International Journal of Gerontology 16 (2022) 271–276
https://doi.org/10.6890/IJGE.202207_16(3).0020

International Journal of Gerontology

journal homepage: http://www.sgecm.org.tw/ijge/

Original Article

# Relationship between Vascular Structure and Function and Thigh Muscle Composition in Normal-Weight Middle-Aged and Older Men

Madoka Ogawa [a,b *], Yuto Hashimoto [c], Naoki Kikuchi [b,c], Takanobu Okamoto [b,c]

[a] *Faculty of Sociology, Kyoto Sangyo University, Kyoto, Japan,* [b] *Faculty of Sport Science, Nippon Sport Science University, Tokyo, Japan,* [c] *Graduate School of Health and Sport Science, Nippon Sport Science University, Tokyo, Japan*

ARTICLE INFO

Accepted 16 December 2021

*Keywords:*
augmentation index,
central systolic blood pressure,
intima-media thickness,
intramuscular fat,
muscle size

SUMMARY

*Background:* Muscle atrophy and increased intramuscular fat (IMF) content in individuals with obesity have been associated with increased cardiovascular disease risk but unknown in normal-weight individuals. We aimed to investigate the relationships between the vascular structure and function and body and thigh compositions among normal-weight men aged 50–79 years.
*Methods:* A total of 45 normal-weight middle-aged and older men were included herein and subsequently measured for mean carotid intima-media thickness (IMT) in near and far walls, central systolic blood pressure, augmentation index at a heart rate of 75 bpm (AIx@75), mid-thigh muscle cross-sectional area (CSA) and IMF contents using magnetic resonance imaging, and body fat percentage via electrical impedance analysis.
*Results:* No difference in body or muscle composition was observed among the age groups. Near wall IMT was significantly positively correlated with IMF content among those in their 70s but not among those in their 50s and 60s. On the other hand, AIx@75 was strongly inversely correlated with muscle CSA per body weight, and positively correlated with body fat percentage among those in their 60s.
*Conclusions:* The findings presented herein suggested that muscle atrophy and increased body fat percentage could affect age-specific changes in vascular structure and function among normal-weight men in their 60s.



## 1. Introduction

Adiposity, especially abdominal obesity, has been a well-known risk factor for the occurrence of cardiovascular disease (CVD) and mortality among the general population.[1] Nonetheless, individuals with less visceral fat and normal body mass index (BMI) may still be at increased risk for CVD through "metabolic obesity" and increased cardio metabolic risk.[2] However, the risk factors for CVD in normal-weight individuals have been poorly documented.

Apart from the well described changes in the neural drive to muscles, fiber type composition, muscle architecture and single-fiber specific tension,[3] aging may cause alterations in muscle composition due to increased infiltration of non-contractile tissues, such as adipose tissue.[4] Excess adipose tissue around the muscle (e.g., intramuscular fat; IMF) could be associated with impaired muscle blood flow, reduced insulin diffusion, and increased fatty acid synthesis via altered lipolysis, which may explain its association with metabolic abnormalities and CVD risk.[5] As such, the aforementioned findings suggest that increased IMF content among normal-weight individuals could increase CVD risk.

Arterial stiffness, an independent risk factor for CVD, has been found to occur with both structural and functional changes in the blood vessel.[6] Similarly, carotid intima-media thickness (IMT), a measure of early atherosclerosis, has been identified as a predictive marker for direct CVD that increases with age, beginning from healthy young individuals.[7] Carotid IMT is measured on the wall nearest to (near wall) and farthest from (far wall) the ultrasound transducer. A previous study reported that far wall IMT showed a stronger association with the incidence of coronary heart disease than near wall IMT.[8] Furthermore, Lee et al. (2015) reported that mean carotid IMT in young overweight and obese individuals was associated with the IMF index determined using thigh muscle attenuation via computer tomography (CT).[9] Their findings also showed that IMT could be associated with IMF content in normal-weight individuals. However, it is unknown whether both near and far walls IMT are associated with IMF accumulation.

On the other hand, central systolic blood pressure (cSBP) might serve as a powerful predictor for the development of hypertension, target organ damage, and cardiovascular events.[10] Moreover, the augmentation index (AIx), an index of arterial stiffness quantifying the reflected wave at the central artery, provides a more accurate estimate of the left ventricle afterload and is thus more strongly related to adverse cardiovascular events compared to brachial BP.[11] A previous study reported that the accumulation of IMF is associated with an increase in IMT,[9] that is the deterioration of vascular morphology, but the relationship between vascular function, such as cSBP and AIx, and IMF content are unknown. Previous studies have reported that vascular dysfunction was related to muscle size in both younger and older individuals;[3,12] thus, muscle composition could

* Corresponding author. Kyoto Sangyo University, Motoyama, Kamigamo, Kita-ku, Kyoto, 603-8555, Japan.
*E-mail address:* madoka.ogawa20@gmail.com (M. Ogawa)

also be associated with vascular function.

The present study therefore aimed to investigate the relationship between vascular structure and function and body and thigh muscle composition among normal-weight men aged 50–79 years. We hypothesized that the mean carotid near wall IMT would be associated with body fat and IMF contents of thigh muscle groups. Furthermore, we hypothesized that vascular function, such as cSBP and AIx, would be related to muscle size.

## 2. Patients and methods

### 2.1. Subjects

Recruited subjects were healthy and normal-weight (BMI < 25.0 kg/m$^2$) volunteers. This prospective study was conducted from 2019 to 2020. Of 63 middle-aged and older men who applied for this study, 11 who were currently taking antihyperlipidemic, antihypertensive, or antihyperglycemic medications or having metal in the body, and 7 overweight men (BMI ≥ 25.0 kg/m$^2$) were excluded. Table 1 summarizes the physical characteristics of the included subjects based on age group (e.g., 50s, 60s, and 70s). Before study commencement, subjects were provided an explanation regarding the procedures, purposes, risks, and benefits associated with the study, after which written consent was obtained. The present study was approved by the ethics committee of Nippon Sport Science University (018-H194) and was performed in accordance with the Declaration of Helsinki.

### 2.2. Assessment of IMT

IMT was evaluated using high-resolution B-mode ultrasonography (Vivid T8, GE Healthcare, Chicago, IL, USA) with a 12-MHz transducer. With patients in the sitting position, carotid IMT was measured 1 cm proximal to the carotid bulb dilatation, with far wall measurements being obtained using an IMT measurement software (IMT measurement system ver.1.0, Takei Scientific Instruments, Niigata, Japan). The mean IMT of near and far wall measurements were utilized for analysis.

### 2.3. Central and brachial blood pressures and the augmentation index

Brachial and aortic blood pressures measurements were obtained with participants placed in a seated position following 5 min of rest. Arterial pulse waveforms of the left radial artery to estimate the cSBP and AIx were measured noninvasively using an automated tonometric system, as previously described.[13] Brachial systolic (bSBP) and diastolic blood pressures (bDBP) were measured on the right arm using an oscillometric manometer (HEM-9000AI, Omron Healthcare Co., Ltd, Tokyo, Japan). Radial arterial pressure wave signals were low-pass-filtered initially at a cut-off frequency of 105 Hz to remove high-frequency noise and then at 25 Hz to extract pressure waveforms. Briefly, the central and brachial blood pressures were obtained, with measurements being repeated when the first two blood pressures were not within 5 mmHg. The average of the two closest readings was used for subsequent analysis. The AIx was defined as the augmented pressure divided by aortic pulse pressure and expressed as a percentage. Heart rate (HR) was defined as the duration between waveforms. AIx values were expressed relative to a HR of 75 bpm (AIx@75). All measurements were taken by one investigator (Y.H.).

### 2.4. Magnetic resonance imaging acquisition

Subjects were assessed using a 1.5 T whole-body magnetic resonance imaging scanner (ECHELON OVAL, Hitachi, Tokyo, Japan). After placing subjects in the supine position, thigh images were acquired using a torso coil. Mid-thigh was defined according to markers attached at the middle point between the greater trochanter and the lateral condyle of the femur. Two-point Dixon transaxial images of the right thigh were obtained using the following sequence parameters: three-dimensional, repetition time = 13.3 ms; echo times = 6.7 and 9.2 ms; flip angle = 60°; optimized field of view = 256 × 256 mm; slice thickness = 5 mm; and interslice gap = 0 mm. All subjects were instructed to remain as motionless as possible.

### 2.5. Thigh composition analysis

Dixon images were analyzed using ImageJ (version 1.44; National Institutes of Health, Bethesda, MD). Thereafter, we measured the muscle cross-sectional area (CSA) of total thigh at the mid-thigh. Serial axial images were used to identify muscle boundaries. Mean signal intensities from water and fat images were measured to create a Dixon-based fat-water ratio using the following equation:[14]

IMF content (%) = 100 × Fat mean intensity / (Water mean intensity + Fat mean intensity)

All images were analyzed in random order by one investigator (M.O.). Test–retest reliability of IMF content has been reported elsewhere.[15] Briefly, intraclass correlation coefficients [ICC (2, 1)] in individual mid-thigh muscles for 10 subjects showed values ranging between 0.97 and 1.00 (all $p < 0.001$).

### 2.6. Body composition assessment

Height and weight were measured using standardized protocols. The BMI was calculated as weight divided by height squared (kg/m$^2$). Body fat percentage was determined using bioelectrical impedance measurements (InBody, Biospace Co. Ltd., Seoul, Korea) with hand grip and foot plate electrodes.

**Table 1**
Subject characteristics.

| | 50s (N = 12) | 60s (N = 18) | 70s (N = 15) | *p* |
|---|---|---|---|---|
| Age (years) | 54.0 (52.0–55.0) | 67.0 (64.5–68.0)[a] | 73.0 (70.0–76.0)[a,b] | < 0.001 |
| Height (m) | 1.7 (1.7–1.8) | 1.7 (1.6–1.7) | 1.7 (1.6–1.7) | 0.005 |
| Weight (kg) | 67.0 (62.6–72.6) | 64.5 (56.0–67.3) | 66.0 (54.0–71.0) | 0.439 |
| BMI (kg/m$^2$) | 22.4 (20.6–24.3) | 22.8 (21.5–24.4) | 22.4 (21.6–24.1) | 0.818 |
| Body fat (%) | 18.7 (16.3–22.3) | 20.5 (14.8–22.3) | 21.0 (20.0–25.0) | 0.197 |
| IMF content (%) | 14.7 (9.1–17.8) | 11.5 (8.8–16.7) | 13.4 (9.4–18.8) | 0.743 |
| Muscle CSA/bw (cm$^2$/kg) | 1.9 (1.8–2.2) | 2.0 (1.8–2.1) | 1.9 (1.7–2.0) | 0.456 |

Medium (25th to 75th percentile), [a] $p < 0.01$ vs. 50s, [b] $p < 0.01$ vs. 60s, BMI: body mass index, CSA: cross-sectional area, IMF: intramuscular fat.

### 2.7. Statistical analysis

All values are reported as the median and 25th to 75th percentile. The Kruskal-Wallis test was used to compare variables according to age group (i.e., 50s, 60s, and 70s). The relationship between vascular structure and function and body fat percentage and muscle composition were determined using Spearman's rank correlation. All statistical analyses were performed using IBM SPSS Statistics (version 24.0, IBM Japan, Tokyo, Japan), with $p < 0.05$ indicating statistical significance.

## 3. Results

Table 1 summarizes the physical characteristics of the subjects. No significant difference in physical characteristics was observed between the groups.

Table 2 summarizes the values of the vascular function and structure for 50s, 60s, and 70s. Subjects in their 60s and 70s had significantly higher mean near and far wall IMTs, cSBP and AIx@75 compared to those in their 50s. Meanwhile, no significant differences in all vascular function and structure measurements were observed between those in their 60s and 70s.

### 3.1. Relationships between near and far wall IMTs and body and muscle composition

Table 3 outlines the relationships between the IMTs and body and muscle composition according to age group. The mean near wall IMT was significantly positively correlated with body fat percentage among those in their 60s in Table 3. Moreover, mean near wall IMT was significantly positively correlated with IMF content among those in their 70s. On the other hand, mean near wall IMT was not significantly correlated with muscle CSA/bw in any of the age groups. Conversely, no significant correlations between mean far wall IMT and muscle CSA/bw or IMF content were observed among those in their 50s and 70s.

### 3.2. Relationships between the AIx@75 and body and muscle composition

Table 4 outlines the relationships between the AIx@75 and body and muscle composition according to age group. No significant correlations between AIx@75 and body fat percentage or IMF contents were observed among those in their 50s and 70s. Meanwhile, AIx@75 was significantly inversely correlated with muscle CSA/bw among those in their 60s. Furthermore, AIx@75 was significantly positively correlated with body fat percentage among those in their 60s.

### 3.3. Relationships between central and brachial blood pressures and body and muscle composition

Tables 5 and 6 outline the relationships between central and brachial blood pressures and body and muscle compositions based on age group. cSBP tended to be correlated with body fat percentage among those in their 50s and 60s. Furthermore, cSBP tended to be correlated with muscle CSA/bw among those in their 60s. On the other hand, no significant correlations between brachial blood pressures and body fat percentage, IMF contents and muscle CSA/bw was observed according to age groups (Table 6).

## 4. Discussion

The three main findings of this study are as follows. First,

**Table 4**
Correlation coefficients between augmentation index at a heart rate of 75 bpm (AIx@75) and body and muscle composition for the age groups 50s, 60s, and 70s.

| | 50s | 60s | 70s |
|---|---|---|---|
| Body fat (%) | 0.049 | 0.499[a] | - 0.002 |
| IMF content (%) | 0.063 | - 0.156 | 0.071 |
| Muscle CSA/bw ($cm^2$/kg) | 0.249 | - 0.739[b] | 0.107 |

[a] $p < 0.05$, [b] $p < 0.01$.
bw: body weight, CSA: cross-sectional area, IMF: intramuscular fat.

**Table 5**
Correlation coefficients between central systolic blood pressure (cSBP) and body and muscle compositions for the age groups 50s, 60s, and 70s.

| | 50s | 60s | 70s |
|---|---|---|---|
| Body fat (%) | 0.280 | 0.453 | 0.330 |
| IMF content (%) | - 0.056 | - 0.165 | 0.020 |
| Muscle CSA/bw ($cm^2$/kg) | - 0.070 | - 0.453 | 0.068 |

bw: body weight, CSA: cross-sectional area, IMF: intramuscular fat.

**Table 2**
Comparison of vascular function and structure for the age groups 50s, 60s, and 70s.

| | 50s | 60s | 70s | *p* |
|---|---|---|---|---|
| IMT (cm) | | | | |
| Near wall | 0.6 (0.6–0.7) | 0.7 (0.7–0.9)[a] | 0.7 (0.7–0.8)[a] | 0.005 |
| Far wall | 0.6 (0.6–0.7) | 0.7 (0.7–0.8)[a] | 0.8 (0.7–0.9)[b] | < 0.001 |
| cSBP (mmHg) | 120.6 (114.8–129.4) | 138.8 (126.4–148.4)[a] | 134.2 (125.0–151.2)[a] | 0.044 |
| bSBP(mmHg) | 120.0 (112.8–127.1) | 132.3 (118.7–140.7) | 126.3 (116.3–137.3) | 0.065 |
| bDBP (mmHg) | 80.0 (73.6–85.1) | 78.3 (74.1–86.1) | 75.0 (72.0–79.7) | 0.272 |
| AIx@75 (%) | 69.1 (50.5–76.1) | 77.6 (71.4–82.8)[a] | 82.1 (71.2–85.7)[b] | 0.020 |

Median (25th to 75th percentile), [a] $p < 0.05$; [b] $p < 0.01$ vs. 50s.
AIx@75: augmentation index at a heart rate of 75 bpm, bDBP: brachial diastolic blood pressure, bSBP: brachial systolic blood pressure, cSBP: central systolic blood pressure, IMT: mean intima-media thickness.

**Table 3**
Correlation between mean intima-media thickness (IMT) and body and muscle compositions for the age groups 50s, 60s, and 70s.

| | Near wall | | | Far wall | | |
|---|---|---|---|---|---|---|
| | 50s | 60s | 70s | 50s | 60s | 70s |
| Body fat (%) | 0.210 | 0.531[a] | - 0.235 | - 0.061 | 0.376 | 0.039 |
| IMF content (%) | - 0.299 | - 0.150 | 0.565[a] | 0.136 | 0.430 | 0.163 |
| Muscle CSA/bw ($cm^2$/kg) | 0.380 | - 0.072 | - 0.300 | - 0.192 | 0.335 | 0.018 |

[a] $p < 0.05$, bw: body weight, CSA: cross-sectional area, IMF: intramuscular fat.

**Table 6**
Correlation coefficients between brachial blood pressures and body and muscle composition for the age groups 50s, 60s, and 70s.

| | bSBP (mmHg) | | | bDBP (mmHg) | | |
|---|---|---|---|---|---|---|
| | 50s | 60s | 70s | 50s | 60s | 70s |
| Body fat (%) | 0.333 | 0.331 | 0.371 | 0.326 | 0.094 | 0.214 |
| IMF content (%) | 0.028 | 0.069 | - 0.007 | 0.125 | 0.281 | - 0.325 |
| Muscle CSA / bw ($cm^2$/kg) | - 0.123 | - 0.168 | 0.179 | - 0.181 | - 0.145 | 0.027 |

bDBP: brachial diastolic blood pressure, bSBP: brachial systolic blood pressure, bw: body weight, CSA: cross-sectional area, IMF: intramuscular fat.

AIx@75 was strongly inversely correlated with muscle CSA/bw among those in their 60s (Table 4). Second, mean near wall IMT was significantly positively correlated with IMF content among those in their 70s but not among those in their 50s and 60s (Table 3). Third, mean near wall IMT and AIx@75% were significantly positively correlated with body fat percentage among those in their 60s (Tables 3 and 4).

The present study focused on vascular structure and function among normal-weight individuals acceding to age group. Notably, no significant difference in body fat percentage, IMF content, and muscle size had been observed among those in their 50s, 60s, and 70s. However, those in their 60s and 70s had higher mean near and far walls IMT compared to those in their 50s. These results indicated that age considerably affected vascular structure and function in normal-weight individuals. On the other hand, although participants in their 60s and 70s had higher cSBP and AIx@75 compared to those in their 50s, bSBP and bDBP did not differ among the age groups (Table 2). The aforementioned results therefore indicate that changes in the aorta, such as increased cSBP and/or arterial stiffening, occur even when the peripheral blood pressure is normal. As such, cSBP may be a powerful predictor of the development of hypertension, target organ damage, and cardiovascular events.[10] Thus, our results suggest the importance of earlier testing for age-related changes in vascular condition through mean IMT, cSBP, and AIx@75 among normal-weight middle-aged and older individuals. Although aging causes linear changes in vascular function, details regarding age-related changes in vascular structure and function have differed depending on the age. Franklin et al. (1997) reported that after the age of 60 years, bDBP declined and pulse pressure increased steeply, which may have been influenced by large artery stiffness.[16] Meanwhile, the present study found that AIx@75, an index of large artery stiffness, was correlated with body fat percentage and muscle size among those in their 60s but not among those in their 50s and 70s (Table 4). Furthermore, our results showed that near wall IMT was correlated with body fat percentage among those in their 60s, but not among those in their 50s and 70s (Table 3). The aforementioned results suggest that body fat percentage and muscle size could be associated with the early changes in blood vessel occurring among participants in their 60s.

The present study showed that muscle CSA/bw had a significant negative correlation with AIx@75 among those in their 60s (Table 4). Several previous studies have reported that the arteriosclerosis index, such as cardio-ankle vascular index and pulse wave velocity and the AIx@75 were associated with muscle mass.[3] The findings presented herein support the aforementioned results. Accordingly, muscle size, an important factor for arteriosclerosis, may be associated with a larger vascular network (e.g., angiogenesis) due to greater muscle mass. Given that skeletal muscle contains more microvessels than any other organ, increasing muscle CSA proportionally increases the number of capillaries.[17] This larger vascular network promotes greater variance in the waveform and a smaller apparent magnitude of the reflected pressure wave.[18] Furthermore, a previous study had shown that muscle perfusion, an indicator of peripheral microvascular health, declined with age.[19] Reduced muscle perfusion could contribute to insulin resistance in the observed muscle with age,[20] eventually leading to muscle mass loss. However, it remains unknown whether the relationship between vascular dysfunction and muscle size is bidirectional, warranting further exploratory studies.

The present study showed that body fat percentage was significantly positively correlated with mean near wall IMT and AIx@75 among participants in their 60s (Tables 2 and 3). Such results are consistent with the findings presented in a previous study, which found body fat percentage to be strongly associated with mean IMT among middle-aged men (mean 53.5 $\pm$ 11.0 years) without CVD, including obesity.[21] As mentioned earlier, body fat percentage may promote atherosclerosis through the secretion of adipocytokines, such as necrosis factor alpha, by adipose tissue via the paracrine and endocrine pathways. On the other hand, the underlying mechanism explaining the relationship between body fat and vascular function has yet to be fully understood. One major mechanism may be related to leptin, a hormone secreted by adipose tissue that promotes sympathetic nerve activation across several organs, such as the kidneys and blood vessels. Notably, a previous study showed that approximately 50% of the association between blood pressure and body weight can be attributed to changes in leptin levels.[22] Although the mechanisms involved have yet to be fully understood, studies have suggested the involvement of increased sympathetic nerve activity.[23] A previous study involving acute infusion of leptin into rabbits confirmed that blood pressure was increased through sympathetic nerve stimulation.[24] Moreover, another study showed that serum leptin levels increased with body fat percentage in normal-weight and obese individuals.[25] The simultaneous increase in leptin with body fat percentage activates the sympathetic nervous system and increases arterial blood pressure even in normal-weight individuals. Therefore, an increase in body fat percentage could increase the risk of CVD among normal-weight middle-aged individuals.

Interestingly, the present study found a significant positive correlation between near wall IMT and IMF content among participants in their 70s (Table 3). The mean far wall IMT showed a tendency to be correlated with IMF content among those in their 60s (Table 3). A previous study reported that far wall IMT showed a stronger association with the incidence of coronary heart disease than near wall IMT,[8] and our hypothesis that there is a high correlation between IMF content and far wall IMT was partly supported. The reason is not clear in this study; however, this may be because the study was focused on healthy middle-aged and older subjects with a BMI of 25 or less. The aforementioned results were consistent with those presented in previous studies, which reported that carotid IMT was associated with the IMF index in the thigh evaluated using muscle attenuation via CT in young individuals with overweight and obesity.[9] IMF accumulation was associated with increased insulin resistance.[26] Moreover, evidence has shown that exposure of vascular cells to elevated insulin concentrations promotes proatherogenic effects. In cultured endothelial cells, elevated insulin concentrations

have been found to promote the secretion of endothelin-1, a potent vasoconstrictor protein, the expression of vascular cell adhesion molecule-1 and monocyte adhesion to cultured endothelial cells.[27] Therefore, IMF accumulation could be associated with IMT thickening through increased insulin resistance. On the other hand, IMF (i.e., the accumulation of adipose tissue surrounding skeletal muscle microvessels) has been widely defined as perivascular adipose tissue (PVAT) involving large arteries and veins and organ-specific vasculature.[28] Previous studies have shown PVAT to be a metabolically active endocrine organ that modulates vascular function.[29] Adipocytes express and secrete a wide range of bioactive molecules, known as adipokines, which can act in a paracrine manner.[30] Given its anatomical proximity to the vessel wall, PVAT may exert more immediate and direct effects on the underlying vasculature compared to distant adipose tissue depots (e.g., subcutaneous adipose tissue).[29] However, the exact biological cascade linking PVAT and changes in vascular function remains incompletely understood, warranting further exploratory studies.

There are several limitations to the present study. First, the data in our study were not normally distributed and the statistical powers (1-$\beta$, $\alpha$ = 0.05) were medium (0.440–0.633); larger studies are needed to further address the effects of age, sex, disease, and BMI on our findings. Incidence of cardiac diseases among women is known to increase 10–20 years later than that among men, which suggests that physiological estrogen levels confer cardioprotective effects.[31] Furthermore, estrogens play a leading role in the pathogenesis and consequences of female obesity, and loss of estrogens after menopause, independent of aging, increases total adipose tissue mass and decreases lean body mass.[32] Therefore, in this study, we focused only on men to examine the effects of aging and minimize the effects of estrogen. Future studies are needed to investigate pre- and postmenopausal women. Second, our study had a cross-sectional design; therefore, we could not affirm whether muscle atrophy and IMF accumulation resulted in the decline of vascular structure and function. Further longitudinal studies are needed to investigate the relationship between muscle atrophy and IMF accumulation and vascular structure and function.

In conclusion, our findings suggest that muscle atrophy and increased body fat percentage could be associated with age-specific changes in vascular structure and function among normal-weight males in their 60s, whereas IMF accumulation may contribute to the decline in vascular structure after the age of 70 years.

## Acknowledgments

This work was supported by Japan Society for the Promotion of Science KAKENHI Grant-in-Aid for Young Scientists (20K19721). The authors graciously thank the volunteers for their participation as well as the laboratory members for their cooperation.

## Conflicts of interest

The authors declare no conflicts of interest.

## References

1. Pischon T, Boeing H, Hoffmann K, et al. General and abdominal adiposity and risk of death in Europe. *N Engl J Med*. 2008;359:2105–2120.
2. Correa-Rodríguez M, González-Ruíz K, Rincón-Pabón D, et al. Normal-weight obesity is associated with increased cardiometabolic risk in young adults. *Nutrients*. 2020;12:1106.
3. Dvoretskiy S, Lieblein-Boff JC, Jonnalagadda S, et al. Exploring the association between vascular dysfunction and skeletal muscle mass, strength and function in healthy adults: a systematic review. *Nutrients.* 2020;12: 715.
4. Csapo R, Malis V, Sinha U, et al. Age-associated differences in triceps surae muscle composition and strength - an MRI-based cross-sectional comparison of contractile, adipose and connective tissue. *BMC Musculoskelet Disord*. 2014;15:209.
5. Yim JE, Heshka S, Albu JB, et al. Femoral-gluteal subcutaneous and intermuscular adipose tissues have independent and opposing relationships with CVD risk. *J Appl Physiol (1985)*. 2008;104:700–707.
6. Shoji T, Maekawa K, Emoto M, et al. Arterial stiffness predicts cardiovascular death independent of arterial thickness in a cohort of hemodialysis patients. *Atherosclerosis*. 2010;210:145–149.
7. Clemente G, Mancini M, Giacco R, et al. Visceral adiposity and subclinical atherosclerosis in healthy young men. *Int J Food Sci Nutr.* 2015;66:466–470.
8. Polak JF, Szklo M, O'Leary DH. Associations of coronary heart disease with common carotid artery near and far wall intima-media thickness: the multi-ethnic study of atherosclerosis. *J Am Soc Echocardiogr.* 2015; 28:1114–1121.
9. Lee JJ, Woodard GA, Gianaros PJ, et al. Ectopic adiposity is associated with autonomic risk factors and subclinical cardiovascular disease in young adults. *Obesity (Silver Spring).* 2015;23:2030–2036.
10. Kollias A, Lagou S, Zeniodi ME, et al. Association of central versus brachial blood pressure with target-organ damage: systematic review and meta-analysis. *Hypertension*. 2016;67:183–190.
11. Roman MJ, Devereux RB, Kizer JR, et al. Central pressure more strongly relates to vascular disease and outcome than does brachial pressure: the Strong Heart Study. *Hypertension*. 2007;50:197–203.
12. Loenneke JP, Fahs CA, Heffernan KS, et al. Relationship between thigh muscle mass and augmented pressure from wave reflections in healthy adults. *Eur J Appl Physiol*. 2013;113:395–401.
13. Okamoto T, Hashimoto Y, Kobayashi R. Isometric handgrip training reduces blood pressure and wave reflections in East Asian, non-medicated, middle-aged and older adults: a randomized control trial. *Aging Clin Exp Res*. 2020;32:1485–1491.
14. Dixon WT. Simple proton spectroscopic imaging. *Radiology.* 1984;153: 189–194.
15. Ogawa M, Yoshiko A, Tanaka N, et al. Comparing intramuscular adipose tissue on T1-weighted and two-point Dixon images. *PLoS One*. 2020;15: e0231156.
16. Franklin SS, Gustin W 4th, Wong ND, et al. Hemodynamic patterns of age-related changes in blood pressure. The Framingham Heart Study. *Circulation*. 1997;96:308–315.
17. McCall GE, Byrnes WC, Dickinson A, et al. Muscle fiber hypertrophy, hyperplasia, and capillary density in college men after resistance training. *J Appl Physiol (1985)*. 1996;81:2004–2012.
18. Hope SA, Tay DB, Meredith IT, et al. Waveform dispersion, not reflection, may be the major determinant of aortic pressure wave morphology. *Am J Physiol Heart Circ Physiol*. 2005;289:H2497–H2502.
19. Mitchell WK, Williams J, Atherton P, et al. Sarcopenia, dynapenia, and the impact of advancing age on human skeletal muscle size and strength; a quantitative review. *Front Physiol*. 2012;3:260.
20. Keske MA, Premilovac D, Bradley EA, et al. Muscle microvascular blood flow responses in insulin resistance and ageing. *J Physiol*. 2016;594: 2223–2231.
21. Landecho MF, Colina I, Sunsundegui P, et al. Comparison of correlations of equation-derived body fat percentage and body mass index with carotid intima-media thickness. *Acta Diabetol*. 2019;56:373–375.
22. Abramson JL, Lewis C, Murrah NV. Body mass index, leptin, and ambulatory blood pressure variability in healthy adults. *Atherosclerosis*. 2011; 214:456–461.
23. Simonds SE, Pryor JT, Ravussin E, et al. Leptin mediates the increase in blood pressure associated with obesity. *Cell*. 2014;159:1404–1416.
24. Matsumura K, Abe I, Tsuchihashi T, et al. Central effects of leptin on cardiovascular and neurohormonal responses in conscious rabbits. *Am J Physiol Regul Integr Comp Physiol*. 2000;278:R1314–R1320.
25. Considine RV, Sinha MK, Heiman ML, et al. Serum immunoreactive-leptin concentrations in normal-weight and obese humans. *N Engl J Med*. 1996; 334:292–295.
26. Goodpaster BH, Krishnaswami S, Resnick H, et al. Association between regional adipose tissue distribution and both type 2 diabetes and impaired glucose tolerance in elderly men and women. *Diabetes Care*. 2003;26:372–379.

27. Madonna R, Pandolfi A, Massaro M, et al. Insulin enhances vascular cell adhesion molecule-1 expression in human cultured endothelial cells through a pro-atherogenic pathway mediated by p38 mitogen-activated protein-kinase. *Diabetologia*. 2004;47:532–536.
28. Britton KA, Fox CS. Perivascular adipose tissue and vascular disease. *Clin Lipidol*. 2011;6:79–91.
29. Lin A, Dey D, Wong DTL, et al. Perivascular adipose tissue and coronary atherosclerosis: from biology to imaging phenotyping. *Curr Atheroscler Rep*. 2019;21:47.
30. Szasz T, Webb RC. Perivascular adipose tissue: more than just structural support. *Clin Sci (Lond)*. 2012;122:1–12.
31. Barrientos G, Llanos P, Basualto-Alarcón C, et al. Androgen-regulated cardiac metabolism in aging men. *Front Endocrinol (Lausanne)*. 2020;11:316.
32. Leeners B, Geary N, Tobler PN, et al. Ovarian hormones and obesity. *Hum Reprod Update.* 2017;23:300–321.