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ABSTRACT

PURPOSE: To evaluate the reliability and validity of retinal vessel diameter measurements by optical coherence tomography (OCT). The effect of age and hypertension on vessel diameter is also examined.

DESIGN: Prospective, cross-sectional study.

METHODS: Two hundred and thirty-eight eyes (238 subjects) with no ocular disease were included. One hundred and six subjects had hypertension and 132 subjects did not. Spectralis HRA+OCT was used to scan a circular region around the optic disc. The outer and inner diameters of the 4 largest retinal arteries and veins were measured using vascular wall reflections on OCT, by which, vessel wall thickness was calculated.

RESULTS: Mean arterial and venous outer diameters were $122.5 \pm 10.9 \mu\text{m}$ and $141.0 \pm 13.1 \mu\text{m}$, respectively. Mean arterial and venous inner diameters were $87.8 \pm 9.4 \mu\text{m}$ and $113.7 \pm 12.5 \mu\text{m}$, respectively. Mean arterial and venous diameter OCT measurements were significantly correlated to mean arterial and venous fundus photography caliber measurements. Arterial and venous wall thicknesses were $17.4 \pm 2.4 \mu\text{m}$ and $13.7 \pm 2.1 \mu\text{m}$, respectively, both of which were highly correlated with subject age (arterial: $r = 0.612$, $P < .001$, venous: $r = 0.455$, $P < .001$). In addition, both mean arterial and venous wall thicknesses were significantly greater in subjects with hypertension than in age-matched subjects without hypertension ($P = .020$ and $.015$, respectively).

CONCLUSIONS: Retinal vessel diameter measurements obtained with OCT were highly reproducible. In addition, vessel walls calculated from outer-and inner-diameters were significantly thickened by both aging and systemic hypertension.

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4 **Age and Hypertension-Dependent Changes in Retinal Vessel Diameter and Wall**
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6 **Thickness: An Optical Coherence Tomography Study**
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4 The retinal vasculature can be directly and noninvasively visualized through the
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6 transparent ocular media. This facilitates assessment of retinal vascular changes
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8 resulting from systemic and/or metabolic diseases.¹⁻³ Classically, funduscopic findings in
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10 the retinal vasculature (i.e., arteriolar narrowing, caliber irregularity, light reflex alterations,
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12 vessel wall sheathing, arteriovenous crossing phenomena)⁴ are categorized according to
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14 the Keith-Wagener-Barker⁵ and/or the Scheie classification systems,⁶ and are used to
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16 diagnose systemic arteriosclerosis or hypertension. In addition, recent semi-automated
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18 measurement software programs allow precise retinal vessel caliber measurements to be
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20 obtained from digital fundus photographs.⁷⁻¹¹ Using these measurements,
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22 population-based studies examined risk factors for various diseases and found that an
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24 increased retinal venous caliber is associated with stroke^{12, 13} and ischemic heart
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26 disease.^{14, 15}
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34 In the past decade, advances in optical coherence tomography (OCT) image
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36 resolution and acquisition speed have allowed more detailed observations of the retinal
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38 architecture, which has greatly enhanced our understanding of the pathomorphology of
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40 various macular diseases.¹⁶⁻¹⁸ Unfortunately, retinal vasculature features have not been
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42 well-studied with OCT and the literature is sparse.¹⁹ Recently, Muraoka and associates²⁰
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44 obtained sectional OCT images of physiologically normal retinal vessels. Vessel
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46 cross-sections were usually oval in shape and appeared as 4 distinct hyper-reflectivities
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48 in a linear configuration. It was concluded that the innermost and outermost
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50 hyper-reflectivities were derived from the vessel wall itself, and that the internal paired
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52 hyper-reflectivities came from flowing blood. Because OCT has a higher resolution in the
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54 direction of the light beam, we assume that OCT allows for precise vertical diameter
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56 measurement of these retinal vessels.
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In the current study, we used Spectralis HRA+OCT to measure retinal vessel diameter and wall thickness in the region surrounding the optic disc. The repeatability and validity of these measurements was also evaluated in subjects without ocular and systemic disease. The influence of age and hypertension on retinal vessels was also examined using measurements from subjects without systemic hypertension, and from age-matched subjects with or without systemic hypertension.

METHODS

The Institutional Review Board and Ethics Committee of Kyoto University approved this study, which was conducted in accordance with the tenets of the Declaration of Helsinki. Written informed consent was obtained from each subject before any study procedures or examinations were performed.

• **PATIENTS:** This prospective study consisted of 238 eyes of 238 subjects with no ocular disease. All subjects had no systemic diseases, except for 106 subjects who had hypertension. Retinal vessels were examined with the Spectralis HRA+OCT (Heidelberg Engineering, Heidelberg, Germany) at the Kyoto University Hospital Department of Ophthalmology between October 2011 and December 2012. Eyes that had any of the following were excluded from the study: keratoconus, high myopia (more severe than -6 diopters), high astigmatism (more severe than ± 3 diopters), best-corrected visual acuity worse than 20/25, prior intraocular surgery, or any co-existing ocular disease (e.g., glaucoma, diabetic retinopathy, retinal vein occlusion, age-related macular degeneration, senile cataract that diminished image quality). Subjects with systemic disease (i.e., diabetes, dyslipidemia, stroke, ischemic heart disease, or collagen disease), except for

hypertension were also excluded.

Presence of hypertension was determined based on antihypertensive agent use or by a physician's diagnosis. After a comprehensive medical interview, refractive error was measured, keratometry was performed, and, after pupil dilation, 45° digital fundus photographs were obtained (TRC-50LX; Topcon, Tokyo, Japan; 3,216 × 2,136 pixels). Finally, OCT examination was performed with the Spectralis HRA+OCT. A circular scan, with a diameter of 3.46 mm, centered around the optic disc center was obtained (Figure 1). Each OCT A-scan had a depth of 2 mm and was comprised of 512 pixels, which provided a depth × transverse digital sampling of 3.87 × 7.50 μm per pixel. An average of 100 scans obtained at the same location was used as the each section.

• **MEASUREMENT OF RETINAL VASCULAR CALIBER ON FUNDUS**

PHOTOGRAPHS: We used 45° digital fundus photographs, centered 5° nasal to the fovea, to measure the retinal vessel caliber, as described previously.^{21, 22} Briefly, fundus photographs were imported into computer-imaging software (IVAN, University of Wisconsin, Madison, WI), and the caliber of each vessel located 0.5–1 disc diameter from the optic disc margin (zone B) was measured in a semi-automated fashion.

• **MEASUREMENT OF RETINAL VESSEL OUTER AND INNER DIAMETER AND VESSEL WALL THICKNESS USING OPTICAL COHERENCE TOMOGRAPHY: A**

Spectralis HRA+OCT circular scan, centered on the optic disc, provides cross-sectional images of all major retinal arteries and veins (Figure 1). Major retinal vessels in the peripapillary area radiate from the optic disc and run parallel to the retinal surface. This allows OCT light to be projected vertically to the vessel walls and lumen. We measured

the vessel diameters of the 4 largest retinal arteries and veins. As reported previously, normal vessels had heterogeneous reflectivity, appeared oval in shape, and often had 4 distinct hyper-reflectivities in a linear configuration (Figure 1).²⁰ When the image horizontal:vertical ratio was changed back to 1:1, the vessel cross-sections became round (Figure 1). While retinal vessels sometimes showed a circular vascular wall (Figure 1), most appeared only as reflections from the inner (towards the vitreous) and outer (towards the retinal pigment epithelium [RPE]) sides of the vessel wall. Additionally, all flowing blood appeared inside the vessel as a paired hyper-reflectivity, which was frequently hourglass-shaped (Figure 1).

Using our OCT measurements, vessel wall thickness was calculated according to the following formula: wall thickness = (outer vessel diameter – inner vessel diameter)/2. Masked retinal specialists measured outer and inner vessel diameter of the 4 largest arteries and 4 largest veins on each OCT scan.

• REPRODUCIBILITY OF OPTICAL COHERENCE TOMOGRAPHY VESSEL

DIAMETER MEASUREMENTS: Intervisit reproducibility of vessel diameter OCT measurements was examined in 25 subjects who underwent 2 OCT examinations by the same examiner at 2 different visits. Evaluations were performed 2 weeks to 3 months apart. Interexaminer reproducibility was examined in 20 subjects who underwent 2 OCT scans on the same date, each by a different independent examiner. Repeatability of vessel diameter measurements was examined by having 2 retina specialists independently measure vessel diameters on the same OCT scan in 50 subjects. The intraclass correlation coefficient (ICC) was used to determine repeatability using 1 randomly selected artery and 1 randomly selected vein in each subject's OCT scan.

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4 Additionally, mean outer and inner vessel diameters were calculated by averaging OCT
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6 measurements from the 4 largest arteries and veins. These were compared with mean
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8 vessel caliber measured on digital color fundus photographs with the assistance of
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10 semi-automated computer software.
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16 • **EFFECT OF BLOOD FLOW ON VERTICAL VESSEL DIAMETER MEASUREMENT:**
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19 The potential effect of blood flow on the OCT representation of retinal vessels was
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21 examined *in vitro*. A fine glass tube with an inner diameter of 140 μm (AS ONE, Osaka,
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23 Japan) was connected to a pump to circulate heparinized human blood collected from
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25 healthy volunteers. Blood flow rates were selected arbitrarily and the tube was scanned
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27 using the Spectralis HRA+OCT. A 25-diopter lens was attached to the OCT machine to
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29 focus on the blood flow within the glass tube. Each final OCT image was obtained by
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31 averaging 100 scans. Previous studies have shown that flow velocity in the major retinal
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33 vessels of healthy eyes is approximately 5–35 mm/s.²³ To be within both physiologically
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35 normal and pathological flow velocity ranges, blood flow rate within the glass tube was
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37 varied between 0.0 and 10.0 ml/h (0.0–180.5 mm/s).
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46 • **STATISTICAL ANALYSES:** Statistical analyses were performed using PASW Statistics
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48 (version 18.0, SPSS, Chicago, IL). Vessel diameter values are presented as mean \pm
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50 standard deviation and were calculated using the 4 selected arteries or 4 selected veins
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52 on fundus photographs or OCT sections. Student's t- tests were used to compare
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54 quantitative data with normal distributions and equal variance. Distributions were
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56 examined for statistical differences with chi-square tests and bivariate relationships were
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58 analyzed using Pearson's correlation coefficient. Statistical significance was defined as a
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P value < .05.

RESULTS

The current study examined 238 eyes from 238 subjects (133 male subjects, 105 female subjects), both with and without hypertension. Mean subject age was 62.1 ± 15.6 years and ranged between 21 and 92 years (Table 1).

Reproducibility and validity of OCT measurements were examined in the eyes of healthy volunteers with no systemic disease. Intervisit ($n = 25$), interexaminer ($n = 20$), and interevaluator ($n = 50$) ICCs of randomly selected vessel measurements were all greater than 0.90 (Table 2). Therefore, OCT measurements of vessel diameter in the circular region around the optic disc are highly reproducible.

Mean outer and inner artery and vein diameters were calculated from the 4 largest arteries and 4 largest veins identified. Mean arterial and venous outer diameter was $122.5 \pm 10.9 \mu\text{m}$ (range: 93.3–172.6 μm) and $141.0 \pm 13.1 \mu\text{m}$ (range: 110.3–183.5 μm), respectively, and mean inner arterial and venous diameters were $87.8 \pm 9.4 \mu\text{m}$ (range: 65.3–143.4 μm) and 113.7 ± 12.5 (range: 78.8–148.5 μm), respectively. Mean vessel calibers of arteries and veins in zone B (0.5–1 disc diameter from the optic disc margin), as measured on fundus photographs, averaged $102.9 \pm 12.0 \mu\text{m}$ (range: 76.2–158.9 μm) and $123.3 \pm 17.1 \mu\text{m}$ (range: 59.2–169.7 μm), respectively. Table 3 shows the correlations between mean vessel diameter, as measured on OCT scans, and mean vessel caliber, as measured on fundus photographs. Images from 50 randomly selected healthy subjects were used. Optical coherence tomography measurements of vessel diameter are done in the plane vertical to the retinal surface, but fundus photograph measurements are done in the coronal plane. Although these disparate methods

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4 measured vessel diameter in different planes, OCT and fundus photograph
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7 measurements were correlated for both arterial and venous diameter/caliber
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9 measurements.

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11 As previously mentioned, vessel wall thickness was calculated from outer and
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13 inner vessel diameter measurements. Mean wall thickness was $17.4 \pm 2.4 \mu\text{m}$ (range:
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15 $10.3\text{--}23.0 \mu\text{m}$) in arteries and $13.7 \pm 2.1 \mu\text{m}$ (range: $10.1\text{--}21.8 \mu\text{m}$) in veins. Additionally,
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18 arterial walls generally had a higher reflectivity than venous walls, and arterial walls were
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20 significantly thicker than venous walls ($P < .001$).

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23 Using measurements from healthy eyes in subjects with no systemic disease,
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25 age-dependent changes in vessels were evaluated (Table 4, Figure 2). Although mean
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27 venous outer and inner diameter was not affected by subject age, mean wall thickness
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29 significantly increased with age ($r = 0.455$, $P < .001$). Mean arterial inner diameter was
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31 also not significantly affected by subject age, but mean outer diameter ($r = 0.384$, P
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33 $< .001$) and vessel thickness ($r = 0.612$, $P < .001$) progressively increased with age.
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38 Subjects were then divided into older (≥ 50 years) and younger (< 50 years) groups (Table
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41 5). Arterial outer diameter, arterial wall thickness, and venous wall thickness were
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43 significantly greater in older subjects than in younger subjects (Figure 3). Vessel walls
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45 were 17% and 22% thicker in veins and arteries, respectively, in older subjects compared
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47 to younger subjects.

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50 The effects of hypertension on the retinal vasculature were also examined in older
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52 (≥ 50 years) subjects. Table 6 shows OCT vessel measurements in eyes of subjects with
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54 hypertension and those of age-matched subjects without hypertension. There were no
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56 significant differences between the 2 groups in mean outer or inner diameter in either
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58 arteries or veins. However, both mean arterial and mean venous wall thicknesses were
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4 significantly greater in subjects with hypertension than in those without ($P = .020$
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6 and .015, respectively). Figure 4 shows the scatter plot of mean arterial and mean
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8 venous wall thickness and age. When eyes were divided into hypertension-positive and
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10 hypertension-negative groups, arterial and venous wall thicknesses in both groups were
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12 positively correlated with age.
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16 We mimicked blood flow within the major retinal vessels using fine glass tubes,
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18 through which blood samples were pumped at different flow velocities and scanned with
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20 Spectralis HRA+OCT. These *in vitro* data were used to confirm that OCT measurements
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22 of retinal vessels were not heavily influenced by blood flow speed. Supplementary Figure
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24 1 shows cross-sections of a glass capillary filled with human blood, as represented on
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26 OCT. Blood in the glass tube without flow (0 mm/sec) showed no paired hyper-reflectivity,
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28 but increasing blood flow velocity resulted in the development of a heterogeneous,
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30 hourglass-shaped hyper-reflectivity within the glass tube. However, blood flow changes
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32 had no effect on the measurement of the glass capillary vertical diameters.
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41 DISCUSSION

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43 Recently, Muraoka and associates²⁰ reported that on OCT, physiologically normal retinal
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45 vessels are represented as oval-shaped configurations with 4 distinct hyper-reflectivities.
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47 In that report they concluded that, intermediate paired hyper-reflectivities, which are
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49 frequently hourglass-shaped and occasionally double “c” patterned, resulted from blood
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51 flow within retinal vessels. Cimalla and associates²⁴ reported similar findings in both
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53 experimental animals and in glass tubes with flowing blood. The authors speculated that
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55 the unique paired hyper-reflectivities arose from a combination of shear-induced red
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57 blood cell alignment in vessels and vessel geometry.²⁴
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4 In the current study, we obtained retinal sections from a circular scan surrounding
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6 the optic disc. Because all major retinal vessels radiate from the optic disc, running
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8 parallel to the retinal surface, and the OCT beam is projected perpendicular to the retinal
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10 surface, vertical sections of all major retinal vessels were reliably obtained with a single
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12 scan. As shown in Figure 1, vascular walls sometimes appeared circular in our OCT
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14 sections, but most OCT sections showed only hyper-reflectivities from the outer and inner
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16 sides of vessel walls, where projected OCT light was perpendicular to the vessel wall. We
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18 used these reflections to measure the outer and inner vessel diameters in the vertical
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20 direction only. It was sometimes difficult to precisely determine vessel borders in the
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22 transverse direction on the OCT scan because of the lower reflectivity from the vessel
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24 walls. Furthermore, vessel diameters in the transverse direction could not be measured
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26 correctly when they were obliquely sectioned. In the present study, all intervisit,
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28 interexaminer, and interevaluator ICCs for OCT vessel diameter measurement were
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30 greater than 0.90, and thus, highly reproducible.
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38 As shown in Table 3, the mean vessel diameters, as measured with OCT, were
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40 significantly correlated with mean vessel calibers, as measured on fundus photographs.
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42 Still, it should be noted that the Spectralis HRA+OCT circular scan (centered on the optic
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44 disc) had a radius of 1.73 mm, slightly outside fundus photograph zone B (0.5–1 disc
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46 diameter from the optic disc margin), where vessel caliber measurements were
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48 measured with the assistance of IVAN. Additionally, vessel diameters were measured in
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50 different directions by fundus photograph.
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55 We also used OCT measurements to calculate the thickness of retinal vessel walls.
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57 Arterial and venous wall thicknesses averaged $17.4 \pm 2.4 \mu\text{m}$ (range: 10.3–23.0 μm) and
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59 $13.7 \pm 2.1 \mu\text{m}$ (range: 10.1–21.8 μm), respectively. This is in agreement with Hogan,²⁵
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4 who reported that the retinal arterial wall had 5–7 layers of smooth muscle cells and a
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6 thickness of approximately 18 μm near the optic disc. Our results also agree with those
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8 reported by Raff and associates²⁶ and Lehmann and Schmieder²⁷ who studied the retinal
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10 arteriolar wall thickness using scanning laser Doppler flowmetry. Retinal arteriole wall
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12 thickness was $13.0 \pm 2.5 \mu\text{m}$ at a location 2–3 mm superotemporal to the optic disc.²⁶
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15 Although only limited information is available on retinal vessel wall thickness, our OCT
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17 measurements are thought to be quite close to previous estimates obtained through
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19 histological measurements, as well as by other modalities.
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24 Previous fundus photography studies, which included a large number of eyes,
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26 have reported an age-dependent reduction in major retinal artery and vein caliber.^{8, 28-30}
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28 We also examined age-dependent changes in the retinal vasculature using OCT in
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30 subjects without hypertension (Table 4, Figure 2). Mean inner diameter did not change
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32 with subject age, and we speculate that this was due, at least in part, to our small sample
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34 size. However, retinal vessel wall thickness, particularly arterial wall thickness,
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36 significantly increased with subject age, despite our small sample size. Additionally, in
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38 older subjects (≥ 50 years old), mean venous and arterial wall thickness was 17% and
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40 22% greater than those in younger subjects.
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46 The effects of systemic hypertension on retinal vascular caliber were studied using
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48 thousands of fundus photographs in various population-based studies. The
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50 Atherosclerosis Risk In Communities³¹ and the Beaver Dam Eye Study³² reported
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52 negative correlations between blood pressure and retinal arteriolar diameter. The Blue
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54 Mountains Eye Study³⁰ showed an increase in retinal venular caliber that was associated
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56 with hypertension. However, in the current study, we did not find any significant effects of
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58 systemic hypertension on either retinal arterial or retinal venous vessel diameter. We
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4 believe this result differed from previous larger studies because of our relatively small
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6 sample size. Interestingly, our OCT examinations did show a significant increase in
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8 retinal arterial and venous wall thickness in subjects with systemic hypertension, in
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10 comparison with the corresponding values in subjects who did not. Retinal vessel wall
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12 thickness may be more sensitive than vessel diameter to systemic hypertension.
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14 Because our data were not adjusted for factors such as diabetes, dyslipidemia, or
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16 smoking, we cannot say how other systemic factors contribute to retinal vessel wall
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18 changes.
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24 Arteriosclerosis in large elastic arteries is characterized by patchy, nodular, and
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26 fatty infiltrations in the intima, and by fibrosis,³³ which often predisposes patients to stroke
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28 and ischemic heart disease. In cardiology and neurology, increased arterial wall
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30 thickness and lumen narrowing are considered important factors in arteriosclerosis
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32 assessments.³⁴⁻³⁷ Intima-media thickness can be precisely measured in the coronary
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34 arteries with intravascular OCT, but this procedure is highly invasive.^{35, 37-41} In a previous
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36 histological study, it was reported that arteriosclerotic changes in retinal arteries were
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38 somewhat different from those seen in large elastic arteries,³³ but that retinal arteries
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40 always accompanied the wall thickening as a result of intimal hyalinization, endothelial
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42 hyperplasia, and medial hypertrophy.⁴² However, to date, there is no established method
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44 to evaluate arteriosclerotic changes in the human retina.
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51 Some investigators believe that vertical elongation of retinal vessels occurs in
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53 OCT sections. In the present study, vessel diameters were measured on OCT images
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55 shown at a vertical/horizontal scale ratio of 2:1, in which retinal vessels were seen to
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57 have an oval configuration. However, when the scale ratio was changed to 1:1, vessel
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59 sections appeared round. Additionally, we examined the effect of blood flow on retinal
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vessel representation on OCT using *in vitro* measurements. Blood samples were pumped through glass tubes at different flow velocities and scanned with the Spectralis HRA+OCT. As shown in Supplementary Figure 1, the measured vertical diameter of the glass capillary tube was not dependent on blood flow velocity. Additionally, the external limiting membrane-line, the photoreceptor inner-outer segment junctions, and the RPE lines showed no faulting beneath major retinal vessels on OCT images. This suggests that the refractive index difference between retinal parenchyma and retinal vessels (red blood cells) was too small to cause image distortion.

- **STUDY LIMITATIONS:** The major limitations of our study are its small sample size, which resulted in a reduced statistical power. In addition, retinal vessel diameters were measured manually on OCT sections and automated software is needed to further standardize measurement methods. Moreover, as noted earlier, systemic conditions such as diabetes, dyslipidemia, and smoking history were not examined in our study. However, despite these shortcomings, we have demonstrated that measurement of retinal vessel diameter with OCT is highly reproducible, and that results are correlated with fundus photograph measurements. In addition, we were able to demonstrate that increased vessel wall thickness in both arteries and veins is strongly correlated with aging. Furthermore, vessel wall thickness is modified by systemic hypertension. Due to its noninvasive nature, OCT measurement of retinal vessel diameters and wall thickness has considerable potential for evaluating retinal arteriosclerotic changes, along with systemic disease associations.

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C. Contributions of Authors: Conception and design of the study (YM, KK, AT); analysis and interpretation (YM, KK, AT, KO, TM, YA-K, KM, NY); writing of the article (YM, KK, AT); critical revision of the article (KO, TM, YA-K, KM, NY); final approval of the article (YM, KK, AT, KO, TM, YA-K, KM, NY); data collection (YM, KK, AT, KO, TM, YA-K, KM).

D. Ethics Committee Approval: The Institutional Review Board and Ethics Committee of Kyoto University approved this prospective study, which adhered to the tenets of the Declaration of Helsinki. Written informed consent for research participation was obtained from each subject before examination.

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FIGURE CAPTIONS

FIGURE 1. Retinal vessel optical coherence tomography (OCT) cross-sections from circular scans surrounding the optic disc. (Top left) Infrared image. A circular OCT scan was performed along the green arrow. (Top right) Corresponding sectional OCT image. All major retinal vessels are represented as a group of heterogeneous reflectivities in an oval-shaped configuration (vertical:horizontal ratio = 2:1). (Bottom left) Magnified OCT image (white square in panel top row-right). The innermost and outermost hyper-reflectivities are the top and bottom of the vessel walls. (Bottom middle) An OCT image with a corrected vertical:horizontal ratio (from 2:1 to 1:1, same area as panel Bottom left). Retinal vessel sections now appear round. (Bottom right) Magnified OCT image of a retinal artery. A circular vascular wall can be seen.

FIGURE 2. Effects of age on mean arterial outer diameter (Top left, $r = 0.384$, $P < .001$), mean arterial inner diameter (Top middle), mean arterial wall thickness (Top right, $r = 0.612$, $P < .001$), mean venous outer diameter (Bottom left), mean venous inner diameter (Bottom middle), and mean venous wall thickness (Bottom right, $r = 0.455$, $P < .001$) in subjects without systemic or ocular disease.

FIGURE 3. Increased thicknesses of arterial and venous walls in an older subject. (Top left) Fundus photograph from a 32-year-old man with no ocular disease. (Top middle) Corresponding infrared image. Optical coherence tomography (OCT) cross-section obtained along the green circular arrow. Red arrows indicate retinal arteries, and blue arrows indicate retinal veins. (Top right) Retinal vessels, as represented on OCT images. (Bottom left) Fundus photograph from a 70-year-old man with no ocular or systemic

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4 disease. (Bottom middle) Corresponding infrared image. The OCT cross-section
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6 obtained along the green circular arrow. Red arrows indicate retinal arteries, blue arrows
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8 indicate retinal veins. (Bottom right) Retinal vessels, as represented on OCT images.
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11 Note the thicker venous and arterial walls and the stronger vessel wall reflectivity in the
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14 older subject.

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19 **FIGURE 4.** Scatter plot of mean arterial and venous wall thickness vs. age. When eyes
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21 were divided into hypertension-positive and hypertension-negative groups, both arterial
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23 (Left) and venous (Right) wall thicknesses were positively correlated with age in both
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25 groups. In the hypertension-positive group, arterial and venous wall thicknesses were
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27 greater than those in the hypertension-negative group.
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33 **Supplementary FIGURE 1.** Cross-sectional optical coherence tomography images
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35 (Spectralis HRA+OCT) of a blood-filled fine glass tube at different blood flow velocities.
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37 Note the homogeneous reflectivity with no blood flow (flow rate = 0 mm/sec). When blood
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39 flow velocity was increased, the single internal reflectivity became multiple
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41 heterogeneous, hourglass-shaped hyper-reflectivities. Glass tube vertical diameter
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43 measurements were not affected by changes in blood flow velocity.
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TABLE 1. Subject Characteristics

Age (years, range)	62.1 ± 15.6 (21–92)
20–39 years (n)	28
40–59 years (n)	51
60–79 years (n)	142
80+ years (n)	17
Gender (male/female)	133/105
Hypertension (+/–)	106/132
Refractive error (diopters)	-0.27 ± 1.94
Keratometer reading (mm)	7.65 ± 0.30

TABLE 2. Intraclass Correlation Coefficient for Measuring Retinal Vessel Diameters by Optical Coherence Tomography

	Intraclass Correlation Coefficient		
	Intervisit (n = 25)	Interexaminer (n = 20)	Interevaluator (n = 50)
Arterial outer diameter	0.964	0.944	0.959
Arterial inner diameter	0.969	0.948	0.972
Venous outer diameter	0.977	0.958	0.980
Venous inner diameter	0.982	0.961	0.978

TABLE 3. Correlations between Mean Retinal Vessel Diameter, as Measured on Optical Coherence Tomography Images and Fundus Photographs (n = 50 eyes)

	r	P value
Mean arterial outer diameter	0.286	.044
Mean arterial inner diameter	0.395	.005
Mean venous outer diameter	0.450	.001
Mean venous inner diameter	0.455	.001

Mean outer and inner vessel diameters were obtained by averaging values of the four largest vessels. Bivariate relationships were analyzed using Pearson's correlation coefficient.

TABLE 4. Effect of Age on Retinal Vessel Measurements by Optical Coherence Tomography
(n = 133 eyes)

	Mean \pm SD	r	P value
Vessel measurements (μm)			
Mean arterial outer diameter	120.6 \pm 10.3	0.384	< .001
Mean arterial inner diameter	87.3 \pm 8.3	0.098	.262
Mean arterial wall thickness	16.6 \pm 2.6	0.612	< .001
Mean venous outer diameter	140.6 \pm 12.5	0.146	.094
Mean venous inner diameter	114.2 \pm 12.1	-0.004	.964
Mean venous wall thickness	13.2 \pm 2.1	0.455	< .001

Means obtained by averaging values of the 4 largest retinal vessels. Subjects with hypertension were excluded from this analysis.

TABLE 5. Retinal Vessel Optical Coherence Tomography Measurements in Younger and Older Subjects Without Systemic Hypertension

	< 50 years (n = 35 eyes)	≥ 50 years (n = 97 eyes)	P value
Age (mean ± MD, range)	31.1 ± 7.7 (21–46)	66.3 ± 9.1 (51–87)	<.001
Gender (male/female)	16/19	53/44	.340
Vessel Measurements (μm)			
Mean arterial outer diameter	115.0 ± 11.3	122.8 ± 9.1	< .001
Mean arterial inner diameter	86.4 ± 8.8	87.7 ± 8.2	.440
Mean arterial wall thickness	14.3 ± 2.5	17.5 ± 2.0	< .001
Mean venous outer diameter	136.8 ± 12.3	142.0 ± 12.4	.036
Mean venous inner diameter	113.5 ± 11.2	114.5 ± 12.5	.667
Mean venous wall thickness	11.7 ± 2.0	13.7 ± 1.8	< .001

Mean outer and inner vessel diameters and mean vessel wall thicknesses were obtained by averaging measurements of the 4 largest vessels.

TABLE 6. Retinal Vessel Optical Coherence Tomography Measurements in Hypertension-Positive and Hypertension-Negative Groups

	Hypertension (-) (n = 83 subjects)	Hypertension (+) (n = 103 subjects)	P value
Age (years)	68.5 ± 7.8	69.1 ± 8.1	.608
Gender (male/female)	47/36	62/41	.623
Vessel measurements (µm)			
Mean arterial outer diameter	122.7 ± 9.3	125.2 ± 10.9	.099
Mean arterial inner diameter	87.3 ± 8.3	88.5 ± 10.6	.416
Mean arterial wall thickness	17.7 ± 2.1	18.3 ± 1.8	.020
Mean venous outer diameter	142.1 ± 12.5	141.7 ± 13.9	.834
Mean venous inner diameter	114.5 ± 12.7	112.8 ± 13.1	.371
Mean venous wall thickness	13.9 ± 1.4	14.5 ± 2.1	.015

Mean outer and inner vessel diameters, and mean vessel wall thickness were obtained by averaging values of the 4 largest vessels in each subject. Subjects under 50 years of age were excluded to match ages of the 2 groups.

Figure 1
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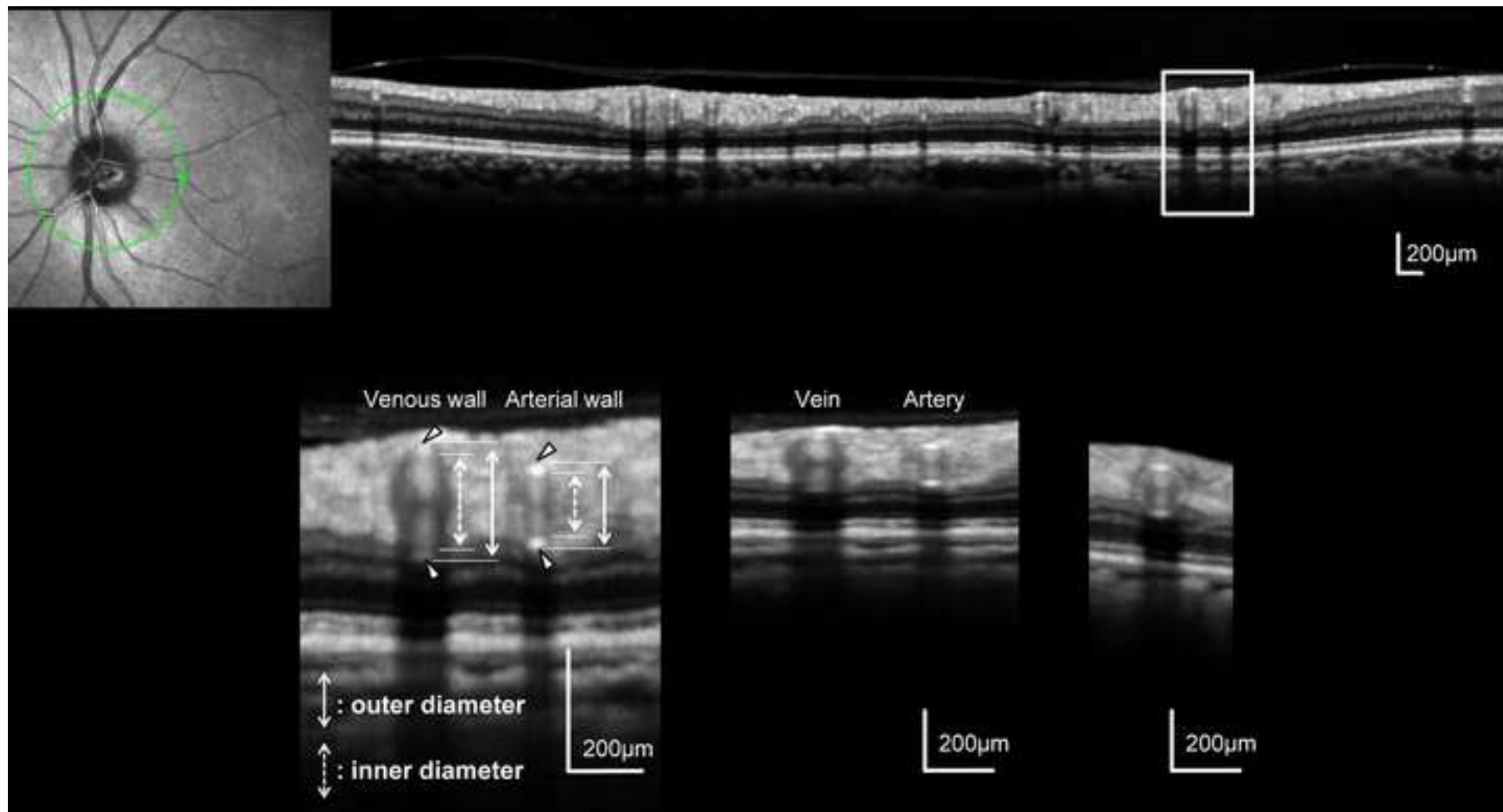


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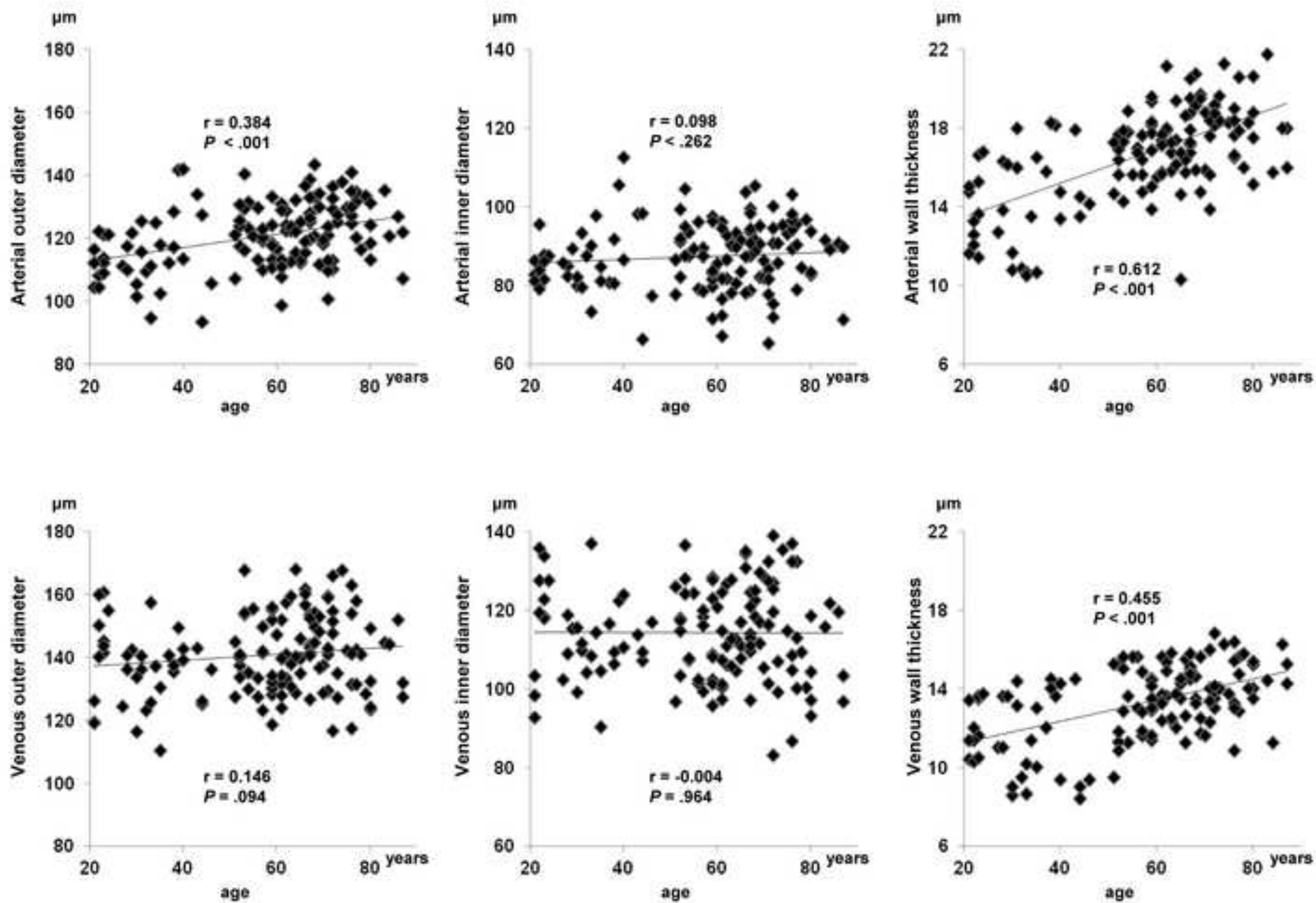


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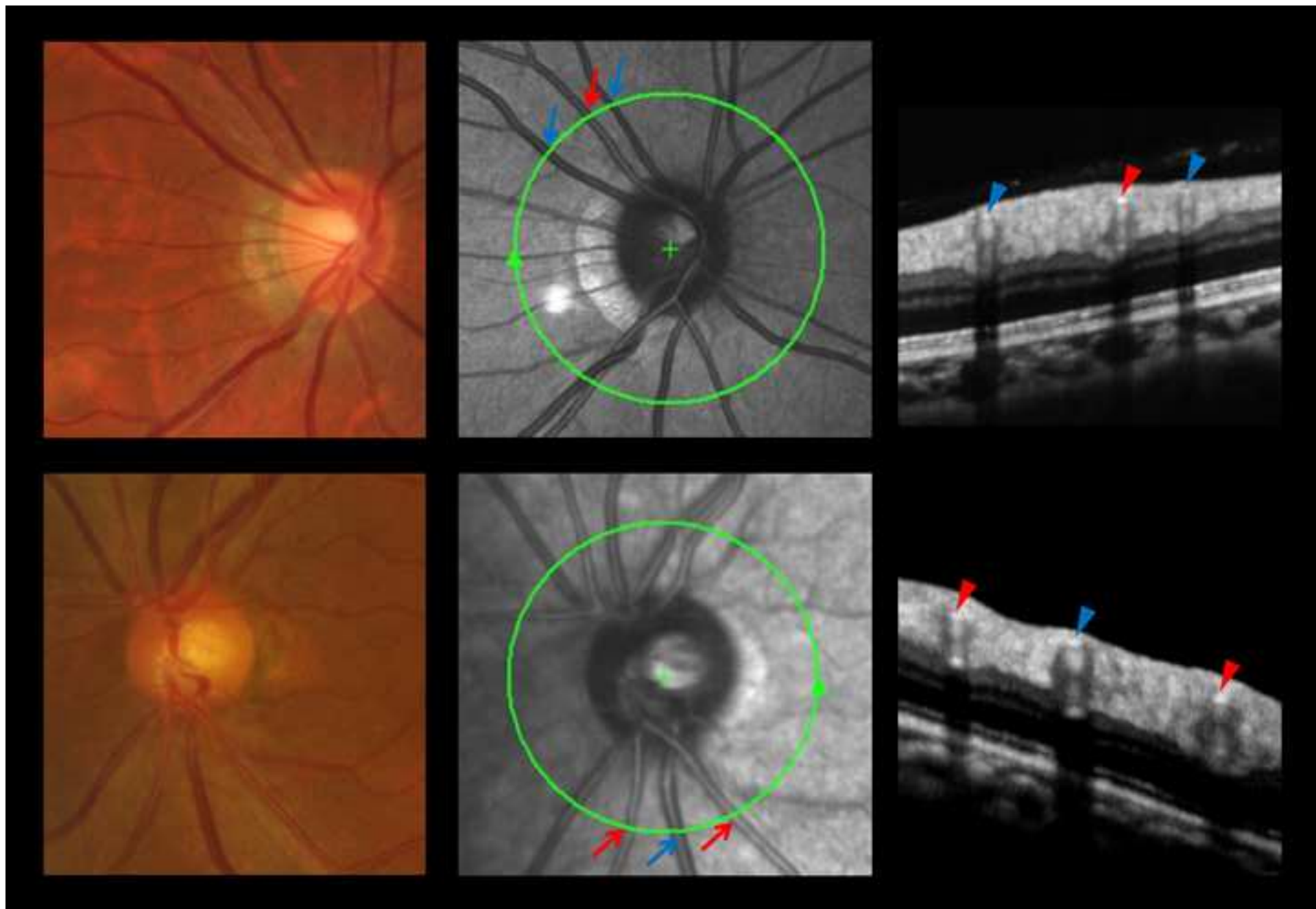
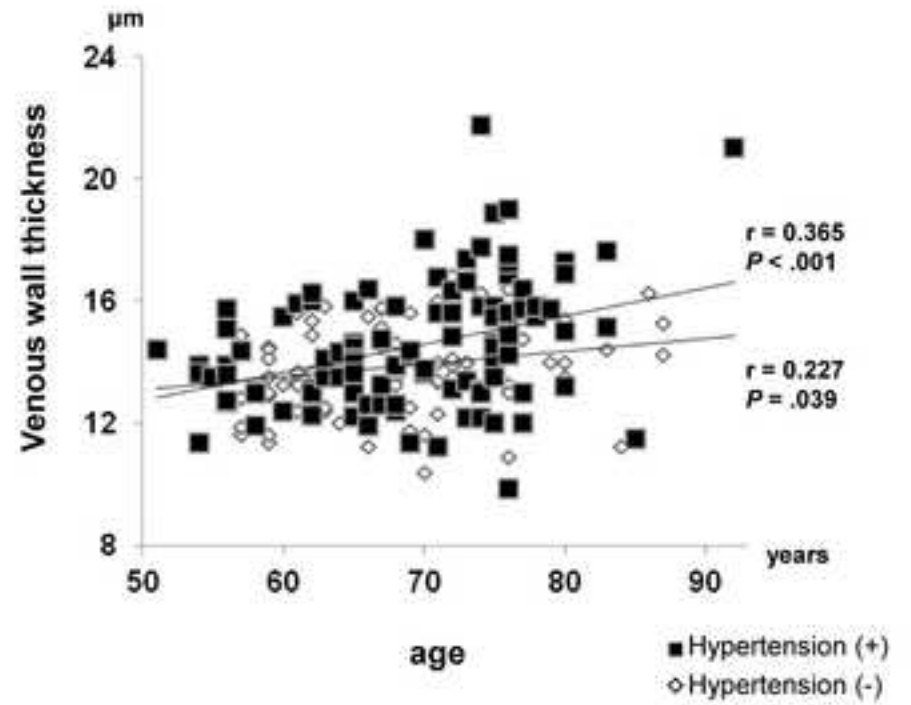
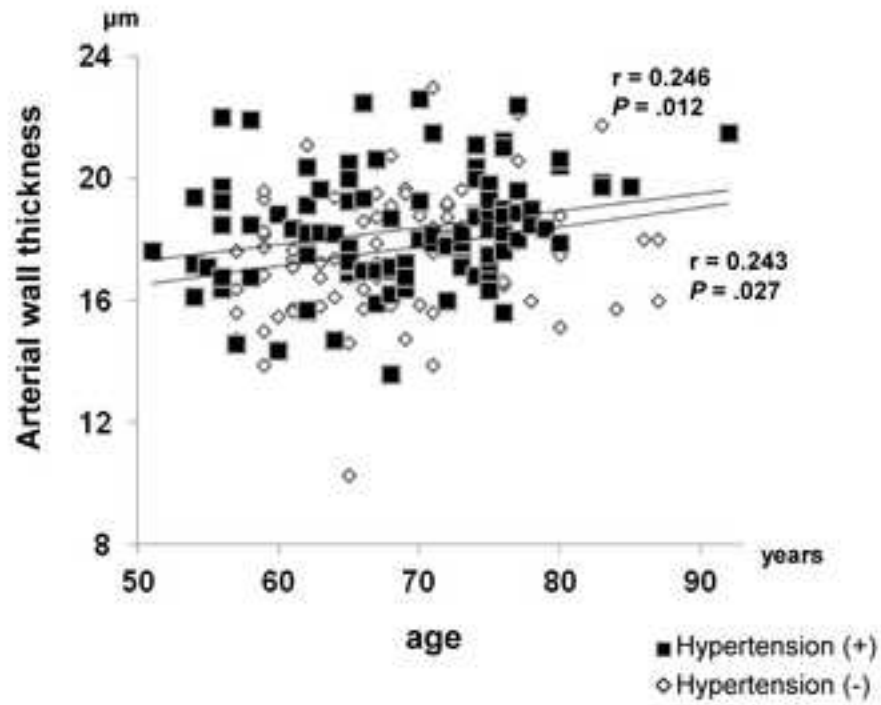


Figure 4
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Supplementary Figure 1

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