprinting speed \[42\], and vertical jump height \[15\]. Decreased stiffness of the MTU after static stretching thus appears to have both positive and negative effects on performance. The decrease in stiffness of the MTU resulting from static stretching may also reduce the risk of injury. No clear conclusions have yet been reached regarding the effects of static stretching on injury risk, with some reports supporting its effectiveness \[2,4\] and others expressing doubts in this regard \[31,37,43\]. One reason for this lack of unambiguous results may be differences in stretching time \[24\]. To reduce the risk of injury after stretching, sufficient decreases in stiffness of the MTU are important \[14,38\], and are believed to require four or five 60-s stretches \[19,20,22,25,28,35\]. Stretching times shorter than this are considered ineffective in preventing injury \[30,39\]. This means that at least 4 min of static stretching is required to lower the risk of injury by decreasing stiffness of the MTU in the target muscle. As stretching is generally performed 15–60 min before exercise \[44\], accurate knowledge of changes over time in the stiffness of the MTU as a result of stretching is important. Ryan et al. \[35\] reported that stiffness of the MTU decreased...
immediately after 2 min, 4 min, and 8 min of stretching, but that the effects of stretching disappeared within 10 min after 2 min of stretching, and within 20 min after 4 min and 8 min of stretching. Mizuno et al. [26] found that although maximal angle of dorsiflexion was increased after 5 min of ankle stretching and this effect was maintained for over 30 min, the decrease in stiffness of the MTU disappeared within 15 min, indicating that the increase in end ROM immediately after stretching is attributable to both decreased stiffness of the MTU and increased “stretch tolerance”, while the increase in end ROM at more than 15 min after stretching is largely attributable to increased “stretch tolerance”. To the best of our knowledge, however, few previous studies have systematically investigated changes in decreased stiffness of the MTU over time. Taking into account the fact that the effect of 5 min of static stretching disappears within 15 min [26], a focused investigation on the shorter period up to 15 min immediately after stretching would be highly advantageous to elucidate the retention time of the effect of stretching on stiffness of the MTU. The purpose of this study was thus to systematically clarify the time course for stiffness of the MTU. This work was conducted under the hypothesis that decrements in stiffness of the MTU would be retained over some interval, but would return to baseline within 15 min.

Materials and Methods

Participants

15 healthy men volunteered for the study, and the final cohort comprised 11 men (mean ± standard deviation (SD)) age, 23.3 ± 3.0 years; height, 172.2 ± 6.8 cm; weight, 64.4 ± 7.5 kg) who could be dorsiflexed > 15° in all passive dorsiflexion tests over the course of 4 test days, as stiffness values of the MTU were determined at 5°, 10° and 15° (Fig. 1b). The 4 volunteers who could not be dorsiflexed > 15° in ≥ 1 test were excluded from all analyses. All participants were recreationally active, but not involved in any structured physical training regime. No participants reported any history of recent musculoskeletal injuries or neuromuscular diseases specific to the lower limb. Written informed consent was obtained from all participants. The study protocols were approved by the Human Subjects Committee of Chukyo University Graduate School of Health and Sport Sciences, and complied with their requirements for human experimentation. The present study was performed in accordance with the ethical standards of the International Journal of Sports Medicine [13].

Experimental protocol

A randomized, repeated-measures, cross-over design (time [pre-stretching] × rest interval [immediately vs. 5 min versus 10 min vs. 15 min]) was used to clarify the time course of stiffness of the MTU. Participants visited the laboratory on 5 occasions, each separated by more than 24 h. Participants completed all experimental trials within 2 weeks. The first visit was a familiarization trial and the subsequent 4 visits were experimental trials. Before each set of measurements, participants were instructed to rest in a sitting position for 15 min in our laboratory. The first passive-dorsiflexion test was performed before stretching. During the passive-dorsiflexion test, we measured passive torque and displacement of the myotendinous junction (MTJ) at different joint angles, and end ROM of the ankle joint and electromyographic (EMG) activities of the medial and lateral heads of the gastrocnemius muscle. After the first passive-dorsiflexion test, static stretching was performed. Participants were then instructed to rest in a sitting position for various intervals before performing the second passive-dorsiflexion test. We designed 4 different rest intervals of 0 (immediately), 5, 10 and 15 min, with testing on 4 different days. The order of these sessions was randomized.

Passive-dorsiflexion test

To determine passive torque, displacement of the MTJ, end ROM and EMG, each participant underwent 1 passive-dorsiflexion test before stretching and after the rest interval (immediately, 5, 10 and 15 min post-stretching). The passive-dorsiflexion test was performed using an approach similar to that adopted by our previous study [26]. Participants were secured to the isokinetic machine (Biodex System3; Biodex, NY, USA) with the knee in full extension, and the footplate fixed to the right foot. The lateral malleolus was aligned with the axis of the dynamometer. In this study, all reported ankle angles were assessed as the angle of the footplate, and ankle angle was defined as 0° when the footplate was perpendicular to the floor. Values were defined as positive for dorsiflexion. Passive end ROM was determined using an approach similar to the method adopted by Morse et al. [27]. In this method, the foot of the participant was passively and isokinetically dorsiflexed at a speed of 1°/s from −30° to the angle at which the participant felt discomfort and stopped the dynamometer by activating a safety trigger. Maximal angle of the footplate was defined as the end ROM. In this process, passive torque generated on the footplate was also determined at ankle angles of submaximal (0°, 5°, 10° and 15°) and maximal dorsiflexion. Throughout the passive-dorsiflexion test, participants were requested to relax completely and not offer any voluntary resistance. Passive torque and ankle joint angle were converted from analog to digital at a sampling rate of 1.5 kHz (LX-10; TEAC, Tokyo, Japan).

To be consistent with our previous study [26], stiffness values of the MTU (Nm/°) were calculated using a second-order polynomial regression model that was fit to the passive torque-angle curves at the 4 points of 0°, 5°, 10° and 15°. Stiffness values of the MTU were calculated as the slope of the polynomial fit passive-torque-angle curves. Stiffness values of the MTU were determined at 5°, 10° and 15° (Fig. 1b).

B-Mode ultrasonography (LOGIQ P5; GE Healthcare, CT, USA) was used to determine displacement of the MTJ for gastrocnemius medialis during the passive-dorsiflexion test. The MTJ was visualized as a longitudinal ultrasonic image using a 4.5-cm, 12.0-MHz linear-array probe (12L probe; GE Healthcare), which was synchronized to the passive torque and joint angle outputs. The probe was secured to the skin using a specially made frame made of styrol. Displacement of the MTJ was measured as the value relative to a reflective marker placed between the skin and ultrasonic probe as a landmark. Ultrasonic images were recorded on videotape at 30 Hz (SR-VSI30; Victor, Kanagawa, Japan). Displacement of the MTJ was analyzed using software we developed. Displacement of the MTJ was manually traced using this software, which was made with Visual C++ (Microsoft, WA, USA) and DirectShow (Microsoft). In this study, the gastrocnemius muscle was elongated, and the MTJ was moved distally, because the ankle joint was dorsiflexed from −30° to end ROM (Fig. 1a).

The reliability of ultrasonic measurement with the ankle joint in a neutral position (0°) to 15° of dorsiflexion was evaluated in...
1 subject by measuring displacement of the MTJ. The coefficients of variation (SD/mean) was 4.7%.

**Static stretching**
Repeated static stretching was performed using the isokinetic dynamometer in the same fashion as the passive dorsiflexion test. Static stretching was administered to the right lower leg of each participant. The leg was secured on an isokinetic machine with the knee in full extension. The footplate attached to the isokinetic machine was fixed securely to the right foot of each participant and passively dorsiflexed at a constant velocity of 1°/s from 30° of plantar flexion to a position of maximal dorsiflexion angle that provoked a sensation in the triceps surae muscle similar to a static stretch manoeuvre without pain and the participant stopped the dynamometer by activating the safety trigger. This position was then held at a constant angle for 1 min. Thereafter, the footplate was returned to a position of 30° of plantar flexion. This stretching procedure was repeated 5 times. Maximal dorsiflexion angle was reassessed at each dorsiflexion. Throughout stretching, participants were requested to relax completely and not offer any voluntary resistance.
EMG
To ensure the passive-dorsiflexion test was truly passive, we measured EMG activities using bipolar, 13 mm Ag/AgCl surface electrodes (S&ME; Biolog, Tokyo, Japan) placed on the most prominent bulge of the gastrocnemius medialis and gastrocnemius lateralis muscles with a 25-mm interelectrode distance. EMG activity was recorded with a band width of 5–2000 Hz. EMG signals were transmitted to a digital data recorder at a sampling rate of 1.5 kHz. To remove any potential contribution of gastrocnemius medialis or gastrocnemius lateralis muscle contraction during dorsiflexion, we monitored EMG tracings < 50 μV above baseline during the passive stretch cycles of passive-dorsiflexion tests [9]. This level of EMG activity corresponds to approximately 2% maximal voluntary contraction [23] and ensures minimal activation of gastrocnemius medialis or gastrocnemius lateralis muscles during passive-dorsiflexion tests [11]. In this study, mean values for all test sessions in terms of root mean square EMG values of gastrocnemius medialis and gastrocnemius lateralis muscles during passive-dorsiflexion test were 7.3 ± 2.6 μV and 8.0 ± 2.2 μV for the initial 10° and 7.5 ± 3.1 μV and 9.3 ± 4.4 μV for the final 5°, respectively.

Statistics
All data are reported as mean ± SD. In this study, all measurement parameters were assumed to show normal distribution. Homogeneity of the variance assumption was assessed by the assumption of sphericity. Parameters that did not meet the assumption of sphericity were corrected for this violation using the Greenhouse-Geisser adjustment. A 3-way analysis of variance (ANOVA) (time [pre-stretch vs. post-stretch] × rest interval [immediately vs. 5 min versus 10 min vs. 15 min] × angle [0° vs. 10° vs. 15°]) was used to analyze submaximal passive torque and submaximal displacement of the MTJ. A 3-way ANOVA (time [pre-stretch vs. post-stretch] × rest interval [immediately vs. 5 min vs. 10 min vs. 15 min] × angle [5° vs. 10° vs. 15°]) was used to analyze stiffness values of the MTU. Two-way ANOVA (time [pre-stretch vs. post-stretch] × rest interval [immediately vs. 5 min vs. 10 min vs. 15 min]) was used to analyze end ROM and passive torque at end ROM. When appropriate, follow-up analyses were performed using lower-order ANOVA and t tests with Bonferroni corrections. The level of significance was set at P < 0.05.

Results

End ROM
No significant 2-way interaction (time × rest interval) and no main effect for rest interval were identified, but a significant main effect was seen for time. Stretching increased end ROM (P < 0.05) (Fig. 2a).

Passive torque at end ROM
No significant 2-way interaction (time × rest interval) and no main effect for rest interval were identified, but a significant main effect was seen for time. Stretching increased passive torque at end ROM (P < 0.05) (Fig. 2b).

Submaximal passive torque
No significant 3-way interaction (time × rest interval × angle) and no significant 2-way interactions for time × angle or rest interval × angle were seen, but a significant 2-way interaction for time × rest interval was identified. Post hoc analyses revealed that stretching decreased submaximal passive torque immediately after stretching (P < 0.05). However, no significant differences in submaximal passive torque were seen at 5, 10 or 15 min after stretching (Table 1).

Stiffness of the MTU
No significant 3-way interaction (time × rest interval × angle) and no significant 2-way interactions for time × angle or rest inter-

Table 1 Passive torque (Nm) at 0°, 5°, 10° and 15°, before and after stretching.

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<td></td>
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</table>

 Values represent mean ± SD. A significant interaction (time × rest interval) was seen *P < 0.05 compared to before stretching.

Fig. 2 Stretching-induced changes in end range of motion (ROM) a and passive torque at end ROM b. † A significant main effect was seen for time on passive torque at end ROM (P < 0.05). Data are expressed as mean ± standard deviation.
val × angle were seen, but a significant 2-way interaction for time × rest interval was identified. Post hoc analyses revealed that stretching decreased submaximal passive torque for immediately and 5 min after stretching (P < 0.05 each). However, no significant differences in submaximal passive torque were seen at 10 and 15 min after stretching (Fig. 3).

Displacement of the MTJ
A significant 3-way interaction (time × rest interval × angle) was noted. Post hoc testing revealed stretching increased displacement of the MTJ for immediately after stretching at 0°, 5°, 10°, and 15°, and for 5 min after stretching at 15° (P < 0.05 each). However, no significant differences in displacement of the MTJ were seen more than 10 min after stretching at any angle (Table 2).

Discussion

The objective of this study was to elucidate the effective time for which decreased stiffness of the MTU is maintained as a result of 1 min of static stretching at maximal dorsiflexion repeated 5 times. We found that although stiffness of the MTU decreased after static stretching, this effect disappeared within 10 min. In this study, all passive-dorsiflexion tests were performed passively, because EMG values of gastrocnemius medialis and gastrocnemius lateralis during passive-dorsiflexion test were < 50 μV above baseline.

Our finding in this study, that 1 min of static stretching at maximal dorsiflexion repeated 5 times increased end ROM of the ankle, was consistent with the results of previous studies [26, 27]. In the present study, end ROM increased significantly by 3.86 ± 5.1° immediately after static stretching compared with pre-stretching. Although the extent of the effect of static stretching on end ROM differed between individuals, this significant increase in end ROM was maintained 5 min (5.1 ± 6.0°), 10 min (2.6 ± 4.1°), and 15 min (3.4 ± 4.4°) after stretching. In addition, a previous study showed the retention time of the effect of 5 min-stretching on end ROM was > 30 min, with no effect of stretching retained by 60 min [26]. The increase in end ROM immediately after and 15 min after stretching was around the same level as that seen in our previous study [26]. The observation in the present study that the increase in end ROM was maintained 15 min after stretching is also supported by the findings of Rafford et al. [33], who reported that this effect is maintained for 5–30 min after static stretching.

In this study, the decrease in stiffness of the MTU disappeared within 10 min after stretching. The increase in passive torque at end ROM as a result of static stretching, however, was still maintained 15 min after stretching. End ROM increases after static stretching for 2 main reasons: 1) changes in mechanical characteristics of the MTU [20]; and 2) increased “stretch tolerance” [7, 41]. The increase in end ROM immediately after and 5 min after stretching seen in our study was probably due to both these factors, with the increase in end ROM 10 min and 15 min after stretching due to increased “stretch tolerance” alone. This result was supported by previous studies suggesting that acute and long-term increases in maximal attainable end ROM are related to subject tolerance to stretch, rather than passive properties of the muscle [20, 22]. The mechanisms underlying altered stretch tolerance remain unclear. However peripheral mechanisms such as afferent information from muscle, tendon and joint receptors may play a role and the potential involvement of central factors cannot be excluded [22].

The decrease in stiffness of the MTU observed in our study was probably mainly due to increased displacement of muscle. A number of tentative theories have been advanced to explain the mechanism underlying the decrease in stiffness of the MTU after static stretching, including increased tendon compliance [17], increased muscle fascicle length [8, 12], and changes in intramuscular connective tissue [10, 22, 27]. In the present study, displacement of muscle was increased significantly at 5°, 10°, and 15° dorsiflexion immediately after stretching, and at 15° dorsiflexion 5 min after stretching. MTU length is the sum of muscle length and tendon length. In this study, therefore, displacement of muscle was increased (i.e., increment of muscle length) and MTU length was constant at the same angle of dorsiflexion before and after stretching, indicating decreased displacement of tendon (i.e., decrement of tendon length) after stretching. When muscle stiffness immediately after and 5 min after stretching was calculated by the same method used in our previous study [26], a tendency toward a reduction in muscle stiffness was observed immediately after stretching (about 22% decrease, from 2.2 ± 1.0 to 1.7 ± 0.8 Nm/mm) and 5 min after stretching (about 30% decrease, from 2.4 ± 1.3 to 1.6 ± 0.6 Nm/mm). This finding was consistent with the results described by Morse et al. [27] that although displacement of muscle increased and muscle
stiffness decreased after stretching, no change in tendon stiffness occurred. This is inconsistent with the findings of Kubo et al. [17, 18], however, that tendon stiffness decreased after static stretching, and that changes in stiffness of the MTU are probably unrelated ($r=0.19$) to tendon stiffness [16]. Working on the hypothesis that stiffness of the MTU is not the only factor affecting passive torque, which is also influenced by other factors including ligaments, joint capsules, connective tissue, and skin, and that the contribution of stiffness of the MTU is constant before and after stretching, decreased muscle stiffness may possibly result in increased displacement of the muscle, with stiffness of the MTU decreasing as a result, causing a reduction in passive torque. As one reason for the decrease in muscle stiffness, Purslow et al. [32] reported the perimysium as the main extracellular contributor to passive stiffness. Gajdosik et al. [10] also suggested that lengthening deformation of the connective tissue within the muscle belly (endomysium, perimysium and epimysium) could influence passive stiffness. The results of our study, however, suggest that changes in displacement of muscle and connective tissue are only short-term.

Decreased stiffness of the MTU due to static stretching is regarded as one factor in the subsequent decrease in muscle strength [36]. Several previous studies have investigated the effect of static stretching on subsequent muscle strength and exercise performance, with no change after static stretching reported by some authors [1] and a decrease described by others [3, 8]. To the best of our knowledge, however, only a few reports have described improved exercise performance after static stretching. This reduction in maximum muscle strength after static stretching is known as a “stretching-induced force deficit,” and 2 tentative theories have been proposed with respect to the mechanisms involved [3, 5, 6, 8, 14]: 1) the neural factors, such as decreases in muscle activation; and 2) the mechanical factors, such as decreases in stiffness of the MTU. The reason the mechanical and contractive characteristics of the MTU cause a “stretching-induced force deficit” is reportedly because stretching increases the length of the resting sarcomere, thus affecting the muscle strength/length relationship and/or sarcomere-shortening velocity [5, 8, 14]. The results of our study, however, suggest that even if a decrease in muscle strength occurs after 5 min of static stretching, this effect is only maintained for ≤10 min. This is supported by the finding of Ryan et al. [36] that after 2 min, 4 min, or 8 min of static ankle joint stretching and 15 min at rest, maximum muscle strength decreased immediately after stretching in all experiments, but this effect disappeared within 10 min. In general, taking into consideration the fact that pre-exercise stretching is usually performed at least 10 min before the start of exercise, and that in practice the actual time spent stretching for a single site is shorter, the negative effects of static stretching on exercise performance are probably extremely small.

In conclusion, our results indicate that 1 min of static stretching at maximal dorsiflexion repeated 5 times can reduce stiffness in the MTU, but this effect disappears within 10 min. This suggests the possibility that although decreased stiffness of the MTU after pre-exercise static stretching is believed to be one factor that can negatively affect performance, the actual effects may be almost non-existent.

References