

The effects of 4 weeks stretching training to the point of pain on flexibility and muscle tendon unit properties

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Abstract

Purpose The purpose of this study was to compare the benefits and possible problems of 4 weeks stretching when taken to the point of pain (POP) and to the point of discomfort (POD).

Methods Twenty-six physically active women (20 ± 1.1 years) took part in group-based stretching classes of the hamstring muscles, 4 times per week for 4 weeks, one group one stretching to POD, the other to POP. Passive stiffness, joint range of motion (ROM), maximal isometric torque and concentric knee flexion torque, were measured before training and 2 days after the last training session.

Results Hip flexion ROM increased by 14.1° (10.1° – 18.1°) and 19.8° (15.1° – 24.5°) and sit-and-reach by 7.6 (5.2–10.0) cm and 7.5 (5.0–10.0) cm for POD and POP, respectively (Mean and 95% CI; $p < 0.001$ within group; NS between groups), with no evidence of damage in either group. Despite the large increases in flexibility there were no changes in either compliance or viscoelastic properties of the muscle tendon unit (MTU).

Conclusion Hamstrings stretching to POP increased flexibility and had no detrimental effects on muscle function but the benefits were no better than when stretching to

POD so there is no justification for recommending painful stretching. The improvements in flexibility over 4 weeks of stretching training appear to be largely due to changes in the perception of pain rather than physical properties of the MTU although less flexible individuals benefited more from the training and increased hamstring muscle length.

Keywords Constant torque stretching · Flexibility · Stretching intensity · Passive stiffness · Physical property

Abbreviations

CON	Concentric contraction
EMG	Electromyography
ISO	Isometric contraction
MTU	Muscle tendon unit
POD	Point of discomfort
POP	Point of pain
PRT	Peak resistive torque
ROM	Range of motion
S&R	Sit and reach
SLR	Straight leg raise
SR	Stress relaxation

Introduction

Most athletes spend a considerable amount of time stretching to improve flexibility and range of motion (ROM) in the belief that it will reduce the chances of injury and improve muscular performance (Magnusson and Renström 2006; Behm et al. 2016). Certain sports require exceptional flexibility and as an example (Moltubakk et al. 2016) showed major differences between elite female rhythmic gymnasts and age matched athletes in other disciplines such as football and cross-country skiing. While training

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studies might not be expected to produce the flexibility seen with gymnasts, significant improvements in flexibility have been reported in many studies stretching either the hamstring or calf muscle groups over several weeks (Wyon et al. 2013; Konrad and Tilp 2014; Blazevich et al. 2014; Freitas and Mil-Homens 2015). What is surprising, however, is that in these studies there has been little or no evidence of a change in either muscle tendon unit (MTU) stiffness or length of the muscle (Kubo et al. 2002; Mahieu et al. 2009; Konrad and Tilp 2014; Lima et al. 2015), the general conclusion being that the increase in ROM is due to an increased tolerance of stretch on the part of the subjects (Magnusson 1998; Blazevich et al. 2014).

As with many aspects of training there is a general belief that the more the better and in particular that stretching to the point of pain will yield better results than stretching to a more comfortable end point (point of discomfort, POD). It seems likely that highly motivated athletes will push themselves harder, possibly to the point of pain (POP), and for longer than is generally the case in training studies. Similar supporting studies of Schleip et al. (2012) and Freitas et al. (2015, 2016) also found that intense stretching could acutely change connective tissue biomechanical properties. Although relatively long training studies, the subjects in Magnusson (1998) study stretched until they felt a “feeling of stretch” while in the study of Blazevich et al. (2014) the subjects were to stretch “within the limit of pain”. Although we saw no benefits in an acute stretching to POP study (Muanjai et al. 2017) it is possible that a longer training study in which subjects stretched to the point of pain might show at least the first signs of adaptations leading to the extreme flexibility seen in some gymnasts. Moltubakk et al. (2016) reported that flexible athletes had longer hamstring muscle length based on the angle at which maximum torque was generated and it is possible that a longer and vigorous training study might show similar changes.

There may, however, be problems with stretching to POP since it could lead to damage the tendons, connective tissue or to the muscle itself and, in this respect, Moore (1984) and Kokkonen et al. (1998) found an acute decrement of force after static stretching to the pain threshold. There may, therefore, be both benefits and risks associated with this practice. We have previously compared the acute effects of stretching to POP with stretching just to POD (Muanjai et al. 2017) and found little or no benefit from stretching to the greater lengths either immediately after the exercise or 24 h later. On the other hand there were no adverse effects in terms of loss of muscle strength or signs of inflammation. However, it is possible that there were small effects, beneficial or otherwise, beneath the sensitivity of our measurements, which over the course of an extended training period may accumulate resulting in significant improvements or problems associated with the

more aggressive stretching procedure. Consequently the purpose of the present study was to compare the effects of a four week training programme in which one group stretched to POP while the other stretched to POD, monitoring changes in the range of motion, and recording any evidence of muscle damage. A second objective was to determine the extent to which changes in the physical properties of MTU may contribute to improvements in flexibility over a long term training programme. It was hypothesised that while stretching to POD and POP would both result in increases in ROM, stretching to POP would result in greater changes and show evidence of increased muscle length in terms of the knee angle at which maximum torque was generated.

Methods

Participants

Twenty-six women were recruited from staff and students at the University. Exclusion criteria were an involvement in resistance or flexibility exercise in the previous 6 months, neuromuscular or skeletal problems associated with lower extremities or lower back and the regular use of analgesic or anti-inflammatory drugs. Subjects were all physically active, taking part in dance or aerobics but none were gymnasts. After the study had been carefully explained the subjects provided written informed consent to the study that had been approved by the Kaunas Regional Biomedical Research Ethics Committee.

Study design

The participants were randomly allocated either to train by stretching to POD or to POP. The physical characteristics of the subjects in the two groups are given in Table 1. Measurements of range of motion (ROM), passive stiffness, stress relaxation (SR), maximal voluntary isometric knee

Table 1 Physical characteristics of the participants

Characteristics	POD group <i>n</i> = 13	POP group <i>n</i> = 13
Age (years)	20 ± 1.2	20 ± 1.1
Weight (kg)	61.4 ± 5.7	63.5 ± 13.6
Height (cm)	168.9 ± 4.0	167.6 ± 5.4
BMI (kg m ⁻²)	21.5 ± 1.9	22.6 ± 4.7
% Body fat	24.6 ± 5.4	25.4 ± 8.9

Data are mean and SD

BMI body mass index, *POD* discomfort group, *POP* pain group

flexion torque (ISO) and concentric isokinetic knee flexion torque (CON) and hamstrings muscle tenderness were made before training began and within 2 days after the last training session, as shown in Fig. 1. A few days before the first testing session, participants took part in a familiarization session where they were introduced to all the measurement procedures. Before each testing session, the subjects warmed up for 8 min on a cycle ergometer at 50 W. All measurements were made on the participants' right leg by the same investigators who were not involved in the training and were not aware of the group to which the subjects belonged. The temperature of the testing and training room was kept at 22 °C throughout the study.

Stretching training

To encourage participation and retention the group-based training sessions were designed to be enjoyable and were

led by a professional instructor. Before each session the participants undertook 10 min of aerobic and callisthenic exercises, mainly of the lower body, after which they performed active stretching of both hamstrings using the three exercises shown in Fig. 2, together with touching toes in the standing position. Stretching continued to the point of maximum tolerable stretch for POD group or a point of pain onset for POP group using the scale of perceived effort in flexibility (PERFLEX) (Magnusson 1998; Blazevich et al. 2014) as a guide. Once the end point had been reached the position was held constant for approximately 10 s while the sensation decreased after which the knee flexors were stretched further to restore the discomfort or pain sensation, several times over a period of 30 s. This procedure was repeated for 8 sets with 15 s rest between sets, 4 times per week for 4 weeks. The two groups were trained separately with the POP group being especially encouraged to fight the pain.

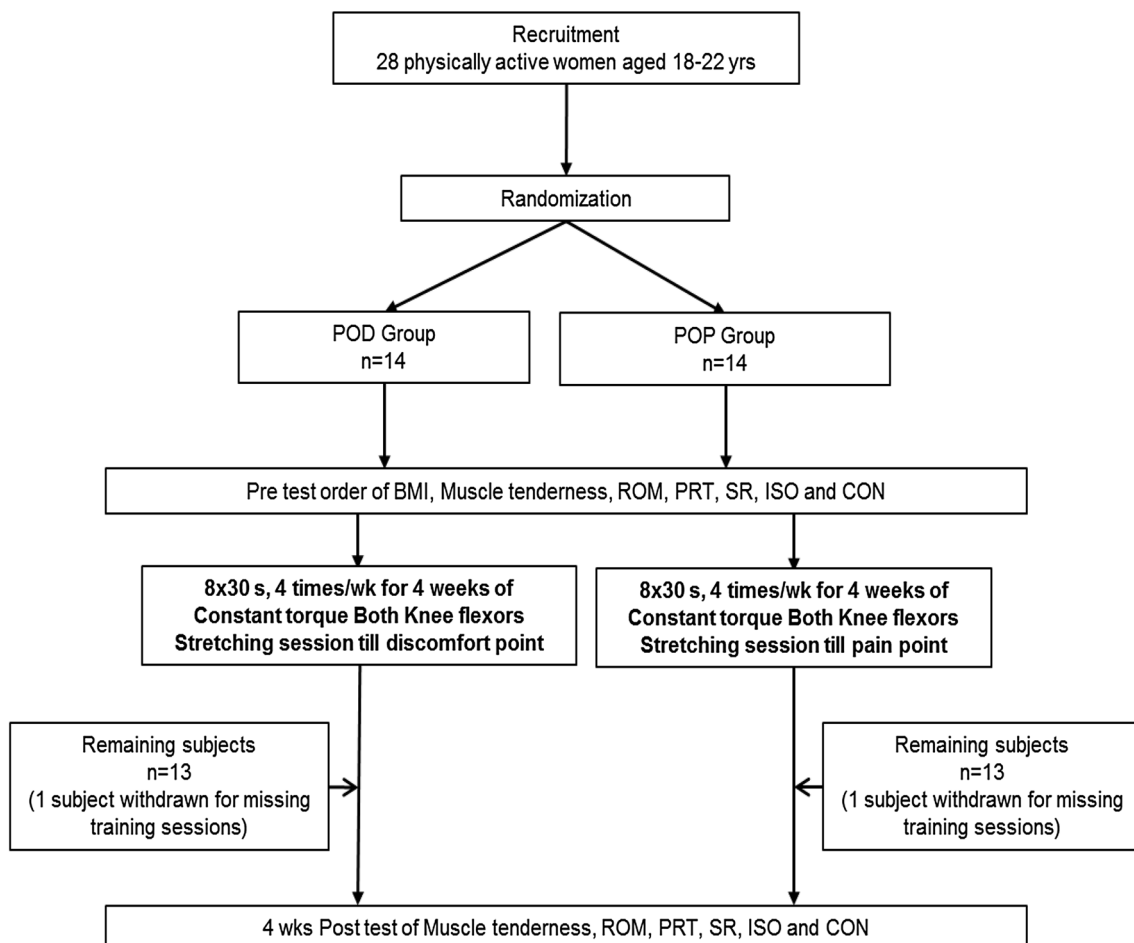


Fig. 1 Study design and flow of subjects. *BMI* body mass index, *ROM* range of motion, *PRT* peak resistive torque, *SR* stress relaxation, *ISO* isometric maximum voluntary contraction, *CON* concentric isokinetic knee flexion torque



Fig. 2 Group-based stretching training. In each training session subjects actively stretched both hamstrings in various positions which included, from left to right, lying bend knee and single leg raise, sit-

ting reach-forward, kneeling toe-up, together with standing reach-downward (touching toes). Each procedure was repeated for 8 sets of 30 s with 15 s rest between sets, 4 times per week for 4 weeks

Flexibility

Flexibility of knee flexors muscle was evaluated by the sit and reach (S&R) method and passive straight leg raise (SLR) test. With the sit and reach box, participants moved their trunk forward as far as possible while the knee was kept straight, holding this position for approximately 2 s (Kamandulis et al. 2013). Best of two trials was recorded. For the passive SLR, participants lay supine on a couch and were told to relax their right leg. The hip flexion angle was determined with a universal goniometer between the lateral epicondyle of the femur and torso with the projection of the greater trochanter of the femur as the axis (Castellote-Caballero et al. 2013). One investigator gradually lifted the participant's leg until maximum resistance was felt, but with no compensatory movement (i.e. knee bending or pelvic tilt), at this point a second investigator placed the movable arm of a goniometer along the alignment line and the hip flexion angle was noted for subsequent analysis.

Maximum knee extension angle, passive resistive torque and passive stiffness

The participants were seated on a Biodex System 3 isokinetic dynamometer (Biodex Medical Systems, Inc., Shiley, New York, USA) with approximately 120° hip flexion and the shank 50° below the horizontal (Muanjai et al. 2017). The pelvis and both thighs were tightened with Velcro straps. The axis of rotation was fixed at the right knee joint and the lever arm pad was attached proximal to the malleolus. EMG activity was recorded from the knee flexor muscles. In this position, the dynamometer passively extended the knee at 5 degree s⁻¹ to the end point of maximum discomfort but without pain. Passive stiffness was determined from the slope of the passive angle-torque relationship, with gravity correction for the weight of the lower leg, as the increase in passive torque between 50 and 80% of maximum ROM using a least-squares method (Cabido

et al. 2014). After 4 weeks of training, stiffness was measured again over the same angles used for the pre-training measurements. Measurements were only accepted if knee flexors muscle EMG activity was less than 5% of EMG max (Whatman et al. 2006). ROM and peak resistive torque (PRT) were measured before and after stretching training as the values where maximal discomfort without pain was reported. Three passive maximum knee extension ROM, peak torque and stiffness measures were averaged for further analysis.

After 1 min rest the procedure was repeated twice more with the knee passively extended at 5 degree s⁻¹ on one occasion to the onset of discomfort and the other to the onset of pain. These positions were held for 30 s and the decline in torque was recorded (Fig. 3). The decline in torque (stress relaxation) was analysed as the difference between peak torque immediately after the end of the stretch and after 30 s while the limb was held in the extended position (Matsuo et al. 2013). The decline in torque was not linear so the SR was further analysed assuming the MTU to behave as a Maxwell spring and dashpot (Vincent 1982; Tian et al. 2010; Meyer et al. 2011) by fitting the SR data to the equation.

$$\text{Torque} = T_0 \cdot e^{-kt} + C$$

where k is the rate constant for decay of torque, t is time, C the torque at infinite time and T_0 the difference between the peak torque at the end of the stretch and the value C . The approximate values of T_0 and C are indicated in Fig. 3a. Data were fitted using a least squares routine running in Excel.

Isometric and concentric voluntary torque

The participants' hip and knee flexion were fixed at 85° and 90° (full extended knee = 0°), respectively, for the measurement of ISO torque (Figure 1 of Matsuo et al. 2015).

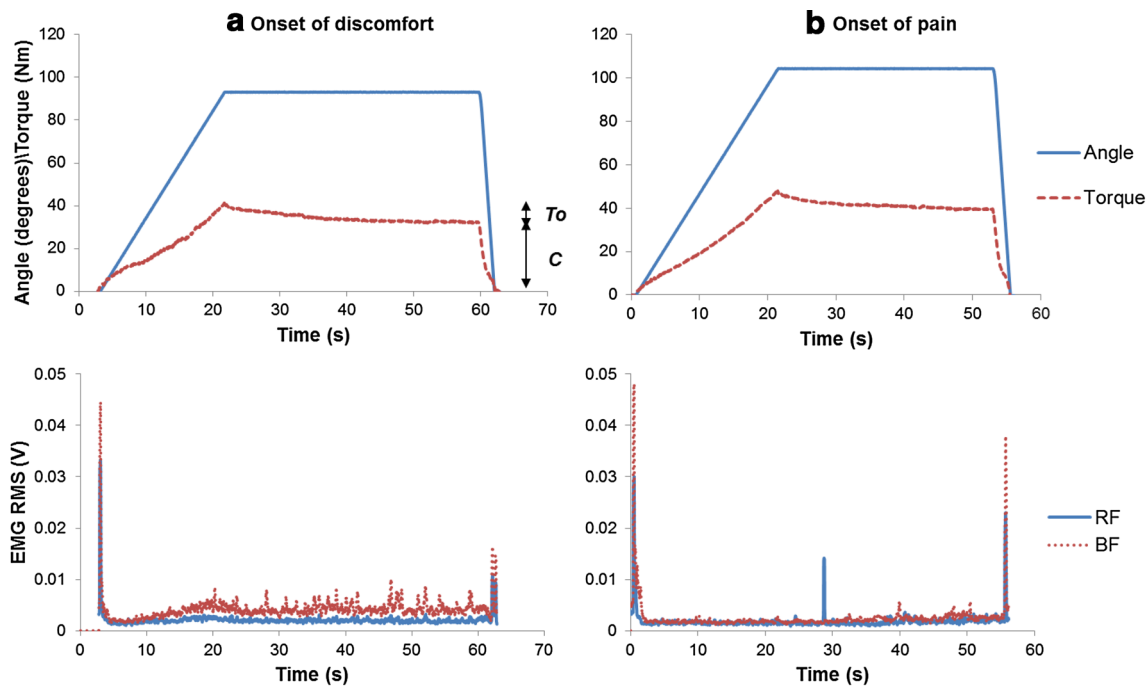


Fig. 3 An example data acquisition of stress relaxation testing at onset of discomfort (a) and pain point (b) in the same person. The approximate values of T_o (the torque decline) and C (the torque at

infinite time) are indicated in (a). The maximum EMG activity was 0.637 V. *EMG* electromyography, *RMS* root mean square, *RF* rectus femoris, *BF* biceps femoris

The trunk, pelvis and thigh were firmly secured with straps and the lever arm tightly fixed 2 cm above the medial malleolus. The participants were verbally encouraged to perform two maximum isometric knee flexion contractions for 3 s with 60 s rest between trials and the highest torque obtained was used for analysis. After the isometric strength test, participants were instructed to execute two maximum voluntary concentric knee flexions from 20° to 110° extended knee during which the lever arm moved at an angular velocity 60 of degree s^{-1} , with verbal encouragement throughout the testing procedure. Peak concentric torque was recorded together with the angle/torque relationship.

Hamstrings muscle tenderness

Muscle tenderness was tested as a pressure pain threshold; the medial and lateral hamstrings were marked at 70, 50, and 30% (representing the upper, middle and musculotendinous portions, respectively) of the distance between the ischial tuberosity and the medial and lateral epicondyle of the tibia. The positions were marked on a transparent sheet together with anatomical markers to enable the same point to be identified 4 weeks later. An algometer (Wagner Instruments, Greenwich, USA) with a 1 cm diameter probe was used to slowly apply increasing pressure perpendicular to the skin at the specific locations working from proximal to

distal of the lateral hamstring first and then distal to proximal of the medial hamstring to avoiding causing hypersensitivity by the repeated testing. The tests were performed with the subjects prone, lying with their feet over the end of the treatment bed. The participants reported the end point when the pressure turned into a painful sensation (Rocha et al. 2012). The applied force at the end point of the three trials at each position was recorded and the average of all 6 point locations used for further analysis. Forces greater than 30 N were taken to indicate no tenderness (Jones et al. 1987).

Sample size and statistical analysis

Change in stiffness during passive extension of the knee flexors as a result of training was considered the critical variable in this study. Duplicate measurements made on 25 subjects showed the measure to be reproducible with a coefficient of variation of 20%. Power calculations indicate that a sample size of 10 or more subjects would be sufficient to detect changes of 10% in paired observations.

Descriptive data are presented as mean \pm SD, and tested for normality by a Shapiro–Wilk test. All data were normally distributed with the exception of maximum knee extension ROM and passive stiffness. Where normally distributed differences are given as mean and 95% confidence

interval. Time and interaction effects were determined by two-way repeated measure ANOVA (time (pre, 4 weeks) × intervention (discomfort, pain)) and where significant interactions were present post hoc multiple comparisons were made, with Bonferroni correction. Where not normally distributed, the differences between times and interventions were examined using the Kruskal–Wallis Test for non-parametric data. An alpha level of 0.05 was used to determine statistical significance. All statistical analysis was performed using Statistic Package for the Social Sciences (SPSS for Window version 22.0, Chicago, IL, USA). A linear least squares linear regression was used to determine the relationship between initial flexibility and change in ROM.

Results

Compliance with training was good. Two subjects, one in each group were withdrawn because they missed eight sessions of training but otherwise subjects completed 100% of the sessions with no adverse reactions to the training.

Range of motion

There were substantial improvements in flexibility as a result of the training (Fig. 4). Hip flexion ROM assessed by straight leg raise increased by 14.1° (10.1°–18.1°) for the POD group with a tendency for a somewhat larger change for the POP group of 19.8° (15.1°–24.5°) (Mean and 95% CI; $p < 0.001$ within group; $p = 0.07$ between groups). Reach, in the sit and reach test, increased by 7.6 cm (5.2–10.0 cm) and 7.5 cm (5.0–10.0 cm) for the POD and POP groups, respectively (Mean and 95% CI; $p < 0.001$ within group; $p > 0.05$ between groups).

Passive muscle properties

After 4 weeks of hamstrings stretching both the ROM and PRT increased as determined by passive knee extension with the isokinetic dynamometer (Fig. 5). The maximum knee extension ROM increased by 15.3° (9.5°–21.0°) for the POD and 16.4° (11.4°–21.4°) in the POP [Mean (95% CI); $p < 0.001$ within groups; $p > 0.05$ between groups]. The maximum PRT also increased by 14.0 Nm (10.3–17.7 Nm) for the POD and by 15.5 Nm (8.7–22.4 Nm)

Fig. 4 Effects of 4 weeks training on flexibility. (Left) Hip flexion in the straight leg raise (SLR). (Right) Reach distance in the sit and reach test. Blank columns are POD; shaded columns, POP. Data are mean and SD. Asterisk statistical difference from the pre-test measure ($p < 0.05$)

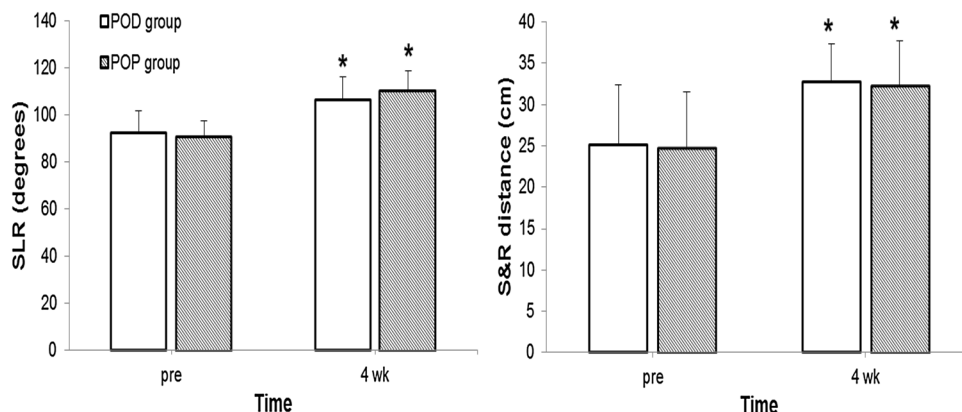


Fig. 5 Torque during passive extension of knee flexors. Circles are the POD group, Squares the POP group; dashed lines and open symbols before training; solid line and filled symbols after 4 weeks stretching training. Data are mean and, where shown, SD. No time or group effect was significantly found in PRT and passive stiffness. Asterisk statistical maximum ROM difference from the pre-test measure ($p < 0.05$)

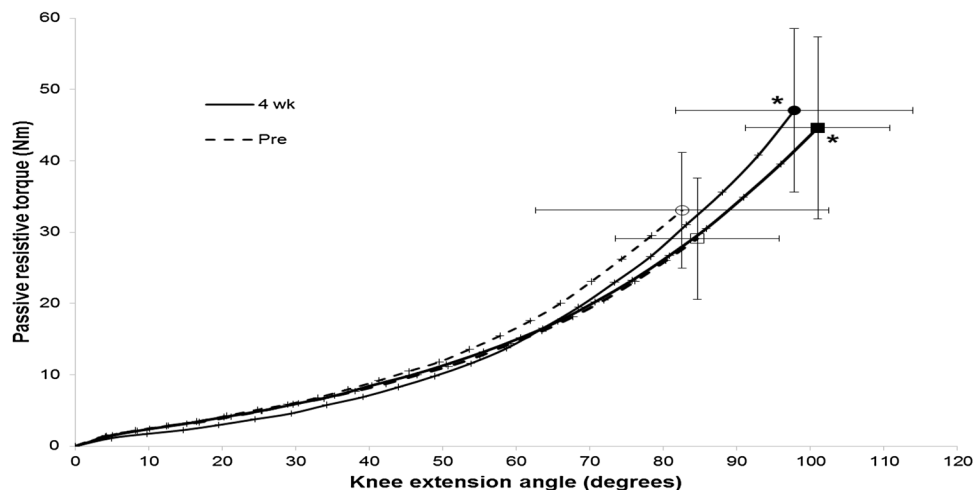
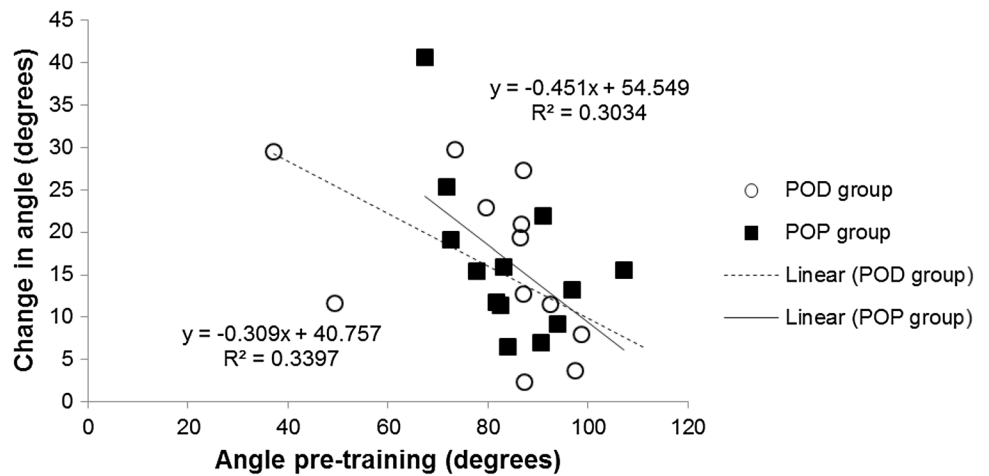


Fig. 6 Changes in maximum knee angle as a result of training in relation to the maximum knee angle pre-training. *Solid line and symbols, POP group; dashed line and open symbols, POD group*



for the POP (Mean (95% CI); $p < 0.001$ within groups; $p > 0.05$ between groups). The increase in ROM after training was related to the initial flexibility of the subjects, those with the smaller angles pre-training had, on average larger improvements than those who were more flexible (Fig. 6; $R^2 = 0.34$, $p = 0.04$ and $R^2 = 0.30$, $p = 0.05$, for POD and POP, respectively). There was no difference between groups in this respect.

Although there was some variation between subjects, it is evident from the data in Fig. 5 that the shape of the angle torque relationship did not change as the result of training. Stiffness before training was 0.47 ± 0.2 and 0.44 ± 0.2 Nm degree⁻¹ after training in the POD. For the POP the stiffness before training was 0.36 ± 0.1 and 0.38 ± 0.1 Nm degree⁻¹ after training. In neither case was the difference significant. There was a tendency for the POD to be slightly stiffer than the POP before training ($p = 0.06$) which was primarily due to two potential outliers (the left most subjects in Fig. 6), although there was no obvious justification for removing them from the analysis.

Stress relaxation

Stress relaxation was measured as the decrease in torque over 30 s after the knee was extended to the onset ROM. There were two measurements for each subject one where the ROM was defined as the onset of discomfort and then a second time when the ROM was taken as the onset of pain. This was repeated for both groups before and after 4 weeks of training. There was greater SR, measured as the difference between peak torque and torque after 30 s rest, after 4 weeks of stretching training and this was true both when measured from POD or POP (Fig. 7). It is notable, however, that SR was larger at the greater angles both before and after training. Analysis of SR in terms of an exponential decay of torque showed similar results with the values of T_0 and C being larger after training and greater at the larger angles (Fig. 8). The average rate constant for torque decay was 0.123 s⁻¹, giving a half time of just over 5 s which did not differ between groups, before and after training, or with the angle.

Fig. 7 Stress relaxation. SR measured before (*dotted columns*) and after (*blank columns*) 4 weeks of stretching training. There are two sets of measurements for each group; on the *left*, where the knee was extended to the point of discomfort and, on the *right*, where it was taken to the point of pain. The angle at which the measurements were made is indicated under each column. Data are mean and SD. Asterisk statistical difference from the pre-test measure ($p < 0.05$)

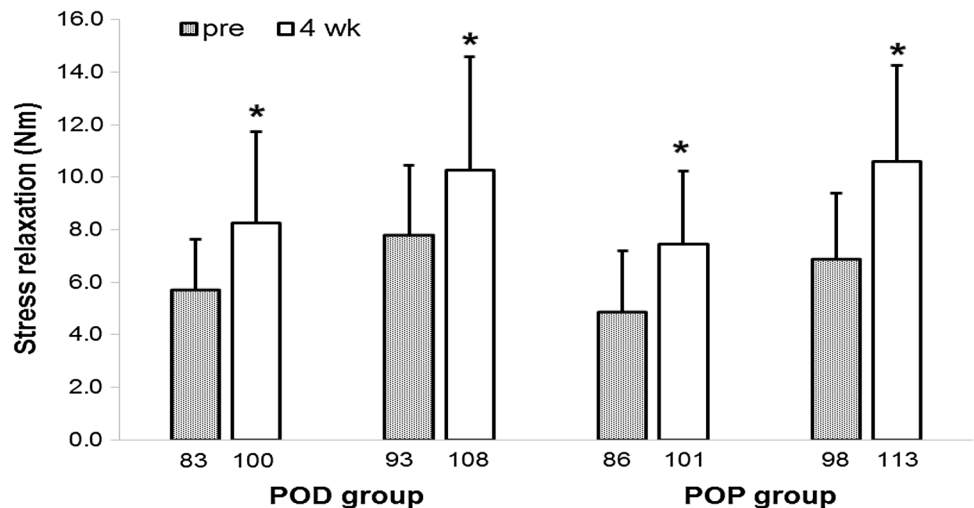


Fig. 8 The relationship between knee angle torque and stress relaxation. Circles peak torque, squares C, triangles To. Open symbols before training, closed symbols after 4 weeks stretch training. Data for the POD group. Data are mean and SD

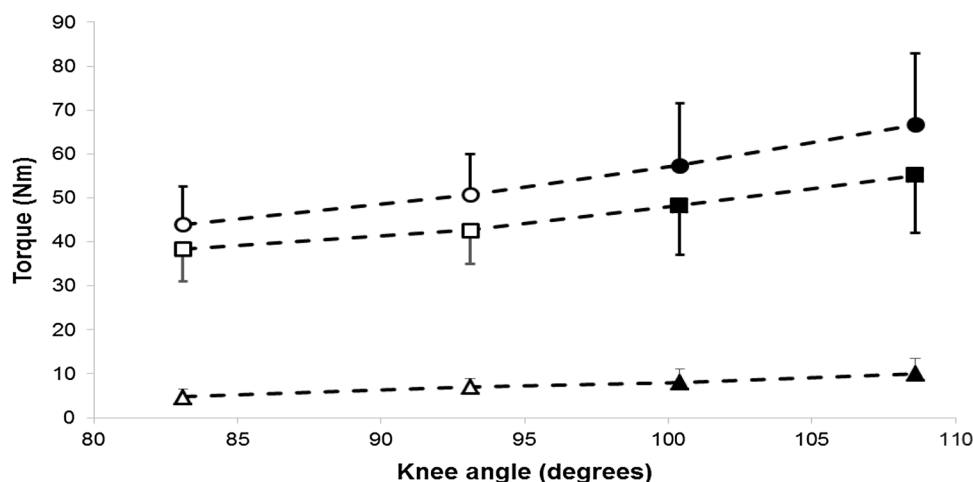


Table 2 Data for muscle strength before and after 4 weeks stretching training

		ISO (Nm)		CON (Nm)	
		Pre	4 week	Pre	4 week
POD group	Torque: mean	84.1	83.7	75.6	78.9
	SD	14.7	14.4	8.9	11.8
POP group	Torque: mean	67.7	70.4	68.3	69.0
	SD	13.5	13.4	14.1	13.6

Data are Isometric torque (ISO) and peak Concentric torque (CON)

Isometric and concentric strength

Data for muscle strength are given in Table 2. There were no significant time, interaction or group effects after 4 weeks training for either isometric or peak concentric strength ($p > 0.05$). The CON torque as a function of knee extension angle is shown in Fig. 9, expressed relative to the torque generated at 0° extension. There were no significant differences in the torque before and after training in either the POD (Fig. 9a) or POP (Fig. 9b) groups.

Muscle tenderness

At no time, and in neither group, were the knee flexor muscles tender.

Differences between subjects in flexibility

During the study it was evident that some subjects were very much more flexible than others and the data in Fig. 6 indicates that the less flexible subjects had larger training responses than those who were more flexible and it was of interest to see if these differences were reflected in the characteristics of the MTU. Nine subjects who had a ROM during passive knee extension of 90° or more (91°–111°)

before training were matched with nine subjects who had the lowest ROM (37°–82°), coming equally from the POD and POP groups.

Before training there were clear differences between the most flexible and least flexible groups in the relationship between the angle of knee extension and CON torque (compare dashed lines in Fig. 9c, d). Relative to the torque generated at 0° extension (lower leg vertical) the most flexible group generated more torque at longer hamstring lengths. After 4 weeks training the least flexible group showed a significant increase in the relative torque developed at the longer muscle lengths (compare dashed and solid lines in Fig. 9c) while there was no change in the relationship after training in the most flexible group (Fig. 9d). After training the relationships were virtually the same in the two groups (compare solid lines in Fig. 9c, d).

Passive stiffness as a function of knee extension also differed between the groups (Fig. 10a) with stiffness measured between 50 and 80% of pre-ROM max being $0.51 \pm 0.18 \text{ Nm degree}^{-1}$ for the least flexible group, and $0.35 \pm 0.10 \text{ Nm degree}^{-1}$ for the most flexible group. With training there was a small and non-significant decrease in stiffness of the least flexible group (Fig. 10b) with no change with the most flexible group (Fig. 10c). As can be inferred from Fig. 6, the least flexible group increased their ROM as a result of training more than the flexible group; by 23° compared with 10°.

Discussion

There is considerable variation in flexibility between people and some groups, such as gymnasts and dancers, demonstrate dramatic differences compared with equally fit and young subjects. Moltubakk et al. (2016) have shown that in one such flexible group the hamstring muscles appear to be lengthened so that torque was generated over a wider

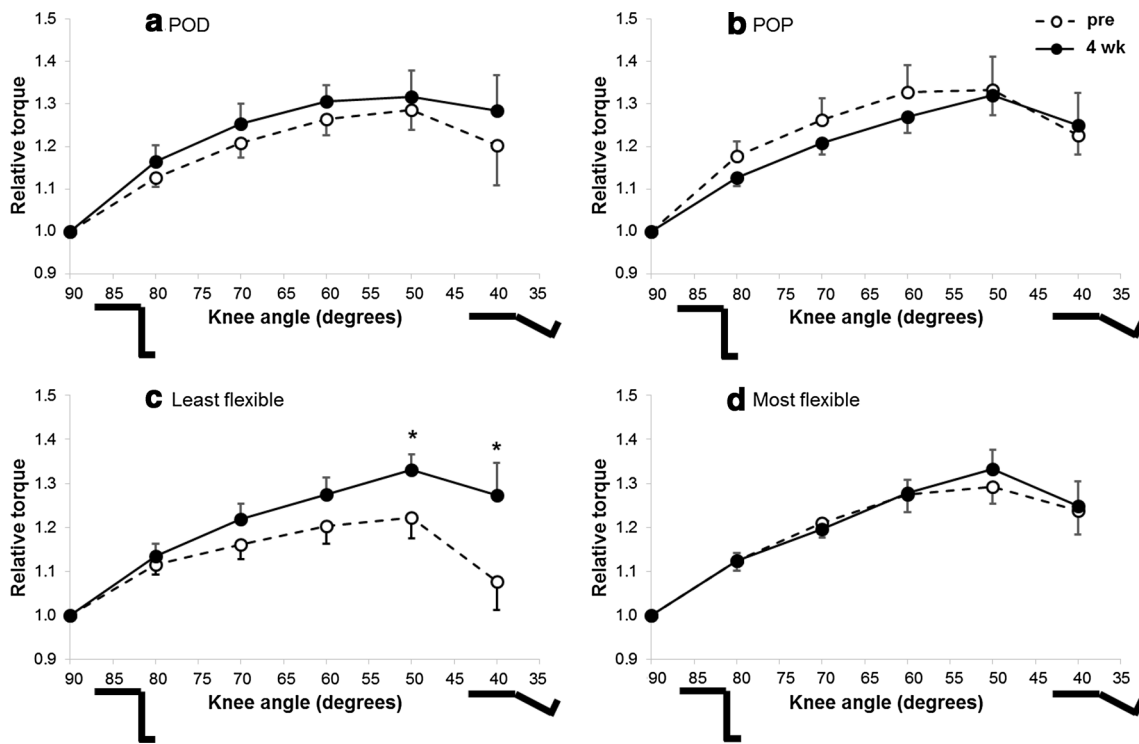


Fig. 9 Angle torque relationships during CON of the knee flexors. **a** POD group. **b** POP group. **c** Least flexible subjects. **d** Most flexible subjects. *Open symbols and dashed lines* before training, *solid sym-*

bols and lines after 4 weeks stretch training. *Asterisk* statistical difference from the pre-test measure ($p < 0.05$). Data are mean and SD

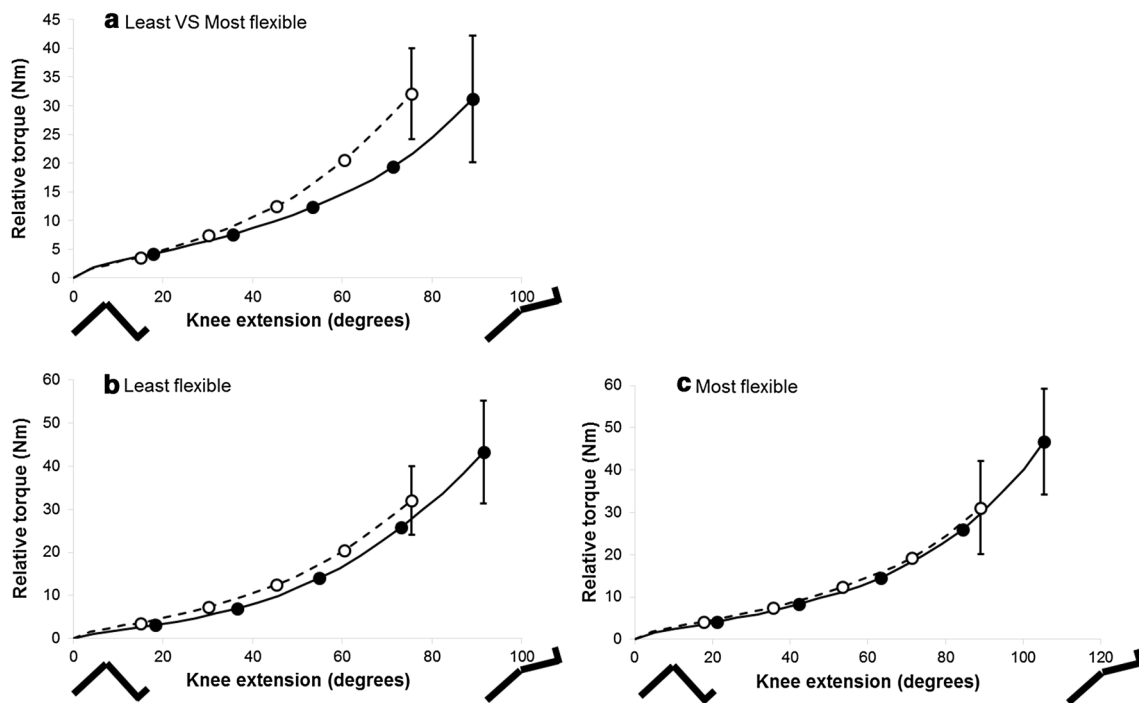


Fig. 10 Passive knee extension. **a** Comparison of the least flexible subjects (*dashed line*) with the most flexible (*solid line*) before training. **b** Least flexible subjects before (*dashed line*) and after 4 weeks

stretch training (*solid line*). **c** Most flexible subjects before (*dashed line*) and after 4 weeks stretch training (*solid line*). Data are means with sample SD shown for the last point

range of knee flexion/extension and peak torque obtained at longer MTU lengths compared with less flexible individuals. The physiological differences and the mechanisms by which such differences in flexibility are established are of considerable interest but the majority of training studies of stretching have not found changes in the physical properties of the MTU, rather they conclude that the improvements in flexibility come about as a result of an increased tolerance to stretch (Magnusson 1998; Blazevich et al. 2014). One possibility is that previous training studies did not stretch far enough, not taking the subjects to POP, and the purpose of the present study was to determine the possible benefits of this more rigorous training, and also to look at possible problems such as muscle damage. The 4 weeks training programme resulted in substantial improvements in flexibility, with the straight leg raise increasing by around 20° while the sit and reach increased by about 7 cm with no differences between the groups stretching either to POD or POP (Fig. 4). These benefits compare with the improvements of 4° for the straight leg raise previously seen 24 h after a single bout of stretching in these same subjects (Muanjai et al. 2017). There was considerable variation in the improvements seen and this was related, to some extent, to the pre-training flexibility (Fig. 6) and was similar for both training groups. There is, therefore, clear evidence that prolonged training results in increased flexibility, as has been widely documented (Folpp et al. 2006; Ben and Harvey 2010; Blazevich et al. 2014; Kay et al. 2016) but no evidence that taking the stretching exercise to POP improves the outcome, although Blazevich et al. (2014) did find the larger stretching to result in greater gains in flexibility.

There was no evidence of muscle damage as a result of the 4 weeks training. None of the subjects showed any evidence or complained of pain or muscle tenderness that is characteristic of the delayed onset muscle soreness associated with stretching active muscle. Possibly the best indicator of muscle damage is loss of force (Warren et al. 1999) and at 4 weeks there was no decrease in either isometric or concentric torque, although others have reported decreases (LaRoche et al. 2008; Kokkonen et al. 2010). It is possible that there may have been some loss of force in the time before the end of the study but we previously reported no significant force loss 24 h after a single training session. Likewise it is possible that there may have been some pain or tenderness in the time before the end of training, but none of the subjects commented on this. There may have been some damage to tendons or ligaments, but this would be expected to result in a degree of tenderness or oedema and none was noticed or commented on during the training. It is clear that while stretching to the point of pain may provide some psychological help for athletes, physiologically it is of no greater benefit than stretching to a more

comfortable limit. On the other hand there is no evidence, in terms of muscle pain or loss of force, that it does any harm.

An additional 20° of hip flexion and 7 cm in the sit and reach test represent very considerable gains in flexibility and might suggest an effective increase in compliance of the MTU, either because of an overall increase in length or compliance of one or more components of the unit. To examine this we determined the passive stiffness of the hamstring muscle group before and after training and found that while the maximum ROM increased after training by 15° and 16° for the POD and POP groups, respectively, the maximum passive torque also increased in proportion so there was no change in the overall angle/torque relationship (Fig. 5) or in the measured stiffness over a specified range of angles. Similar findings have been reported by Magnusson (1998), Folpp et al. (2006), Ylinen et al. (2009), Ben and Harvey (2010), and Weppeler and Magnusson (2010) although these studies did not take their subjects the point of pain. While acute effects of stretching on MTU compliance have been reported (Morse et al. 2008) the changes may be relatively short lived since Mizuno et al. (2013) found that MTU stiffness recovered within 15 min after static stretching.

Muscle demonstrates a characteristic hysteresis when stretched implying the presence of a viscoelastic component, seen here as stress relaxation, and we show that this decays away with a half time of approximately 5 s. The magnitude of the viscoelastic component can be judged from the value of T_0 , derived from the exponential analysis of SR. This value is very similar to that obtained by simply comparing the difference between peak torque and the value 30 s later (Fig. 7) since 30 s represents about 6 half-lives. The rate constant or half time for torque decay during SR was found to be very constant, not differing between pre- and post-training or with the angle to which the knee was extended. This suggests that the physical nature of the viscoelastic component of the MTU did not change as a result of the training. However, the amplitude of T_0 and the decline in torque between the peak and 30 s later (Fig. 8) were greater following 4 weeks training. This could be as a result of an increase in the quantity of the viscoelastic component but this is unlikely and the important point to note is that SR was determined at a longer MTU length after training. Any viscoelastic structure will show greater SR when stretched to a longer length and this is what appears to happen here and is consistent with the work of Freitas et al. (2015) who found SR to be greater when the ankle plantarflexors were stretched to 80° compared to 40°. Consequently it appears that, on average, stretching training, either to POD or POP, has little effect on the quality or quantity of the MTU and the major reason for improvements in flexibility is a change in the subjects' tolerance

to stretch. In our previous study of the acute effects of stretching (Muanjai et al. 2017) we commented on the fact that although only one leg was stretched there was, nevertheless, an improvement in the sit and reach test which involves both legs, suggesting that the change is a greater tolerance of pain, rather than any change in the physical properties of the MTU. A similar explanation seems to be required to explain the present results as has also been suggested (Folpp et al. 2006; Weppler and Magnusson 2010; Blazeovich et al. 2014). It is possible that the stretching training increased blood flow to the MTU reducing pain sensation since Mense and Stahnke (1983) noted that the C-fibre activity is inversely dependent on the muscle blood flow.

Differences between subjects in flexibility

There was considerable variation in the flexibility with the ROM of subjects in the POD group ranging from 37° to 111° during the passive knee extension test although the range was somewhat less in the POP group (67°–107°), with no obvious explanations for this variation in terms of ethnic origin, lifestyle or sporting background. There was a tendency for the least flexible subjects to improve the most (Fig. 6) and we have taken the opportunity to explore possible reasons for this difference. Dividing the subjects into the most and least flexible, essentially the upper and lower tertiles, has demonstrated differences both in the untrained state and in the responses to training.

The least flexible group had stiffer MTUs (Fig. 10a) and while the torque developed at the end of the passive knee extension was similar in the two groups both before and after training, the final angle was approximately 10° larger for the flexible group. If the sensation of stretch is related to the passive torque generated in the MTU, as seems likely, then these results suggest that difference in perception of discomfort or pain are not the explanations for the differences in flexibility. In support of this is the observation that the difference between POD and POP was about 10° in both groups. If the less flexible group was more sensitive to the sensations of stretch then the difference between POD and POP might be rather less than for the most flexible group. The data concerning the length/torque relationship (Fig. 9c, d) indicate that the muscle portion of the MTU was shorter in the least flexible subjects and the effects of training on the length/torque relationship also differed between the groups. For the least flexible subjects the relative torque increased at the longer muscle lengths implying an increase in muscle length with training, the relationship becoming very similar to that of the most flexible group for whom there was no change with training. Despite the increased length of the muscle of the least flexible group there was no significant change in the stiffness of the whole MTU. This

may indicate that the passive stiffness of the MTU largely reflects the characteristics of the connective tissue and tendons. Blazeovich et al. (2012) investigated the differences between flexible and inflexible subjects and concluded that the greater flexibility was mainly due to both longer muscle and tendon length and our observations of the two groups before training are largely consistent with this except that our two groups had similar pain perception (similar torque at end of stretch) while the flexible subjects of Blazeovich et al. (2012) study were prepared to tolerate higher torques. It is likely that in this latter study the flexible subjects were accustomed to the stretch sensations. Six weeks stretching training is reported to increase gastrocnemius fascicle length with the increase reaching a plateau after about 3 weeks, which is a comparable time scale to our training study (Simpson et al. 2017), thus being consistent with our suggestion of an increase in muscle length. Unfortunately that study did not measure MTU stiffness or tendon length so there is no independent evidence to support our suggestion that tendon length did not increase over this time scale. For the most flexible group there was no change in either of the two parameters measured, the passive stiffness and the hamstring muscle length, in which case the improvements in flexibility with training appear to be entirely due to changes in tolerance to stretch, and resulted in an average increased ROM of 10°. For the least flexible subjects, however, there may have been two factors contributing to their improvement, one being the increase in muscle length, the other an increased tolerance of stretch and it is possible that each contributed 10° to the overall 20° improvement in this group. We suggest that muscle length and tolerance of stretch sensations can adapt fairly rapidly to stretch training, within 4 weeks, but the connective tissue elements of the MTU which dominates the passive length tension relationship responds much more slowly, possibly over the course of years.

Limitations of this study

Although every effort was made to ensure that the POP group stretched further than the POD group, there was no way of documenting this in the group training sessions. One way of being sure would be to train a single muscle group over a prolonged period with the isokinetic dynamometer, as we did with the acute training study (Muanjai et al. 2017) although this would be very labour intensive. The present study has been restricted to young female subjects because the degree of stretching during training was determined by the subjects' perception of discomfort and pain and there are well documented differences between the sexes in pain perception and in the extent to which pain limits extensibility in the SLR (Marshall and Siegler 2014). Future studies might redress this imbalance and focus on

male subjects and, possibly on older subjects since loss of flexibility is one of the aspects of ageing. The fact that the less flexible subjects responded to training with an increase in muscle length that did not occur with the more flexible subjects suggests that future studies of flexibility training might use a more homogeneous group, although there was no indication in our data that there was any differences in the response to POD and POP training. The concentric angle/torque relationship depends on subjects making maximal efforts and this is not necessarily easy to with hamstrings at extended length and while shortening. A better determination of the length tension relationship of the hamstrings might be to measure isometric strength at a range of angles.

In conclusion, hamstrings stretching to POP for 4 weeks increased flexibility and had no detrimental effect in terms of loss of muscle function. However, the benefits were not significantly different from those achieved by stretching to POD so there is no justification for recommending painful stretching. The improvements in flexibility over 4 weeks of stretching training appear to be due to reduced perception of discomfort and pain although in less flexible subjects there may also be an increase in hamstring muscle length. It is likely that very much longer training may be necessary to modify the connective tissue elements of the MTU and achieve the degree of flexibility seen with many gymnasts and dancers.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval All procedures performed 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.

Informed consent Informed consent was obtained from all individual participants included in the study.

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