

RESEARCH ARTICLE

The effects of static stretching programs on muscle strength and muscle architecture of the medial gastrocnemius

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Abstract

Introduction

Static stretching (SS) programs are widely used in clinical and athletic settings. Many previous studies investigate the effect of SS program on muscle strength and muscle architecture (muscle thickness, and pennation angle). However, no consensus has been reached about the effect of SS programs on muscle strength and muscle architecture. The aim of this study was to investigate the effects of 6-week SS programs performed at different weekly frequencies on muscle strength, muscle thickness and pennation angle at different ankle joint positions.

Methods

A total of 24 healthy male volunteers were performed 6-week SS programs (2,160 s of SS: 360 s/week*6 weeks) and were randomized to a group that performed SS once a week, or a group that performed SS three times per week. Total time under stretching was equated between groups. The muscle strength (maximum voluntary isometric contraction) at three different ankle joints were assessed before and after the 6-week SS program. In addition, muscle thickness and pennation angle were assessed by ultrasonography before and after 6-week SS program.

Results

There were no significant changes in all variables before and after the 6-week SS program, regardless of weekly frequency ($p > 0.05$).

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Conclusions

Our results suggest that 6-week SS programs do not increase muscle strength or muscle architecture at different ankle joint positions, regardless of stretching frequency; however, no negative effect on these outcomes was observed, contrary to evidence on the immediate, detrimental effects of SS.

Introduction

Static stretching (SS) is widely used in clinical setting and has been reported to increase range of motion (ROM) [1, 2] and decrease muscle stiffness of the hamstring or gastrocnemius muscles [3–5]. In athletic settings, SS program is usually performed to prevent sport injury.

Many previous studies investigating SS have reported only the acute effects on ROM, muscle stiffness, and muscle strength. Although ROM is increased immediately following SS [6, 7], muscle strength is decreased [6, 8–10]; this phenomenon is called the stretching-induced force deficit. Consistently, data from reviews demonstrate a detrimental effect of SS on muscle strength and athletic performance [11–13]; therefore, Simic *et al.* recommended avoiding the use of SS alone during warm-up routines [12]. By contrast, the effect of SS programs of at least several weeks duration on muscle strength and athletic performance remains equivocal. Interestingly, there have been no studies reporting the stretching-induced force deficit after several weeks SS program. In addition, although some studies reported that there were significant changes in muscle strength and athletic performance after SS program [14, 15], other studies reported no significant effects [16–18], which have not reached the consensus. In addition, a systematic review about the effect of stretching program on muscle strength and athletic performance concluded that more studies were needed to confirm whether stretching programs could positively affect muscle strength and performance [19].

In animal models, previous studies showed that passive stretching might trigger mechanisms that are important for muscle hypertrophy, such as insulin-like and myogenic growth factors, stretch-activated channels, the AKT/mTOR pathway and protein synthesis [20–22]. Indeed, the studies reporting chronic effects of an SS program showed robust hypertrophy and perhaps hyperplasia after several weeks of SS program intervention [23, 24]. Conversely, in human studies the muscle thickness and pennation angle are usually assessed using ultrasonography. Previous studies showed that muscle size (cross-sectional area, muscle thickness and muscle volume) has a strong influence on muscle strength [25, 26]. In addition, the pennation angle of muscle fibers also influences muscle strength [27, 28]. Taken together, these elements of muscle architecture (muscle thickness and pennation angle) could strongly influence muscle strength. Some previous studies measured these indexes before and after SS program and reported that there were no significant changes in muscle thickness, pennation angle and fascicle length of medial gastrocnemius (MG) after 3–6 week SS program [5, 16–18]. However, Freitas and Mil-Homens (2015) investigated the effect of 8-week high-intensity SS program on muscle architecture and reported significant increase in the fascicle length of biceps femoris. In addition, there were reported significant increases in the muscle thickness and fascicle length of MG after six weeks of a machine-assisted SS program [29]. Since no consensus has been reached about the effect of SS program on muscle architecture as well as muscle strength, it is necessary to reconsider the effect of SS program on muscle architecture. Furthermore, if SS program could change the muscle architecture, it is expected to have a positive effect on muscle strength. In particular, it is possible that the change in fascicle length might affect the

muscle force at different angles (joint position). Therefore, along with investigating the effect of SS program on the muscle architecture, it is necessary to clarify the effect of SS program on muscle strength at different joint angles.

A SS program is considered to comprise intensity, duration, and repetition of stretches; rest intervals; and intervention frequency. In previous studies, increased duration and intensity of stretching resulted in greater increases and decreases, respectively, in ROM and passive stiffness [30–32]. In addition, Marques *et al.* (2009) found that a stretching frequency of three times per week was more effective than that of one or five times per week in increasing ROM. It is possible that stretching frequency could determine the effect of SS programs on muscle strength and architecture, in addition to ROM. However, the effects of stretching frequency on muscle strength and architecture under volume-equated conditions are unclear. This information could be useful for coaches or therapists when prescribing SS programs.

Therefore, the aim of this study was to compare the effects of two 6-week SS programs with different stretching frequencies under volume-equated conditions on muscle strength and architecture at different ankle joint positions.

Methods

Experimental design

A quasi-randomized controlled trial was conducted, which investigated the changes in muscle strength and architecture (defined as muscle thickness and penetration angle) in two routine SS programs with different stretching frequencies as follows: 360 s of SS conducted 1 time per week (1 time/week group) versus 120 s of SS conducted 3 times per week (3 times/week group). Muscle strength, muscle thickness, and penetration angle were measured at baseline (before intervention [PRE]) and after the 6-week SS program (after intervention [POST]) in both groups. A familiarization trial of muscle strength measurement was performed >3 days before the PRE evaluation. Following the PRE evaluation, participants were randomly allocated to either of the comparison groups in a 1:1 ratio using the alternation method. To control for immediate SS effects, all outcome measurements were performed ≥ 24 h after the final SS session. Participants were instructed not to initiate any other SS or strength-training programs during the study period.

Participants

A total of 24 healthy male volunteers, who were non-athletes, participated in this study (mean \pm SD; age, 20.8 ± 0.9 years; height, 168.9 ± 5.0 cm; body mass, 61.3 ± 6.2 kg). All participants engaged in sports at a recreational level, but were not involved in any regular resistance or flexibility training. All participants were fully informed of the procedures and purpose of the study, and provided written informed consent. The Ethics Committee of the Niigata University of Health and Welfare, Niigata, Japan (Procedure #17677) approved the study and complied with the requirements of the Declaration of Helsinki.

Maximum voluntary isometric contraction

Participants were seated in an isokinetic dynamometer chair at a 0° knee angle (that is, the anatomical position) with adjustable belts fixed over their trunk and pelvis (Biodex System 3.0, Biodex Medical Systems, Inc., Shirley, NY, USA). Participants were reclined (70° hip angle and 0° full extension) to prevent tension in the posterior knee. The trunk and pelvis were firmly fixed with straps, and trunk movement was restricted by holding the handle with both hands. The maximal voluntary isometric contraction (MVIC) of the MG was measured with

the ankle joint at 30° plantarflexion, in the neutral position, and at 15° dorsiflexion, which were determined by the ROM through which all participants could exert force. To obtain measurements, the ankle joint of the dominant leg was securely attached to the footplate of the dynamometer using a velcro strap. A soft cloth was inserted between the velcro strap and instep to prevent movement of the ankle joint. Two MVICs were performed for 5 s at each ankle position, and the average value of both MVICs used for analyses. Strong, verbal encouragement was provided to promote participants' maximal effort during contractions.

Muscle thickness and pennation angle

Participants were instructed to lie on a dynamometer table in the prone position, with their hips secured in place using an adjustable lap belt. The knee of the dominant leg was maintained in full extension, and the foot of the same leg was attached to the dynamometer footplate with adjustable belts.

B-mode ultrasonography (Aplio 500, Toshiba Medical Systems, Tochigi, Japan) with a 5–14 MHz linear probe was used to assess the muscle thickness and pennation angle of the MG. The longitudinal ultrasound image of the MG at 30% of the lower-leg length, measured from the popliteal crease to the lateral malleolus near the point of the maximal cross-sectional area of the lower leg, was obtained [33, 34]. Muscle thickness, defined as the distance between the inside edges of the fascia, and pennation angle, defined as the angle of the fascicle insertion into the deep aponeurosis, were measured with the ankle joint at 30° plantarflexion, in the neutral position, and at 15° dorsiflexion, consistent with the MVIC measurements; each measurement was performed once. Muscle thickness and pennation angle were determined using image processing software (ImageJ, National Institutes of Health, Bethesda, MD, USA). Muscle thickness was determined as the mean of the distances between the deep and superficial aponeuroses measured at both ends of each image [35, 36]. In addition, pennation angle was determined as the mean of the three fascicles as the angle between fascicle and deep aponeurosis. The test-retest reliability of the ultrasound measurements was determined by coefficient variation (CV) for eight participants. The CVs for muscle thickness were $3.7 \pm 1.9\%$ in 30° plantarflexion, $3.5 \pm 2.4\%$ in neutral position and $3.5 \pm 2.4\%$ in 15° dorsiflexion. In addition, the CVs for pennation angle were $4.0 \pm 3.2\%$ in 30° plantarflexion, $3.7 \pm 2.4\%$ in neutral position and $2.2 \pm 1.8\%$ in 15° dorsiflexion.

Static stretching programs

Participants in both groups were instructed to perform the SS program they had been assigned to for 6 weeks using a stretching board (Asahi stretching board, Asahi Corp., Gifu, Japan). Participants stood erect with one foot on the stretching board and the other on its edge and both arms against a wall in front of the body to provide balance [16, 33]. Stretching intensity was defined as the greatest tolerated dorsiflexion angle, determined during a test conducted on the stretching board. However, participants who could tolerate $>35^\circ$ dorsiflexion, which was the maximal angle permitted by the stretching board, were instructed to maintain the stretching intensity by moving their body mass forward. All SS programs were performed in the laboratory under direct supervision of research team. The total weekly duration of SS was 360 s in both groups. The frequency of performing SS was every 7 days and every 2–3 days in the 1-time/week and 3-times/week groups, respectively.

Statistical analyses

IBM SPSS Statistics version 24.0 (IBM Corp., Armonk, NY, USA) was used to conduct statistical analyses. Between-group differences in anthropometric characteristics, muscle strength,

muscle thickness, and pennation angle relative to PRE evaluation values were determined using unpaired *t*-tests. For muscle strength, muscle thickness, and pennation angle, a split-plot analysis of variance (ANOVA) using two factors [time (PRE versus POST evaluation) and group (1-time/week versus 3-times/week)] was used to determine the interaction and main effects. Classification of effect size (ES) was set where $\eta^2 < 0.01$ was considered small, 0.02–0.1 was considered medium and over 0.1 was considered to be a large effect size based on previous studies [37, 38]. A *post-hoc* analysis was conducted, using a paired *t*-test in each group, to determine differences between PRE and POST evaluation scores. ES were calculated as differences in the mean value between PRE- and POST-evaluation divided by the pooled standard deviation (SD) [37]. In addition, an ES of 0.00–0.19 was considered as trivial, 0.20–0.49 as small, 0.50–0.79 as moderate and ≥ 0.80 as large [37, 39]. Statistical significance was defined as $p < 0.05$. Descriptive data are reported as means \pm SD.

Results

All participants completed the SS program they were assigned to in full, and there were no drop-outs. The characteristics of study participants are reported in Table 1. There were no significant differences in age, height, or body mass between the two study groups at baseline.

Effects of static stretching programs on maximum voluntary isometric contraction

The effects of the SS program on MVIC in both groups are reported in Tables 2 and 5. There were no significant differences between groups in PRE evaluation scores, determined by the unpaired *t*-test. The split-plot ANOVA indicated no significant interaction effects for MVIC at 30° plantarflexion or in the neutral position. There was a significant interaction effect for MVIC at 15° dorsiflexion. The *post-hoc* test revealed that there were no significant differences between PRE and POST evaluation in both groups (1-time/week, $p = 0.08$, ES = 0.33, and 3-times/week, $p = 0.149$, ES = -0.37).

Table 1. Characteristics of participants participating in two static stretching programs.

	One-time/week group (N = 12)	Three-times/week group (N = 12)	P-value
age (years)	21.0 \pm 0.9	20.5 \pm 0.8	P = 0.177
height (cm)	167.7 \pm 4.2	170.0 \pm 5.4	P = 0.273
weight (kg)	61.8 \pm 6.1	60.8 \pm 6.2	P = 0.684

Data presented as mean \pm standard deviation.

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Table 2. Values for muscle strength before and after 6-week static stretching intervention program.

(Nm)		One-time/week group (N = 12)	Three-times/week group (N = 12)	Interaction effect
MVIC at 30° plantarflexion	PRE	64.8 \pm 23.4	65.7 \pm 23.8	F = 0.001, P = 0.975
	POST	64.0 \pm 20.4	65.1 \pm 29.5	$\eta^2 = 0.000$
MVIC at neutral position	PRE	172.0 \pm 38.5	189.3 \pm 42.1	F = 0.08, P = 0.78
	POST	169.4 \pm 26.3	183.7 \pm 49.8	$\eta^2 = 0.004$
MVIC at 15° dorsiflexion	PRE	200.2 \pm 56.1	244.2 \pm 52.5	F = 5.41, P = 0.03
	POST	216.6 \pm 42.7	222.3 \pm 65.5	$\eta^2 = 0.197$

PRE, before static stretching intervention program; POST, after static stretching intervention program; MVIC, maximum voluntary isometric contraction. Data presented as mean \pm standard deviation.

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Table 3. Values for muscle thickness before and after 6-week static stretching intervention program.

(cm)		One-time/week group (N = 12)	Three-times/week group (N = 12)	Interaction effect
Muscle Thickness at 30° plantarflexion	PRE	1.61 ± 0.19	1.60 ± 0.22	F = 0.589, P = 0.451
	POST	1.62 ± 0.16	1.54 ± 0.26	$\eta^2 = 0.026$
Muscle Thickness at neutral position	PRE	1.68 ± 0.23	1.59 ± 0.25	F = 0.030, P = 0.865
	POST	1.66 ± 0.16	1.60 ± 0.29	$\eta^2 = 0.001$
Muscle Thickness at 15° dorsiflexion	PRE	1.65 ± 0.29	1.55 ± 0.28	F = 0.021, P = 0.887
	POST	1.68 ± 0.16	1.59 ± 0.25	$\eta^2 = 0.001$

PRE, before static stretching intervention program; POST, after static stretching intervention program. Data presented as mean ± standard deviation.

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Table 4. Values for pennation angle before and after 6-week static stretching intervention program.

(cm)		One-time/week group (N = 12)	Three-times/week group (N = 12)	Interaction effect
Pennation Angle at 30° plantarflexion	PRE	22.9 ± 3.8	21.8 ± 3.3	F = 0.804, P = 0.380
	POST	21.9 ± 2.8	21.7 ± 3.2	$\eta^2 = 0.035$
Pennation Angle at neutral position	PRE	19.4 ± 3.5	18.7 ± 2.4	F < 0.001, P = 0.996
	POST	19.2 ± 2.6	18.5 ± 2.3	$\eta^2 < 0.001$
Pennation Angle at 15° dorsiflexion	PRE	17.7 ± 2.7	17.1 ± 2.8	F = 0.064, P = 0.803
	POST	17.1 ± 2.2	16.8 ± 2.0	$\eta^2 = 0.003$

PRE, before static stretching intervention program; POST, after static stretching intervention program. Data presented as mean ± standard deviation.

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Effects of static stretching programs on muscle thickness and pennation angle

The effects of the SS program on muscle thickness and pennation angle in both groups are reported in Tables 3, 4 and 5, respectively. The split-plot ANOVA indicated no significant interaction effects for muscle thickness at 30° plantarflexion the neutral position or 15° dorsiflexion. In addition, there were no significant interaction effects for pennation angle at 30° plantarflexion or the neutral position or 15° dorsiflexion.

Table 5. Effect size values for maximum voluntary isometric contraction (MVIC), muscle thickness and pennation angle according to groups after 6-week static stretching intervention program.

	One-time/week group (N = 12)	Three-times/week group (N = 12)
MVIC at 30° plantarflexion	-0.04	-0.02
MVIC at neutral position	-0.08	-0.12
MVIC at 15° dorsiflexion	0.33	-0.37
Muscle Thickness at 30° plantarflexion	0.08	-0.24
Muscle Thickness at neutral position	-0.06	0.01
Muscle Thickness at 15° dorsiflexion	0.14	0.18
Pennation Angle at 30° plantarflexion	-0.29	-0.03
Pennation Angle at neutral position	-0.05	-0.07
Pennation Angle at 15° dorsiflexion	-0.03	-0.12

Data presented as mean ± standard deviation.

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Discussion

We investigated the effects of a 6-week SS program on muscle strength and architecture of the MG at different ankle joint positions, comparing two different stretching frequencies under volume-equated conditions. We found no significant effects of the SS program on these outcomes, regardless of stretching frequency. It is important to mention that according to the literature presented in a recent review on the topic [40] and our updated knowledge, this is the first study that compared effects of different frequencies of SS on muscular adaptations that equalized the weekly volume of training. Equating the volume of training when studying training frequency is necessary to determine causality as to verify the actual influence of weekly frequency of training; otherwise, the effects of volume confound the ability to draw proper inferences [41].

In previous studies of animal models, SS programs induced muscle hypertrophy [21, 23, 24]; therefore, it was hypothesized that an SS program could produce a similar effect in humans, which could lead to increased muscle strength and athletic performance. However, our results were inconsistent with this hypothesis and revealed no significant changes in muscle thickness and pennation angle, which suggested that there was no change in fascicle length, or in muscle strength at different ankle joint positions. This discrepancy between our *in vivo* study and previous *in vitro* studies might be explained by the stretching intensity and duration of our program [42]. Nonetheless, the lack of SS effects on muscle architecture (muscle thickness, pennation angle, and fascicle length) were also reported in other previous studies [5, 16, 17, 43]. One work reported that there was a positive effect for increased muscle thickness, from Simpson *et al.* (2017), albeit with the caveat that raw data showed similar changes versus control [44, 45], calling into question the practical relevance of the findings.

One point that seems to be important to induce architectural changes in muscle is SS training intensity [46]. Although we were unable to measure stretching intensity because we used a stretching board in our SS program, the stretching intensity was expected to be much lower than the required for muscle hypertrophy, such the achieved during resistance-training. The stretching program in the previous study was carried out in a leg-press loaded with 20% of MVIC, which induced muscle hypertrophy [29]. It is possible that higher stretching intensity in the previous study contributed to the discrepancy with the current study. Nonetheless, in addition to training load, total exercise volume, defined as the product of training load and repetition number, has become recognized as an important factor for muscle hypertrophy. In this study, each participant performed 2,160 s of SS (360 s/week*6 weeks); however, the total exercise volume might be insufficient to induce muscle hypertrophy because the SS intensity and duration were low. For instance, in studies from Freitas and Mil-Homens (2015) and Simpson *et al.* (2017), which verified increases in fascicle length, the total time under stretching of the SS programs were of approximately 11,250 s (450 s*3.1 ± 0.8 sessions per week*8week) or 5,400 s, respectively; too higher than performed in our study.

Chen *et al.* (2011) reported a significant increase in concentric hamstring strength following an 8-week SS program, whereas Nakao *et al.* (2019) found no significant changes in isometric and concentric hamstring strength, but a significant increase in the peak angle of concentric strength after a 4-week SS program. In addition, Freitas and Mil-Homens (2015) reported a significant increase in the fascicle length of the biceps femoris following an 8-week high-intensity SS program. These studies demonstrate that, in the case of hamstring, an SS program may change the muscle architecture (e.g., muscle thickness or pennation angle), which may result in increased strength or performance. Among studies evaluating the gastrocnemius muscle, Akagi *et al.* (2014) reported no significant changes in MVIC and muscle thickness And Blazevich *et al.* (2014) reported no significant change in fascicle length after a 3-week

SS program; the latter finding was supported by Nakamura *et al.* (2012) and Konrad *et al.* (2014). Previous studies have noted the difference in muscle architecture (muscle thickness, pennation angle, fascicle length and tendon length) between the biceps femoris and medial gastrocnemius muscle [47, 48]. This could affect differences arising in prior literature on the chronic effect of SS training. Collectively, these results suggest that the effects of SS training on muscle strength and architecture may differ on the basis of the target muscle and total exercise volume (stretching intensity and duration). Therefore, future studies should elucidate the variation in effects of SS programs on strength and architecture of different muscles.

An important factor regarding changes in muscle architecture might be nutrition. Our study did not incorporate a nutritional component to the SS program. However, Simpson *et al.* (2017) reported increased MG thickness and fascicle length after a 6-week SS program which included subsequent protein intake, suggesting this might have moderated the hypertrophic effect of the SS program. Future studies should consider the combined effects of SS programs, nutritional interventions, and resistance training.

This study has several limitations. The major limitations of this study were the small sample size and the short duration of the intervention, although it is similar to several previous studies on SS [5, 16, 17, 29, 43]. In this study, we calculated the sample size needed for split-plot ANOVA (alpha error = 0.05, power = 0.80, effect size = 0.4 [large]), and the requisite number of participants was 14 in each group. Therefore, it is feasible to be underpowered in this study. We calculated the effect sizes for the split-plot ANOVA (η^2) and different between PRE and POST in each group. As the results of effect sizes, all variables were trivial or small. Therefore, we have assumed that the results of this study have not been underpowered. However, further study is needed to investigate the chronic effect of SS programs on muscle strength and muscle architecture using more subjects for a longer duration. In addition, the effects of SS programs on muscle performance (for example, jumping or sprinting) should be considered. In addition, the effect of a 6-week SS program on athletes is not known, and future studies are needed to investigate the effect of long-term SS program on muscle performance in other populations, such as athletes.

Conclusion

In conclusion, a 6-week SS program targeting the MG muscle did not increase muscle strength or hypertrophy; however, we also found no negative effect on these outcomes, contrary to evidence on the immediate, detrimental effects of SS.

Supporting information

S1 Data.
(XLSX)

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References

1. Bandy WD, Irion JM, Briggler M. The effect of time and frequency of static stretching on flexibility of the hamstring muscles. *Physical therapy*. 1997; 77(10):1090–6. <https://doi.org/10.1093/ptj/77.10.1090> PMID: 9327823.
2. Bandy WD, Irion JM. The effect of time on static stretch on the flexibility of the hamstring muscles. *Physical therapy*. 1994; 74(9):845–50; discussion 50–2. <https://doi.org/10.1093/ptj/74.9.845> PMID: 8066111.
3. Nakamura M, Ikezoe T, Umegaki H, Kobayashi T, Nishishita S, Ichihashi N. Changes in Passive Properties of the Gastrocnemius Muscle-Tendon Unit During a 4-Week Routine Static-Stretching Program. *Journal of sport rehabilitation*. 2017; 26(4):263–8. <https://doi.org/10.1123/jsr.2015-0198> PMID: 27632863.
4. Ichihashi N, Umegaki H, Ikezoe T, Nakamura M, Nishishita S, Fujita K, et al. The effects of a 4-week static stretching programme on the individual muscles comprising the hamstrings. *Journal of sports sciences*. 2016; 34(23):2155–9. <https://doi.org/10.1080/02640414.2016.1172725> PMID: 27113325.
5. Nakamura M, Ikezoe T, Takeno Y, Ichihashi N. Effects of a 4-week static stretch training program on passive stiffness of human gastrocnemius muscle-tendon unit in vivo. *European journal of applied physiology*. 2012; 112(7):2749–55. <https://doi.org/10.1007/s00421-011-2250-3> PMID: 22124523.
6. Ryan ED, Beck TW, Herda TJ, Hull HR, Hartman MJ, Stout JR, et al. Do practical durations of stretching alter muscle strength? A dose-response study. *Medicine and science in sports and exercise*. 2008; 40(8):1529–37. Epub 2008/07/11. <https://doi.org/10.1249/MSS.0b013e31817242eb> PMID: 18614936.
7. Hoge KM, Ryan ED, Costa PB, Herda TJ, Walter AA, Stout JR, et al. Gender differences in musculotendinous stiffness and range of motion after an acute bout of stretching. *Journal of strength and conditioning research / National Strength & Conditioning Association*. 2010; 24(10):2618–26. Epub 2010/10/05. <https://doi.org/10.1519/JSC.0b013e3181e73974> PMID: 20885189.
8. Fowles JR, Sale DG, MacDougall JD. Reduced strength after passive stretch of the human plantarflexors. *J Appl Physiol*. 2000; 89(3):1179–88. Epub 2000/08/24. <https://doi.org/10.1152/jappl.2000.89.3.1179> PMID: 10956367.
9. Avela J, Kyrolainen H, Komi PV. Altered reflex sensitivity after repeated and prolonged passive muscle stretching. *J Appl Physiol*. 1999; 86(4):1283–91. Epub 1999/04/08. <https://doi.org/10.1152/jappl.1999.86.4.1283> PMID: 10194214.
10. Behm DG, Button DC, Butt JC. Factors affecting force loss with prolonged stretching. *Can J Appl Physiol*. 2001; 26(3):261–72. Epub 2001/07/07. PMID: 11441230.
11. Behm DG, Chaouachi A. A review of the acute effects of static and dynamic stretching on performance. *European journal of applied physiology*. 2011; 111(11):2633–51. <https://doi.org/10.1007/s00421-011-1879-2> PMID: 21373870.
12. Simic L, Sarabon N, Markovic G. Does pre-exercise static stretching inhibit maximal muscular performance? A meta-analytical review. *Scandinavian journal of medicine & science in sports*. 2013; 23(2):131–48. <https://doi.org/10.1111/j.1600-0838.2012.01444.x> PMID: 22316148.
13. Kay AD, Blazevich AJ. Effect of acute static stretch on maximal muscle performance: a systematic review. *Medicine and science in sports and exercise*. 2012; 44(1):154–64. Epub 2011/06/11. <https://doi.org/10.1249/MSS.0b013e318225cb27> PMID: 21659901.
14. Chen CH, Nosaka K, Chen HL, Lin MJ, Tseng KW, Chen TC. Effects of flexibility training on eccentric exercise-induced muscle damage. *Medicine and science in sports and exercise*. 2011; 43(3):491–500. <https://doi.org/10.1249/MSS.0b013e3181f315ad> PMID: 20689450.

15. Nelson AG, Kokkonen J, Winchester JB, Kalani W, Peterson K, Kenly MS, et al. A 10-week stretching program increases strength in the contralateral muscle. *Journal of strength and conditioning research / National Strength & Conditioning Association*. 2012; 26(3):832–6. Epub 2012/02/03. <https://doi.org/10.1519/JSC.0b013e3182281b41> PMID: 22297415.
16. Akagi R, Takahashi H. Effect of a 5-week static stretching program on hardness of the gastrocnemius muscle. *Scandinavian journal of medicine & science in sports*. 2014; 24(6):950–7. <https://doi.org/10.1111/sms.12111> PMID: 23944602.
17. Blazeovich AJ, Cannavan D, Waugh CM, Miller SC, Thorlund JB, Aagaard P, et al. Range of motion, neuromechanical, and architectural adaptations to plantar flexor stretch training in humans. *Journal of applied physiology*. 2014; 117(5):452–62. Epub 2014/06/21. <https://doi.org/10.1152/jappphysiol.00204.2014> PMID: 24947023.
18. Konrad A, Tilp M. Increased range of motion after static stretching is not due to changes in muscle and tendon structures. *Clinical biomechanics*. 2014; 29(6):636–42. <https://doi.org/10.1016/j.clinbiomech.2014.04.013> PMID: 24856792.
19. Medeiros DM, Lima CS. Influence of chronic stretching on muscle performance: Systematic review. *Human movement science*. 2017; 54:220–9. Epub 2017/05/21. <https://doi.org/10.1016/j.humov.2017.05.006> PMID: 28527424.
20. Riley DA, Van Dyke JM. The effects of active and passive stretching on muscle length. *Physical medicine and rehabilitation clinics of North America*. 2012; 23(1):51–7, x. Epub 2012/01/14. <https://doi.org/10.1016/j.pmr.2011.11.006> PMID: 22239873.
21. Tatsumi R. Mechano-biology of skeletal muscle hypertrophy and regeneration: possible mechanism of stretch-induced activation of resident myogenic stem cells. *Animal science journal = Nihon chikusan Gakkaiho*. 2010; 81(1):11–20. Epub 2010/02/19. <https://doi.org/10.1111/j.1740-0929.2009.00712.x> PMID: 20163667.
22. Mohamad NI, Nosaka K, Cronin J. Maximizing hypertrophy: Possible contribution of stretching in the interset rest period. *Strength and conditioning journal*. 2011; 33(1):81–7. <https://doi.org/10.1519/SSC.0b013e3181fe7164>
23. Goldspink DF, Cox VM, Smith SK, Eaves LA, Osbaldeston NJ, Lee DM, et al. Muscle growth in response to mechanical stimuli. *The American journal of physiology*. 1995; 268(2 Pt 1):E288–97. Epub 1995/02/01. <https://doi.org/10.1152/ajpendo.1995.268.2.E288> PMID: 7532362.
24. Goldberg AL, Etlinger JD, Goldspink DF, Jablecki C. Mechanism of work-induced hypertrophy of skeletal muscle. *Medicine and science in sports*. 1975; 7(3):185–98. Epub 1975/01/01. PMID: 128681.
25. Akagi R, Takai Y, Ohta M, Kanehisa H, Kawakami Y, Fukunaga T. Muscle volume compared to cross-sectional area is more appropriate for evaluating muscle strength in young and elderly individuals. *Age and ageing*. 2009; 38(5):564–9. Epub 2009/07/15. <https://doi.org/10.1093/ageing/afp122> PMID: 19596739.
26. Trezise J, Collier N, Blazeovich AJ. Anatomical and neuromuscular variables strongly predict maximum knee extension torque in healthy men. *European journal of applied physiology*. 2016; 116(6):1159–77. Epub 2016/04/15. <https://doi.org/10.1007/s00421-016-3352-8> PMID: 27076217.
27. Wakahara T, Kanehisa H, Kawakami Y, Fukunaga T, Yanai T. Relationship between muscle architecture and joint performance during concentric contractions in humans. *Journal of applied biomechanics*. 2013; 29(4):405–12. Epub 2012/08/29. <https://doi.org/10.1123/jab.29.4.405> PMID: 22927507.
28. Ikegawa S, Funato K, Tsunoda N, Kanehisa H, Fukunaga T, Kawakami Y. Muscle force per cross-sectional area is inversely related with pennation angle in strength trained athletes. *Journal of strength and conditioning research / National Strength & Conditioning Association*. 2008; 22(1):128–31. Epub 2008/02/26. <https://doi.org/10.1519/JSC.0b013e31815f2fd3> PMID: 18296965.
29. Simpson CL, Kim BDH, Bourcet MR, Jones GR, Jakobi JM. Stretch training induces unequal adaptation in muscle fascicles and thickness in medial and lateral gastrocnemii. *Scandinavian journal of medicine & science in sports*. 2017; 27(12):1597–604. Epub 2017/02/01. <https://doi.org/10.1111/sms.12822> PMID: 28138986.
30. Ryan ED, Herda TJ, Costa PB, Defreitas JM, Beck TW, Stout J, et al. Determining the minimum number of passive stretches necessary to alter musculotendinous stiffness. *Journal of sports sciences*. 2009; 27(9):957–61. <https://doi.org/10.1080/02640410902998254> PMID: 19629845.
31. Boyce D, Brosky JA Jr., Determining the minimal number of cyclic passive stretch repetitions recommended for an acute increase in an indirect measure of hamstring length. *Physiotherapy theory and practice*. 2008; 24(2):113–20. <https://doi.org/10.1080/09593980701378298> PMID: 18432514.
32. Nakamura M, Ikezoe T, Takeno Y, Ichihashi N. Time course of changes in passive properties of the gastrocnemius muscle-tendon unit during 5 min of static stretching. *Manual therapy*. 2013; 18(3):211–5. <https://doi.org/10.1016/j.math.2012.09.010> PMID: 23294911.

33. Akagi R, Takahashi H. Acute effect of static stretching on hardness of the gastrocnemius muscle. *Medicine and science in sports and exercise*. 2013; 45(7):1348–54. <https://doi.org/10.1249/MSS.0b013e3182850e17> PMID: 23299765.
34. Nakamura M, Ikezoe T, Kobayashi T, Umegaki H, Takeno Y, Nishishita S, et al. Acute effects of static stretching on muscle hardness of the medial gastrocnemius muscle belly in humans: an ultrasonic shear-wave elastography study. *Ultrasound in medicine & biology*. 2014; 40(9):1991–7. <https://doi.org/10.1016/j.ultrasmedbio.2014.03.024> PMID: 24973829.
35. Ema R, Wakahara T, Miyamoto N, Kanehisa H, Kawakami Y. Inhomogeneous architectural changes of the quadriceps femoris induced by resistance training. *European journal of applied physiology*. 2013; 113(11):2691–703. <https://doi.org/10.1007/s00421-013-2700-1> PMID: 23949789.
36. Blazeovich AJ, Gill ND, Zhou S. Intra- and intermuscular variation in human quadriceps femoris architecture assessed in vivo. *Journal of anatomy*. 2006; 209(3):289–310. Epub 2006/08/25. <https://doi.org/10.1111/j.1469-7580.2006.00619.x> PMID: 16928199
37. Cohen J, editor. *Statistical power analysis for the behavioral sciences*. 2nd ed. ed. Routledge: Hillsdale; 1988.
38. Akiyama K, Akagi R, Hirayama K, Hirose N, Takahashi H, Fukubayashi T. Shear Modulus of the Lower Leg Muscles in Patients with Medial Tibial Stress Syndrome. *Ultrasound in medicine & biology*. 2016; 42(8):1779–83. <https://doi.org/10.1016/j.ultrasmedbio.2016.03.010> PMID: 27129903.
39. Nakamura M, Sato S, Hiraizumi K, Kiyono R, Fukaya T, Nishishita S. Effects of static stretching programs performed at different volume-equated weekly frequencies on passive properties of muscle-tendon unit. *Journal of biomechanics*. 2020:109670. Epub 2020/02/10. <https://doi.org/10.1016/j.jbiomech.2020.109670> PMID: 32035662.
40. Freitas SR, Mendes B, Le Sant G, Andrade RJ, Nordez A, Milanovic Z. Can chronic stretching change the muscle-tendon mechanical properties? A review. *Scandinavian journal of medicine & science in sports*. 2018; 28(3):794–806. Epub 2017/08/13. <https://doi.org/10.1111/sms.12957> PMID: 28801950.
41. Schoenfeld BJ, Grgic J, Krieger J. How many times per week should a muscle be trained to maximize muscle hypertrophy? A systematic review and meta-analysis of studies examining the effects of resistance training frequency. *Journal of sports sciences*. 2018:1–10. Epub 2018/12/19. <https://doi.org/10.1080/02640414.2018.1555906> PMID: 30558493.
42. Lowe DA, Alway SE. Animal models for inducing muscle hypertrophy: are they relevant for clinical applications in humans? *The Journal of orthopaedic and sports physical therapy*. 2002; 32(2):36–43. Epub 2002/02/13. <https://doi.org/10.2519/jospt.2002.32.2.36> PMID: 11838579.
43. Lima KM, Carneiro SP, Alves Dde S, Peixinho CC, de Oliveira LF. Assessment of muscle architecture of the biceps femoris and vastus lateralis by ultrasound after a chronic stretching program. *Clinical journal of sport medicine: official journal of the Canadian Academy of Sport Medicine*. 2015; 25(1):55–60. Epub 2014/01/24. <https://doi.org/10.1097/jsm.000000000000069> PMID: 24451696.
44. Nunes JP, Nakamura M, Schoenfeld BJ, Cyrino ES. The data do not seem to support the effect of stretch training on increasing muscle thickness. *Scandinavian journal of medicine & science in sports*. 2018; 28(12):2767–8. Epub 2018/08/26. <https://doi.org/10.1111/sms.13285> PMID: 30144180.
45. Jakobi JM, Simpson CL, Smart RR, O'Connor B. Response to Nunes and colleagues letter: The data do not seem to support the effect of stretch training in increasing MT. *Scandinavian journal of medicine & science in sports*. 2018; 28(12):2769–71. Epub 2018/09/16. <https://doi.org/10.1111/sms.13300> PMID: 30218619.
46. Freitas SR, Mil-Homens P. Effect of 8-week high-intensity stretching training on biceps femoris architecture. *Journal of strength and conditioning research / National Strength & Conditioning Association*. 2015; 29(6):1737–40. Epub 2014/12/09. <https://doi.org/10.1519/jsc.0000000000000800> PMID: 25486299.
47. Stenroth L, Peltonen J, Cronin NJ, Sipila S, Finni T. Age-related differences in Achilles tendon properties and triceps surae muscle architecture in vivo. *Journal of applied physiology*. 2012; 113(10):1537–44. <https://doi.org/10.1152/jappphysiol.00782.2012> PMID: 23042907.
48. Kellis E, Galanis N, Natsis K, Kapetanios G. Muscle architecture variations along the human semitendinosus and biceps femoris (long head) length. *Journal of electromyography and kinesiology: official journal of the International Society of Electrophysiological Kinesiology*. 2010; 20(6):1237–43. Epub 2010/08/24. <https://doi.org/10.1016/j.jelekin.2010.07.012> PMID: 20727788.