European Journal of Applied Physiology (2021) 121:331–337
https://doi.org/10.1007/s00421-020-04524-0

ORIGINAL ARTICLE

Acute effect of passive one-legged intermittent static stretching on regional blood flow in young men

Yosuke Yamato[1,2] · Yuya Higaki[2] · Shumpei Fujie[2] · Natsuki Hasegawa[2] · Naoki Horii[2,3] · Hiroki Aoyama[1] · Yoshihiro Yamashina[1] · Shigehiko Ogoh[4] · Motoyuki Iemitsu[2]

Received: 23 April 2020 / Accepted: 3 October 2020 / Published online: 20 October 2020

Abstract

Purpose Passive stretching reduces stiffness in the lower limb arteries of the stretched limb. To address this physiological mechanism, we measured the change in shear rate in the posterior tibial artery during a single bout of one-legged passive calf stretching compared with that in the non-stretched leg.

Methods The diameter, mean blood velocity, blood flow, and shear rate in the posterior tibial artery were measured using Doppler ultrasound before (baseline), during, and after a one-legged passive intermittent calf stretching procedure (six repetitions of 30-s static stretch with 10-s relaxation) in nine healthy young men.

Results In the posterior tibial artery of the stretched leg, the arterial diameter significantly decreased from baseline during the stretching period (baseline vs. stretching period of the 6th set, 0.19 ± 0.01 vs. 0.18 ± 0.01 cm, $P < 0.05$) without any change in shear rate and mean blood velocity. In contrast, during the relaxation period, the mean blood velocity (baseline vs. relaxation period of the 5th set, 2.98 ± 0.54 vs. 6.25 ± 1.48 cm/s) increased, and consequently, the shear rate (baseline vs. relaxation period of the 5th set, 66.75 ± 15.39 vs. 122.85 ± 29.40 s^{-1}) increased (each $P < 0.01$); however, there was no change in arterial diameter. In contrast, these values in the non-stretched leg were unchanged at all-time points.

Conclusions The stretching procedure increased the shear rate in the peripheral artery of the stretched leg during the relaxation period. This finding indicates that the local hemodynamic response (possibly through endothelial function), resulting from an increase in shear stress, may contribute to stretching-induced attenuation of local arterial stiffness.

Keywords Stretching · Skeletal muscle · Shear rate · Blood flow · Hemodynamics

Abbreviations

ANOVA	Analysis of variance
NO	Nitic oxide
SE	Standard error
V_{mean}	Mean blood velocity

Communicated by Toshio Moritani.

✉ Motoyuki Iemitsu
iemitsu@fc.ritsumei.ac.jp

[1] Department of Physical Therapy, Aino University, Osaka, Japan

[2] Faculty of Sport and Health Science, Ritsumeikan University, 1-1-1 Nojihigashi, Kusatsu, Shiga 525-8577, Japan

[3] Research Fellow of Japan Society for the Promotion of Science, Tokyo, Japan

[4] Department of Biomedical Engineering, Toyo University, Saitama, Japan

Introduction

Some epidemiological studies suggest that poor trunk flexibility is associated with high systemic and central arterial stiffness; thus, measurement of trunk flexibility may be a useful a marker of the risk of future cardiovascular events (Yamamoto et al. 2009; Nishiwaki et al. 2014). Additionally, in training investigations, it has been demonstrated that 4 weeks or 6 months of static stretching intervention reduces systemic arterial stiffness (Nishiwaki et al. 2015; Shinno et al. 2017). These findings indicate that habitual stretching accompanied by prevention of arterial stiffening may decrease the risk of cardiovascular diseases. Interestingly, in our recent study, besides habitual stretching, it has been demonstrated that one bout of passive intermittent leg stretching acutely decreased pulse wave velocity as an index of arterial stiffness, and this acute reduction in arterial stiffness occurred only in the artery of the stretched leg but not in the artery of the non-stretched leg (Yamato et al. 2017).

However, the mechanism of static stretching-induced acute attenuation of arterial stiffness remains unknown.

One possible mechanism may be an acute increase in nitric oxide (NO) synthesis during stretching. Indeed, attenuated arterial stiffness (during exercise) is associated with vasodilation in the peripheral vasculature induced by an increase in shear stress (e.g., shear rate) accompanied by NO synthesis in endothelial cells (Higashi and Yoshizumi 2004; Zhou et al. 2014; Schmitt et al. 2005). Therefore, the acute reduction in arterial stiffness caused by our stretching procedure may be due to the acute increase in shear rate. Indeed, Venturelli et al. (2019) did not measure shear rate, but a 15-s relaxation after a 45-s static stretching increased blood flow in the femoral artery of the stretched leg without a change in arterial diameter, indicating the likely that shear rate increases after this stretching procedure. Additionally, Kruse et al. (2016) observed an increase in shear rate in the popliteal artery immediately after a single bout of a 4-min continuous passive calf stretch despite a reduction in shear rate during stretching. However, the stretching procedure in these previous studies was different from ours, which decreased arterial stiffness. For example, hemodynamics (e.g., heart rate, arterial blood pressure, and stroke volume), which affects arterial stiffness, was changed during stretching in these previous studies but not in our previous study. These hemodynamic responses also increase the shear rate (Green et al. 2017; Carter et al. 2017). Therefore, it remains unknown whether our stretching procedure, which decreases arterial stiffness without a change in central hemodynamics, increases the shear rate.

Given this background, we hypothesized that acute passive calf static stretching partially increases the shear rate in the peripheral artery of the stretched leg, and consequently, it may cause a reduction in arterial stiffness via local vasodilation. In the present study, to test this hypothesis, we need to exclude the effect of exercise-induced hemodynamics on vasculature. Thus, we measured mean blood velocity (V_{mean}), blood flow, and shear rate in the posterior tibial artery of the stretched and non-stretched legs in healthy young men before, during, and after one-legged passive intermittent calf static stretching previously described.

Methods

Participants

Nine healthy young men (age 20.4 ± 0.2 years; height 170.4 ± 1.7 cm; weight 63.5 ± 2.7 kg, body mass index 21.8 ± 0.7 kg/m^2; sit-and-reach test 40.2 ± 2.8 cm) were recruited for voluntary participation in the study. All participants had no habitual exercise (e.g., aerobic and/or resistance training), and were non-smokers, free from known cardiovascular disease, and not using prescription or over-the-counter medications. All volunteers provided written informed consent before participating in the study, which was approved by the Ethics Committee of Aino University and conducted in accordance with the Declaration of Helsinki.

Experimental design

Participants were advised not to participate in any strenuous physical activity and avoid alcohol or caffeine consumption for 24 h before any of the scheduled experiments. Participants were also instructed not to eat or drink fluids other than water for at least 6 h prior to the experiments. On the day of the experiments, the participant's height, body weight, and sit-and-reach measurements were obtained (T.K.K.5112; Takeikiki, Tokyo, Japan). Posterior tibial artery diameter, V_{mean}, and heart rate were measured in the supine position after at least 30 min of rest. Participants were subjected to passive calf intermittent static stretching on a randomly selected leg by one researcher. The posterior tibial artery diameter, V_{mean}, and heart rate were measured in the supine position during stretching, relaxation, and 1, 5, and 10 min after stretching. Diameter, V_{mean}, and heart rate were measured for the last 5 s of each point, and 3-beat (approximately 2–3 s) averages were used. The room temperature was maintained at 24 °C throughout the experiment.

Stretching protocol

The participants underwent a single session of passive calf intermittent static stretching on one leg as previously described (Yamato et al. 2017). In each participant, the stretched and non-stretched (control) legs were randomly selected from the right or left leg. With each participant placed in the supine position, the researcher passively moved the ankle joint of the stretched leg slowly (for at least 5 s) through the full range of motion. In contrast, the non-stretched leg was not moved. The stretches were passively held for 30 s at the end range (point of minimal discomfort), followed by a 10-s relaxation period for six repetitions.

Measurement of peripheral hemodynamics and heart rate

The measurements of posterior tibial artery diameter and V_{mean} were performed in the passively stretched leg and non-stretched legs, behind the malleolus medialis of the tibia using two LOGIQ e ultrasound system (GE Healthcare, Tokyo, Japan). Each LOGIQ e ultrasound system was equipped with a linear array transducer operating at an imaging frequency of 10 MHz. The posterior tibial artery diameter was determined at a perpendicular angle along

the central axis of the scanned area using B-mode imaging. The blood velocity was measured using the PW mode of the same probe at a frequency of 4.2 MHz. All blood velocity measurements were obtained with the probe appropriately positioned to maintain an insonation angle ≤ 60°. The sample volume was maximized according to the vessel size and centered within the vessel based on real-time ultrasound visualization. The arterial diameter was measured, and V_{mean} values was automatically calculated using available software (LOGIQ e). Diameter and V_{mean} were used to calculate the posterior tibial artery blood flow and shear rate. Blood flow was calculated as $V_{mean}\pi$ (vessel diameter/2)$^2 \times 60$ (Kruse et al. 2016; Wray et al. 2006), where blood flow is in milliliters per minute, and shear rate (per second), a useful estimator of shear stress that does not account for blood viscosity, was defined as $[4 \times V_{mean}]$/vessel diameter (Kruse et al. 2016; Wray et al. 2006). Heart rate was automatically calculated using the LOGIQ e software. In this study, the coefficient of variation for interobserver reproducibility of arterial diameter and V_{mean} were 2.1% and 9.4%, respectively.

Statistical analysis

Values are expressed as mean ± standard error (SE). A two-way (leg × time) repeated-measures analysis of variance (ANOVA) was performed on the posterior tibial artery diameter, V_{mean}, blood flow, and shear rate. One-way repeated-measures ANOVA was performed on the heart rate data. Fisher's post hoc test was used where significant values were found. Simple regression analysis was used to assess the associations between the following: percentage change in arterial diameter from baseline or 1–4 sets of relaxation period to stretching period and percentage change in blood flow from 1 to 5 sets of stretching period to relaxation period. We used the data from our previous study (Yamato et al. 2017) to calculate the effect size, which in turn was used to determine the sample size needed for this study. To detect this effect size at 80% power and with a two-tailed α of 5%, the sample size should set eight participants using G*Power. Thus, nine participants were recruited to ensure a statistical power > 0.80. A P-value < 0.05 was considered statistically significant. All statistical analyses were performed using StatView 5.0 (SAS Institute, Tokyo, Japan).

Results

Posterior tibial artery diameter, V_{mean}, blood flow, and shear rate in passive stretching

Figure 1 shows the representative images of both B-mode and ultrasound dopplers for the posterior tibial artery of the stretched leg during baseline, stretching, and relaxation. Baseline diameter, V_{mean}, blood flow, and shear rate of the posterior tibial artery did not differ between the stretched and non-stretched legs (Figs. 2, 3, Table 1). There were significant interactions between leg and time, and main effect of time on the posterior tibial artery diameter, V_{mean}, blood flow, and shear rate ($P < 0.0001$, respectively). In contrast, there were no significant differences in the main effect of leg. Posterior tibial artery diameter in the stretched leg was significantly decreased during each passive stretching period (Fig. 2a, $P < 0.05$) but not during each relaxation period and after the stretching procedure compared with that at baseline (Table 1). In contrast, the posterior tibial artery diameter in the non-stretched leg was not significantly altered at any time point (Fig. 2a, Table 1). Posterior tibial artery V_{mean}, blood flow, and shear rate in the stretched leg were significantly increased during each relaxation period compared with those at baseline (Figs. 2b, c, 3, $P < 0.05$). Additionally, the posterior tibial artery blood flow was significantly increased but not blood velocity and shear rate 1 min after the session of passive stretching compared with that at baseline (Table 1, $P < 0.05$). In contrast, the posterior tibial artery V_{mean}, blood flow, and shear rate in the non-stretched leg were not significantly altered at any time point (Figs. 2b, c, 3 and Table 1).

The percentage change in arterial diameter at the stretching period was significantly negatively correlated with blood flow during the relaxation period in the stretched leg ($r = -0.38$, $P < 0.05$; Fig. 4a). In contrast, in the non-stretched leg, there was no significant relationship

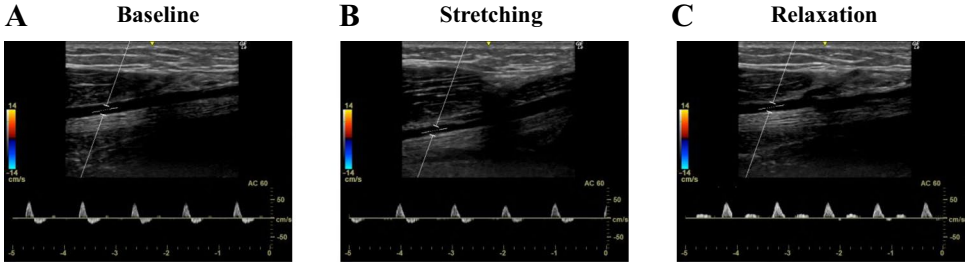

Fig. 1 Representative images of B-mode and ultrasound dopplers for the posterior tibial artery of the stretched leg at baseline (**a**), during stretching (**b**), and relaxation (**c**)

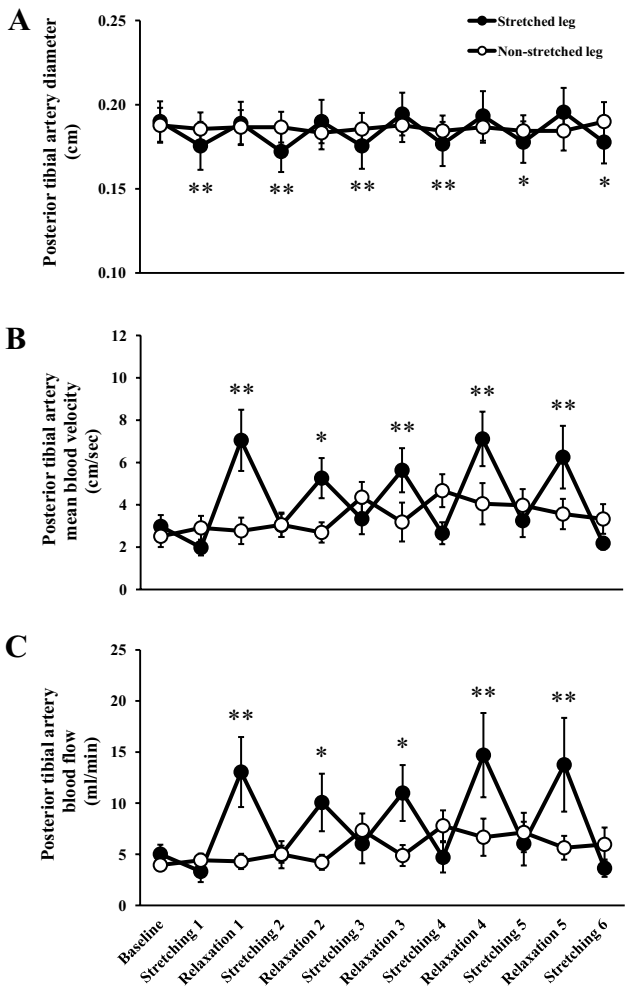

Fig. 2 Vessel diameter (**a**), mean blood velocity (**b**), and blood flow (**c**) of the posterior tibial artery before and during passive one-legged stretching. Data are expressed as the mean ± SE. *$P < 0.05$, **$P < 0.01$ vs. baseline

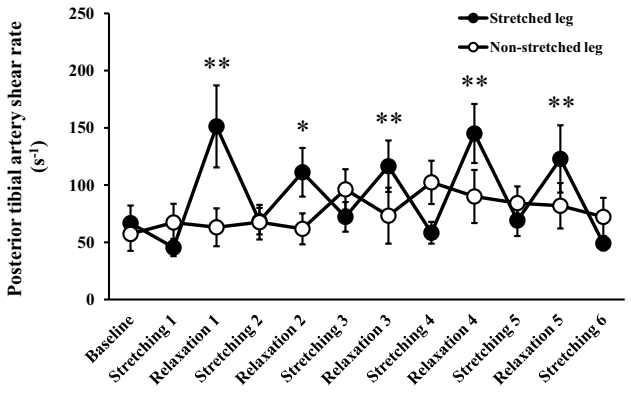

Fig. 3 Shear rate of the posterior tibial artery before and during passive one-legged stretching. Data are expressed as mean ± SE. *$P < 0.05$, **$P < 0.01$ vs. baseline

between these changes in arterial diameter and blood flow (Fig. 4b).

Heart rate during passive static stretching

The heart rate was not significantly altered at any time point (Fig. 5 and Table 1).

Discussion

The present study aimed to investigate the effect of acute passive calf intermittent static stretching on the shear rate in the peripheral artery of the stretched leg. Similar to our previous study (Yamato et al. 2017), hemodynamics, such as heart rate and blood pressure, did not change during and/or after passive calf intermittent static stretching. In the posterior tibial artery of the stretched leg, the shear rate was unchanged during stretching despite a reduction in the diameter, while it increased during the relaxation period because of the increase in blood velocity, indicating a likely increase in the total shear rate during this stretching procedure. These findings suggest that the stretching procedure used in this study increases the shear rate without central hemodynamic effects.

Our previous study demonstrated that acute passive one-legged stretching induced a reduction in peripheral arterial stiffness in the stretched leg (Yamato et al. 2017), while physiological mechanism remains unclear. The findings of the present study demonstrated that the total shear rate likely increases during stretching; thus, this result provides one possible mechanism that stretching-induced local hemodynamic response (e.g., activation of endothelial function) may contribute to the reduction in arterial stiffness in the stretched leg. Importantly, this stretching procedure did not change the hemodynamics, such as heart rate, which causes an increase in the shear rate. Therefore, it is reasonable that mechanical stress on the artery in the stretched leg, not hemodynamic effects, increases the shear rate and consequently causes activation of endothelial function that contributes to reduced arterial stiffness.

It is well-established that increased shear stress acutely causes the release of vasodilatory substances, such as NO, from the endothelial cell via activation of endothelial NO synthase (Higashi and Yoshizumi 2004; Zhou et al. 2014; Schmitt et al. 2005; Corson et al. 1996). Indeed, in human studies, flow-mediated dilation following ischemic cuff occlusion that increases the shear rate and consequently NO production is widely used as an index of endothelial function (Green et al. 2014; Mullen et al. 2001; Nishiyama et al. 2008; Pyke et al. 2004; Kooijman et al. 2008). Moreover, in vitro studies have shown that pulsatile flow only and pulsatile flow and tube compression conditions caused a

Table 1 Posterior tibial artery vascular measures at baseline and after stretching

	Baseline	After stretching		
		1 min	5 min	10 min
Diameter, cm				
Stretched leg	0.19 ± 0.01	0.20 ± 0.02	0.19 ± 0.01	0.19 ± 0.01
Non-stretched leg	0.19 ± 0.01	0.19 ± 0.01	0.19 ± 0.01	0.19 ± 0.01
Mean blood velocity, cm/s				
Stretched leg	2.98 ± 0.54	4.61 ± 1.18	3.82 ± 0.92	3.52 ± 0.54
Non-stretched leg	2.51 ± 0.51	3.20 ± 0.76	2.90 ± 0.56	2.83 ± 0.60
Blood flow, ml/min				
Stretched leg	5.01 ± 0.92	9.99 ± 3.86 *	6.93 ± 1.68	6.29 ± 1.18
Non-stretched leg	3.95 ± 0.58	5.79 ± 1.98	4.60 ± 0.74	4.62 ± 0.92
Shear rate, s^{-1}				
Stretched leg	66.75 ± 15.39	92.50 ± 19.69	82.08 ± 22.74	75.89 ± 14.36
Non-stretched leg	57.28 ± 14.78	67.69 ± 13.63	65.68 ± 15.93	63.05 ± 15.53
Heart rate, beats/min	60 ± 3	59 ± 3	60 ± 3	60 ± 3

Values are mean ± SE

*$P < 0.05$ vs. baseline

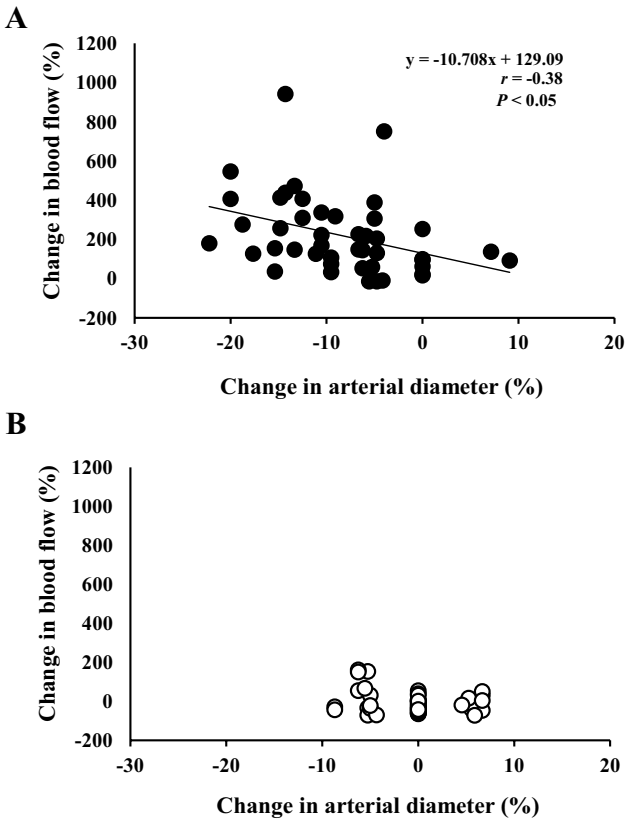

Fig. 4 Relationship between the changes (%) in posterior tibial artery diameter during the stretching period and blood flow during the relaxation period in the stretched leg (**a**) and non-stretched leg (**b**)

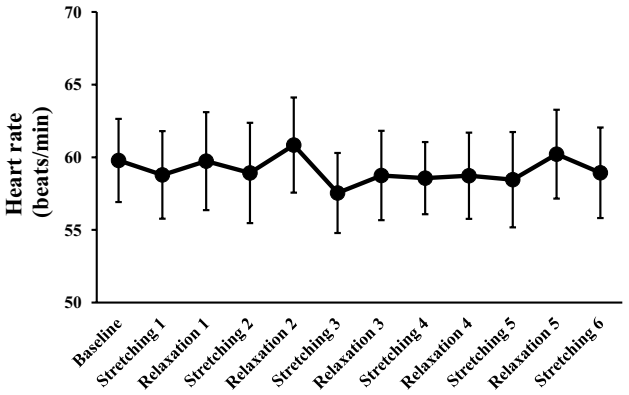

Fig. 5 Heart rate before and during passive one-legged stretching. Data are expressed as mean ± SE

rapid release of NO followed by a sustained increase (Dai et al. 2002). Thus, in the present study, the increase in shear rate as a result of acute passive intermittent static stretching may affect NO production in endothelial cells. Moreover, some previous studies have demonstrated that endothelial dysfunction contributes to arterial stiffening (Kinlay et al. 2001; Wilkinson et al. 2002). Taken together, acute stretching-induced reduction in arterial stiffness may be associated with activation of endothelial function via an increase in shear rate in the peripheral artery.

The mechanism of stretching-induced increase in shear rate should be considered. Similar to the previous studies (Venturelli et al. 2019; Kruse et al. 2016), in the present study, the V_{mean} of the posterior tibial artery in the stretched leg decreased during the stretching period but significantly increased during the relaxation period (Fig. 2). This large increase in blood velocity contributes to an increase in the shear rate because there is no change in diameter during the relaxation period. Additionally, heart rate and blood pressure did not change during and/or after passive calf intermittent

static stretching protocol used previous study (Yamato et al. 2017). Thus, this increase in blood velocity may be associated with mechanical microvascular compression in the muscle caused by stretching. This concept is supported by previous studies (Venturelli et al. 2019). Intermittent pneumatic leg compression increased blood flow in the treated limb during cuff deflation, whereas cuff inflation has an opposite effect (Sheldon et al. 2012; Roseguini et al. 2010). Moreover, in animal studies, as the length of the muscle increased, the diameters of the arterioles, feed arteries, and capillary arteries decreased, with decreased blood flow of feed arteries and capillary arteries (Poole et al. 1997; Welsh and Segal 1996). These findings suggest that the loss of microvascular blood volume and decrease in arterial diameter during each stretching period may induce reactive hyperemia in the relaxation period after each stretching period. Indeed, in the present study, an increase in blood flow during the relaxation period was significantly associated with a decrease in diameter during the stretching period ($r = -0.38$, $P < 0.05$; Fig. 4a). Moreover, in the stretching protocol used in the present study, the heart rate remained unchanged throughout the stretching procedure; therefore, it is likely that stretching-induced muscle compression, and not change in central hemodynamics, mechanically elevates V_{mean} during the relaxation period and consequently increases the total shear rate in the stretched leg during the stretching protocol.

This study has some limitations. First, although the present study used the stretching procedure similar to our previous study (Yamato et al. 2017), we did not measure the arterial stiffness index in this study. Therefore, the effect of arterial stiffness in this study may be the same, since the stretch procedure was repeatable. Second, we did not measure NO production in the posterior tibial artery of the stretched leg. Further studies are needed to examine the circulating NO level in the stretched leg. Third, in this study, we did not evaluate several other factors, including central and peripheral hemodynamics (e.g., cardiac output, sympathetic nerve activity, speed of passive movement). Stroke volume, heart rate, and cardiac output measurements using ultrasonography may be necessary to obtain an accurate assessment of the central hemodynamic response. Moreover, a previous animal study showed that muscle length-induced vasomotor responses, which were triggered by norepinephrine release from the perivascular sympathetic nerves, were independent of the central nervous system (Welsh and Segal 1996), while a previous human study evaluated the effects of the movement speed and range of motion associated with leg blood flow on passive leg movement-induced hyperemia (Gifford et al. 2019). Therefore, local hemodynamic responses may be affected by perivascular sympathetic nerve activity via increased peripheral muscle length, and movement speed and range of motion may also have some impact on passive leg movement. Finally, we could not assess the continuous shear rate throughout the stretching procedure in this study. Since we passively moved the ankle joint of the stretched leg slowly (using at least 5 s) through the full range of motion, we measured blood flow after the completely stretched and returning base position. Therefore, beat-to-beat blood velocity was not measured during and after the stretching period. Therefore, to examine the total shear rate, it is necessary to continuously measure the blood velocity.

Thus, passive calf intermittent static stretching mechanically reduced arterial diameter during stretching and consequently increased the shear rate during the relaxation period in the artery of the stretched leg. This finding suggests that the mechanical effect of acute passive stretching on artery (decrease in diameter)-induced increase in shear rate may be associated with the reduction in arterial stiffness after the stretching.

Acknowledgements This work was supported by Grants-in-Aid for Scientific Research from the Ministry of Education, Culture, Sports, Science and Technology of Japan (KAKENHI: 17H02182 and 19K22828 for M. Iemitsu).

Author contributions MI, SO and YY conceived and designed research. YY, YH, SF, NH, N. Horii, HA, YY and MI conducted experiments. YY, SO and MI analyzed data. MI, SO, YY, YH, SF, and NH wrote the manuscript. All authors read and approved the manuscript.

Compliance with ethical standards

Conflict of interest The authors declare no conflicts of interest.

Ethical approval This study was approved by the Ethics Committee of Aino University and conducted in accordance with the Declaration of Helsinki.

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

References

Carter HH, Spence AL, Ainslie PN, Pugh CJA, Naylor LH, Green GJ (2017) Differential impact of water immersion on arterial blood flow and shear stress in the carotid and brachial arteries of humans. Physiol Rep 5:e13285. https://doi.org/10.14814/phy2.13285

Corson MA, James NL, Latta SE, Nerem RM, Berk BC, Harrison DG (1996) Phosphorylation of endothelial nitric oxide synthase in response to fluid shear stress. Circ Res 79:984–991. https://doi.org/10.1161/01.res.79.5.984

Dai G, Tsukurov O, Chen M, Gertler JP, Kamm RD (2002) Endothelial nitric oxide production during in vitro simulation of external limb compression. Am J Physiol Heart Circ Physiol 282:H2066–H2075. https://doi.org/10.1152/ajpheart.00288.2001

Gifford JR, Bloomfield T, Davis T, Addington A, McMullin E, Wallace T, Proffit M, Hanson B (2019) The effect of the speed and range of motion of movement on the hyperemic response to passive leg movement. Physiol Rep 7:e14064. https://doi.org/10.14814/phy2.14064

Green DJ, Dawson EA, Groenewoud HM, Jones H, Thijssen DH (2014) Is flow-mediated dilation nitric oxide mediated? A meta-analysis. Hypertension 63:376–382. https://doi.org/10.1161/HYPERTENSIONAHA.113.02044

Green DJ, Hopman MTE, Padilla J, Laughlin MH, Thijssen DHJ (2017) Vascular adaptation to exercise in humans: role of hemodynamic stimuli. Physiol Rev 97:495–528. https://doi.org/10.1152/physrev.00014.2016

Higashi Y, Yoshizumi M (2004) Exercise and endothelial function: role of endothelium-derived nitric oxide and oxidative stress in healthy subjects and hypertensive patients. Pharmacol Ther 102:87–96. https://doi.org/10.1016/j.pharmthera.2004.02.003

Kinlay S, Creager MA, Fukumoto M, Hikita H, Fang JC, Selwyn AP, Ganz P (2001) Endothelium-derived nitric oxide regulates arterial elasticity in human arteries in vivo. Hypertension 38:1049–1053. https://doi.org/10.1161/hy1101.095329

Kooijman M, Thijssen DH, de Groot PC, Bleeker MW, van Kuppevelt HJ, Green DJ, Rongen GA, Smits P, Hopman MT (2008) Flow-mediated dilatation in the superficial femoral artery is nitric oxide mediated in humans. J Physiol 586:1137–1145. https://doi.org/10.1113/jphysiol.2007.145722

Kruse NT, Silette CR, Scheuermann BW (2016) Influence of passive stretch on muscle blood flow, oxygenation and central cardiovascular responses in healthy young males. Am J Physiol Heart Circ Physiol 310:H1210–H1221. https://doi.org/10.1152/ajpheart.00732.2015

Mullen MJ, Kharbanda RK, Cross J, Donald AE, Taylor M, Vallance P, Deanfield JE, MacAllister RJ (2001) Heterogenous nature of flow-mediated dilatation in human conduit arteries in vivo: relevance to endothelial dysfunction in hypercholesterolemia. Circ Res 88:145–151. https://doi.org/10.1161/01.res.88.2.145

Nishiwaki M, Kurobe K, Kiuchi A, Nakamura T, Matsumoto N (2014) Sex differences in flexibility-arterial stiffness relationship and its application for diagnosis of arterial stiffening: a cross-sectional observational study. PLoS ONE 9:e113646. https://doi.org/10.1371/journal.pone.0113646

Nishiwaki M, Yonemura H, Kurobe K, Matsumoto N (2015) Four weeks of regular static stretching reduces arterial stiffness in middle-aged men. Springerplus 4:555. https://doi.org/10.1186/s40064-015-1337-4**(eCollection 2015)**

Nishiyama SK, Wray DW, Richardson RS (2008) Aging affects vascular structure and function in a limb-specific manner. J Appl Physiol 105:1661–1670. https://doi.org/10.1152/japplphysiol.90612.2008

Poole DC, Musch TI, Kindig CA (1997) In vivo microvascular structural and functional consequences of muscle length changes. Am J Physiol 272:H2107–H2114. https://doi.org/10.1152/ajpheart.1997.272.5.H2107

Pyke KE, Dwyer EM, Tschakovsky ME (2004) Impact of controlling shear rate on flow-mediated dilation responses in the brachial artery of humans. J Appl Physiol 97:499–508. https://doi.org/10.1152/japplphysiol.01245.2003

Roseguini BT, Mehmet Soylu S, Whyte JJ, Yang HT, Newcomer S, Laughlin MH (2010) Intermittent pneumatic leg compressions acutely upregulate VEGF and MCP-1 expression in skeletal muscle. Am J Physiol Heart Circ Physiol 298:H1991–H2000. https://doi.org/10.1152/ajpheart.00006.2010

Schmitt M, Avolio A, Qasem A, McEniery CM, Butlin M, Wilkinson IB, Cockcroft JR (2005) Basal NO locally modulates human iliac artery function in vivo. Hypertension 46:227–231. https://doi.org/10.1161/01.HYP.0000164581.39811.bd

Sheldon RD, Roseguini BT, Thyfault JP, Crist BD, Laughlin MH, Newcomer SC (2012) Acute impact of intermittent pneumatic leg compression frequency on limb hemodynamics, vascular function, and skeletal muscle gene expression in humans. J Appl Physiol 112:2099–2109. https://doi.org/10.1152/japplphysiol.00042.2012

Shinno H, Kurose S, Yamanaka Y, Higurashi K, Fukushima Y, Tsutsumi H, Kimura Y (2017) Evaluation of a static stretching intervention on vascular endothelial function and arterial stiffness. Eur J Sport Sci 17:586–592. https://doi.org/10.1080/17461391.2017.1284267

Venturelli M, Rampichini S, Coratella G, Limonta E, Bisconti AV, Cè E, Esposito F (2019) Heart and musculoskeletal hemodynamic responses to repetitive bouts of quadriceps static stretching. J Appl Physiol 127:376–384. https://doi.org/10.1152/japplphysiol.00823.2018

Welsh DG, Segal SS (1996) Muscle length directs sympathetic nerve activity and vasomotor tone in resistance vessels of hamster retractor. Circ Res 79:551–559. https://doi.org/10.1161/01.res.79.3.551

Wilkinson IB, Qasem A, McEniery CM, Webb DJ, Avolio AP, Cockcroft JR (2002) Nitric oxide regulates local arterial distensibility in vivo. Circulation 105:213–217. https://doi.org/10.1161/hc0202.101970

Wray DW, Uberoi A, Lawrenson L, Richardson RS (2006) Evidence of preserved endothelial function and vascular plasticity with age. Am J Physiol Heart Circ Physiol 290:H1271–H1277. https://doi.org/10.1152/ajpheart.00883.2005

Yamamoto K, Kawano H, Gando Y, Iemitsu M, Murakami H, Sanada K, Tanimoto M, Ohmori Y, Higuchi M, Tabata I, Miyachi M (2009) Poor trunk flexibility is associated with arterial stiffening. Am J Physiol Heart Circ Physiol 297:H1314–H1318. https://doi.org/10.1152/ajpheart.00061.2009

Yamato Y, Hasegawa N, Fujie S, Ogoh S, Iemitsu M (2017) Acute effect of stretching one leg on regional arterial stiffness in young men. Eur J Appl Physiol 117:1227–1232. https://doi.org/10.1007/s00421-017-3609-x

Zhou J, Li YS, Chien S (2014) Shear stress-initiated signaling and its regulation of endothelial function. Arterioscler Thromb Vasc Biol 34:2191–2198. https://doi.org/10.1161/ATVBAHA.114.303422