Microstructural Alterations of Retinal Arterial Blood Column along the Vessel Axis in Systemic Hypertension

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PURPOSE. Image analysis by the retinal vessel analyzer (RVA) observes retinal vessels in their dynamic state online noninvasively along a chosen vessel segment. It has been found that high-frequency diameter changes in the retinal artery blood column along the vessel increase significantly in anamnestically healthy volunteers with increasing age and in patients with glaucoma during vascular dilation. This study was undertaken to investigate whether longitudinal sections of the retinal artery blood column are altered in systemic hypertension.

METHODS. Retinal arteries of 15 untreated patients with essential arterial hypertension (age, 50.9 ± 11.9 years) and of 15 age-matched anamnestically healthy volunteers were examined by RVA. After baseline assessment, a monochromatic luminance flicker (530–600 nm; 12.5 Hz; 20 s) was applied to evoke retinal vasodilation. Differences in amplitude and frequency of spatial artery blood column diameter change along segments (longitudinal arterial profiles) of 1 mm in length were measured and analyzed using Fourier transformation.

RESULTS. In the control group, average reduced power spectra (ARPS) of longitudinal arterial profiles did not differ when arteries changed from constriction to dilation. In the systemic hypertension group, ARPS during constriction, baseline, and restoration were identical and differed from ARPS during dilation (P < 0.05). Longitudinal arterial profiles in both groups showed significant dissimilitude at baseline and restoration (P < 0.05).

CONCLUSIONS. The retinal artery blood column demonstrates microstructural alterations in systemic hypertension and is less irregular along the vessel axis during vessel dilation. These microstructural changes may be an indication of alterations in vessel wall rigidity, vascular endothelial function, and smooth muscle cells in this disease, leading to impaired perfusion and regulation. (Invest Ophthalmol Vis Sci. 2010;51:2165–2172) DOI:10.1167/iovs.09-3649

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Early diagnosis and prevention of vascular endothelial dysfunction, as well as structural atherosclerotic and hypertensive changes in human vessels is gaining increased importance, especially in the industrialized countries due to sedentary life styles, longer life expectancy, and increasing monetary restrictions within the health systems.

The eye possesses a few unique features in the body. One of them is transparency of the optic media, which allows for direct observation of the microvascular bed in the retina. The fact that retinal vessels are similar in structure, function, and regulation to cerebral vasculature underlines the importance of ophthalmic assessment of the retina for the diagnosis of systemic hypertension. Ophthalmoscopy was introduced a long time ago to detect severe changes in retinal vasculature and to diagnose systemic hypertension in its advanced stages. However, there is evidence that this method does not allow early diagnosis of the disease. One of the earliest and most classic signs of hypertensive retinopathy is arteriolar narrowing, the ophthalmoscopic detection of which is sometimes not specific in earlier stages, and its clinical value is controversial. A correlation of retinal arterial diameter or the arteriole-to-venule ratio with systemic blood pressure was found in several epidemiologic studies. The semiautomatic method, called static vessel analysis, was developed to assess this parameter from retinal photographs and to use it for early detection of ocular and systemic diseases including systemic hypertension. Nagel et al. introduced the retinal vessel analyzer (RVA; Imedos, Ltd., Jena, Germany) to measure temporal reactions of retinal arterioles after stimulation with flickering light. These functional reactions were significantly different in patients with moderate systemic hypertension when compared with healthy volunteers.

From clinical and experimental studies, it is known that arteries of both the microcirculation and the macrocirculation can be afflicted by pathologic changes that include local microirregularities that can interfere with blood flow. A pattern of alternating constrictions and dilations along the vessel has been shown in the microcirculation by in vivo microscopy and in larger vessels by arteriography. This structure of longitudinal vessel profiles has been termed stationary arterial waves or corrugated arteries by different investigators. Primarily, the phenomenon was reported in femoral arteries. Subsequently, it has been observed in carotid, radial, splenic, superior mesenteric, and renal arteries.

Another peculiar structure of longitudinal arterial profiles, the so-called sausage-string appearance, occurs when severe experimental hypertension is provoked in a microcirculation system. The sausage-string pattern in microcirculation is related to the development of vascular damage. Thus, it seems that vessels in the elderly and in some diseases possess a less regular longitudinal profile, for several possible reasons: instability of thin arterial walls, which become more rigid with age; partial endothelial damage of retinal arterioles; and...
partial degradation of smooth musculature of retinal arteriolar walls.\textsuperscript{8,14}

Previously, we found that high-frequency diameter changes in the retinal arterial blood column along the vessel increase significantly in anamnestically healthy volunteers with increasing age\textsuperscript{15} and in patients with glaucoma\textsuperscript{16} during vascular dilatation. These changes may signal endothelial damage.

In the present study, we used an RVA to detect possible structural changes in the retinal arterial vasculature in moderate systemic hypertension. Such alterations in peripheral arterial vessels have been reported earlier in severe systemic hypertension.\textsuperscript{8,11}

\section*{METHODS}

\subsection*{Subjects}

Fifteen patients with essential untreated arterial hypertension (8 men, 7 women, age 50.5 ± 12.2 years) and 15 age- and sex-matched anamnestically healthy volunteers (8 men, 7 women; age 50.9 ± 11.9 years) were entered into the prospective clinical study. In the study, all patients with arterial hypertension and an age-matched corresponding number of the healthy volunteers reported by Nagel et al.\textsuperscript{6} were included. Healthy subjects had no systemic disease and took no medication, with the exception of hormonal contraception in two cases. Patients and volunteers with any ocular disease or affections were excluded: clouding of the optic media, visual acuity less than 0.5, astigmatism more than 2.0 D, myopia more than 7.0 D, hyperopia more than 4.0 D, history of eye surgery or injury in the examined eye, wearing of contact lenses within the previous 24 hours, acute infection, known diabetes mellitus, pregnancy, and nursing.

At the start of the study, a clinical ophthalmic examination was performed, including measurement of visual acuity, objective refraction, slit lamp microscopy, applanation tonometry, and funduscopy. Then, the pupil was dilated with tropicamide 0.5% eye drops. The examination was performed, including measurement of visual acuity, objective refraction, slit lamp microscopy, applanation tonometry, and funduscopy. Then, the pupil was dilated with tropicamide 0.5% eye drops. The preparation for the examination took at least 30 minutes.

Informed consent was obtained from all the subjects after explanation of the nature and possible consequences of the study. The study design was reviewed and approved by the Ethics Committee of the Thuringia State Medical Board and was performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki.

\subsection*{Measurements}

Retinal vessels were assessed dynamically with the RVA. Details of this device and its process of vessel diameter measurement have been described elsewhere.\textsuperscript{14,17} Briefly, the device allows noninvasive online assessment of the retinal arterial and venous red blood cell column diameter depending on time and location along the vessel before, during, and after provocations. For that purpose, the RVA consists of a retinal camera (model 450 FF; Carl Zeiss Meditec, Oberkochen, Germany), a CCD camera for electronic online imaging, and a computer for system control and analysis and recording of the obtained data.\textsuperscript{18}

\subsection*{Functional Stimulation}

During examination, retinal vessels were stimulated with flickering light relying on the principles of neurovascular coupling.\textsuperscript{17-19} This vascular stimulation is described in detail elsewhere.\textsuperscript{6} An optoelectronic shutter was inserted in the retinal camera in place of an additional optical filter. The shutter interrupted the observation light with a frequency of 12.5 Hz.\textsuperscript{6,14} Measurement of the baseline vessel diameter for 100 seconds in continuous light was followed by five cycles of 20-second flicker provocation and an 80-second observation. An arterial segment approximately 1 mm in length was evaluated in each eye. Selection criteria for the segment were location within a circular area of 2 disc diameters, no crossing or bifurcation in the measured segment, curvature of not more than 30°, a distance from neighboring vessels of at least 1 vessel diameter and sufficient contrast to the surrounding fundus. The vessel segment was scanned 25 times per second in the measurement window under optimal conditions. Since the image scale of each eye was unknown, the measured values were expressed in relative units (RU). These units correspond to micrometers if the examination eye has the dimensions of the normal Gullstrand eye. For each measurement time, an interval of 30 seconds before every flicker provocation was considered as the baseline. The statistical mean of arterial diameters during the baseline was calculated, to which the subsequent vessel diameter response was normalized. Relative vessel diameters are reported in percentage of baseline mean.

\subsection*{Pulse and Blood Pressure}

Automatic blood pressure (BP) measurement was obtained with the intensive care monitor (Cardiocup II; Datex Ohmeda, Louisville, CO). Each measurement took approximately 33 seconds and was repeated at 1-minute intervals. From those data the mean systemic arterial blood pressure (MAP) was calculated as: MAP = BP_{diastole} + \frac{1}{3}(BP_{systole} - BP_{diastole}) mm Hg.

\subsection*{Off-line Remeasurement of Data}

To provide higher precision and accuracy, we took the measurements of arterial reactions later off-line from videotape recordings with a novel version of the RVA, the dynamic vessel analyzer (DVA). The peculiarities of the DVA and its improvements on the RVA have been published.\textsuperscript{20} The most important improvement in our study is that the DVA scans each image pixel of a measured vessel segment at a rate of 25 times per second under optimal conditions.

\subsection*{Assessment of Spatial Arterial Blood Column Diameter Change: The Longitudinal Vessel Profile}

Temporal assessment of retinal arterial behavior in response to stimuli is the most common feature of the device already published in several studies.\textsuperscript{6,21,22} Changes in mean vessel segment diameter over time are traced.

The data assessment with DVA allows observation of spatial changes in vessel blood column diameter along a chosen vessel segment at chosen time intervals (Fig. 1). Through this feature, it is possible to assess in vivo dynamic variations noninvasively in the vessel longitudinal structure in humans at different stages of a vessel reaction (Fig. 2). Differences in diameter along the vessel segment during a defined time period can be measured. The method of data acquisition for local vessel analysis with RVA has been explained in detail elsewhere.\textsuperscript{15,16} For each position along a measured vessel segment, the mean of all measurements in this location during the chosen time interval was calculated. We termed the result the longitudinal vessel profile (Figs. 1, 2). Profiles obtained at different time intervals can be

\section*{FIGURE 1. Definition of a longitudinal vessel profile (spatial diameter changes of retinal vessel blood column along its longitudinal section).}
compared. External data transfer of spatial curves obtained by RVA is performed in measurement units (MU); 1 MU equals 12.5 μm in a normal Gullstrand eye.

Data Evaluation and Statistical Methods

Definition of Observation Time Intervals. The time course of vessel diameter change in both groups was plotted. This time response has been partially reported, and it was not the subject of the present study. Average vessel diameter over time demonstrated a diameter increase during provocation. A reactive vessel constriction was observed after cessation of the stimulus, mainly in the control group with an ensuing return to initial baseline values (Fig. 3).

From one complete arterial examination one temporal cycle (from five assessed) was chosen for the evaluation of each subject as basis for our spatial analysis. It consisted of 30 seconds of baseline, 20 seconds of flicker stimulation, and 80 seconds of observation period (Fig. 3). Five time intervals for investigation were defined as shown in Figure 3. Longitudinal vessel profiles during the selected time segments were evaluated for each subject at the defined time intervals (Figs. 2, 3). The start of time segment IV was assigned individually. For each subject, the individual time interval included the point of maximum constriction.

Mathematical and Statistical Analysis. To characterize longitudinal vessel profiles, we obtained their power spectra by Fast Fourier transformation. Each power spectrum was reduced by dividing each value in the frequency distribution by the whole area of the power spectrum, as described in detail elsewhere. For each type of spatial curves and for each group, average power spectra were derived from these reduced individual power spectra by calculation of the median value in the group for each point of frequency distribution, as suggested by others for the analysis of electroencephalograms. The following parameters of the averaged reduced power spectrum (ARPS) were evaluated: average frequencies of peaks and average area under the spectrum within the chosen frequency bands.

A program was created (MatLab 6.1; MathWorks Inc., Natick, MA) to plot power spectra and evaluate the chosen parameters. Longitudinal vessel profile segments of ~800 μm in length (which corresponds to 2^6 = 64 MU) were analyzed for each subject.

A template with corresponding macros was created (MS Excel 2000; Microsoft Corp., Redmond, WA) for each subject to filter, process, and analyze the numerical data from DVA. Since it was impossible to prove the normal distribution of most measurement data (using the Kolmogorov-Smirnoff test), the Mann-Whitney test and the Wilcoxon test were used to assess the significance of differences in the evaluated characteristics. Nonparametric data are expressed as the median (1st quartile; 3rd quartile) and parametric data as the mean ± SD. For comparison of the five different phases of vessel reaction regarding one parameter, necessary adjustment for multiple comparisons was made by the Dunnett method, with a coefficient of 5. Because of the small number of subjects, the nonparametric tests were applied on the level of significance of P = 0.05 for each evaluated parameter, and nonparametric statistics were calculated (SPSS ver. 11.0 for Windows; SPSS Inc., Chicago, IL, and Primer of Biostatistics, ver. 4.03 by Glantz).

FIGURE 2. Assessment of longitudinal vessel profiles. Longitudinal vessel profiles during baseline (green), constriction (red), dilation (blue and light blue), and restoration (brown), as displayed in the DVA system (right bottom). Top right: an average longitudinal vessel profile during the whole examination. Assessed temporal cycle with flicker stimulation is marked within the whole temporal vessel diameter course (left top). Bottom left: assessed arterial (red) and venous (blue) segments are shown within the fundus. 1 RU = 0.08 μm in a normal Gullstrand eye; 1 MU equals 12.5 μm in a normal Gullstrand eye.

FIGURE 3. Example of temporal arterial vessel reaction to flicker provocation and set of time intervals of local vessel reaction assessment.
RESULTS

The mean systemic arterial blood pressure amounted to 110.3 ± 9.7 mm Hg in the hypertensive group and to 95.6 ± 8.5 mm Hg in the control group. No significant changes in blood pressure occurred during the examination. The blood pressure was significantly higher in the hypertensive group (P < 0.01).

There were no statistically significant differences between initial (baseline) average diameters of the measured arterial segments in both groups (P = 0.52). This parameter amounted to 119.6 (109.2; 137.1) RU in the hypertensive group and 112.6 (109.5; 127.0) RU in the control group.

Typical longitudinal arterial profiles are demonstrated in Figure 4. The calculated ARPS of longitudinal arterial profiles for the five phases of arterial reaction in both groups are represented in Figure 5. The sequence of phases was chosen according to the increase in vessel tonus from left to right.

In the control group ARPS did not differ when arteries changed from constriction to dilation. No statistically significant differences in primary or secondary peak frequencies, peak values, or average areas under ARPS within any frequency band were found between the different phases of arterial reaction in the control group (P > 0.2). This confirms our results reported previously in healthy subjects. Typical examples in Figure 4, right, display this finding. The longitudinal arterial profile shifted almost parallel during vessel reaction without changing its configuration. ARPS at all phases possessed three peaks (Fig. 5). The primary one varied from 0.016 to 0.022 Hz at different phases. This range corresponds to one oscillation in 45.5 to 62.5 MU or in 568 to 781 μm—a low-frequency oscillation of a high magnitude. This characteristic frequency is superimposed with other oscillations in Figure 4, right. The secondary peak of the power spectrum varied from 0.035 to 0.042 Hz at different phases and corresponded to one oscillation.
oscillation in 23.8 to 28.6 MU (298–357 μm)—a midfrequency oscillation (Fig. 4, right). There was also a third small peak at 0.07 to 0.08 Hz at all phases, which corresponds to one oscillation in 12.5 to 14.2 MU (156–179 μm)—a high-frequency oscillation of a low magnitude (Fig. 4, right).

In the systemic hypertension group, ARPSs during constriction, baseline, and restoration were identical and differed from both ARPSs during dilation (Fig. 5, bottom). This difference between dilation and other phases is shown in Figure 4, bottom. The average area under the reduced power spectra within the frequency band of 0.03 to 0.06 Hz differed significantly in dilation 1 (0.226 [0.15; 0.364]) from constriction (0.371 [0.241; 0.425]), and restoration (0.352 [0.243; 0.412]; P < 0.05; Fig. 5). This parameter differs significantly in dilation 2 (0.226 [0.15; 0.364]) compared with baseline (P < 0.05).

ARPS during constriction and baseline possessed three main peaks: one primary and two secondary ones of almost the same height. At restoration, two secondary peaks merged into one (Fig. 5). The primary peak varied from 0.015 to 0.018 Hz at different phases corresponding to one oscillation in 55.5 to 66.7 MU (694–853 μm). In Figure 4, left, the frequency is superimposed on other oscillations. The secondary peak of the power spectrum (0.038–0.042 Hz at different phases) corresponds to one oscillation in 18.1 to 19.2 MU (227–240 μm)—a midfrequency oscillation (Fig. 4, left). A third peak at 0.052 to 0.055 Hz at baseline and constriction corresponds to one oscillation in 23.8 to 26.5 MU (298–328 μm)—a midfrequency oscillation (Fig. 4, left). The primary peak frequency at baseline longitudinal arterial profiles did not change during functional vessel reaction to the stimulus of flicker light (Fig. 6, right). In contrast to that finding, the microstructure of the longitudinal arterial profiles in the healthy volunteers did not change during the different phases of the functional vessel reaction (Fig. 5). The profiles in systemic hypertension became less irregular during vessel dilation than during other phases of the vessel reaction to the stimulus of flicker light (Fig. 6, right).

We distinguish between functional and structural alterations of longitudinal arterial profiles. The former are those alterations that change during functional vessel reaction. The latter are the alterations that are persistent and do not change during functional vessel reaction. The longitudinal arterial profiles in the systemic hypertension group do not differ from those of the control group at both dilation phases, although...
they show differences compared with the control group. There are also differences in the dilation phases of hypertensive subjects compared with their baseline and restoration. We therefore postulate that we are able to demonstrate functional changes in the retinal arterial blood column in patients with systemic hypertension. Previously, we demonstrated that healthy subjects display age-related structural changes in their retinal arterial blood column: its microstructure changes with age. The change, however, is independent of the phase of vessel reaction. Functional changes in retinal arterial behavior were also revealed in the patients with primary open-angle glaucoma: Their microstructure of longitudinal arterial profiles did not differ during baseline configuration from that of age-matched healthy volunteers but it changed during vessel dilatory response. A discussion of the possible reasons for our findings in the patients with systemic hypertension follows.

The lumen diameter of a vessel represents its primary functional characteristic. The diameter of a vessel determines its resistance to blood flow. Vessel diameter is determined by its functional and structural properties. The effectors for displaying active functional properties of a vessel are smooth muscle cells, their number, their arrangement, and their state of contraction. The latter is massively influenced by the endothelial cells and their mediators.

Fuhrmann (unpublished data, 2002) and Vilser et al. studied forced retinal arterial constriction during oxygen breathing. They reported that retinal vessels of a patient with hypertension lose their constriction ability in contrast to those in a healthy person. Some segments along a vessel can still react, whereas other segments do not react any longer (Fig. 7).

These results, together with those in the present work, may be explained by the effects of endothelial dysfunction in systemic hypertension, which presumably has focal distribution along the vessel in primary stages of the disease and would lead to nonuniform vessel reaction. Yamada described the irregularity of endothelial nuclei arrangement and their elongation in retinal arteries of renal hypertensive rats.

We propose irregularities in smooth muscle cell status and segmental smooth muscle loss in vessels in systemic hypertension to be a more plausible explanation of the effects on longitudinal vessel structure in systemic hypertension reported in the present work and previously by Vilser et al. (Fig. 7, top). This hypothesis is supported by light and electron microscopic studies. Kimura et al. reported that in the walls of sclerotic blood vessels the smooth muscle cells have been replaced by collagen fibers, proteoglycan filaments, and retinum red-positive materials. Such a pathologic effect had also been described in the human diabetic retina (Rungger-Brändle E, et al. IOVS 1997; 38:ARVO Abstract 3563) (Modified with kind permission of Elisabeth Rungger-Brändle). Fuhrmann (unpublished data, 2002) and Vilser et al. studied longitudinal arterial profiles (changes in vessel diameter along the vessel segment) of a patient with systemic hypertension measured with RVA before and after 100% oxygen breathing. Segmental vessel constriction. (Modified with permission from Vilser W, Nagel E, Lanzl I. Retinal vessel analysis: new possibilities. Biomed Tech (Berlin). 2002;47(suppl 1):682-685.) The top and bottom are adjusted to each other according to the position along the vessel. The loss of smooth muscle cells is assumed in unreactive arterial segments (grey arrows); 1 RU = 1 μm in a normal Gullstrand eye.

**Figure 7. Top:** Segmental loss of smooth muscle cells along a vessel (Runigger-Brändle E, et al. IOVS 1997; 38:ARVO Abstract 3563) (Modified with kind permission of Elisabeth Rungger-Brändle). **Bottom:** Longitudinal arterial profiles (changes in vessel diameter along the vessel segment) of a patient with systemic hypertension measured with RVA before and after 100% oxygen breathing. Segmental vessel constriction. (Modified with permission from Vilser W, Nagel E, Lanzl I. Retinal vessel analysis: new possibilities. Biomed Tech (Berlin). 2002;47(suppl 1):682-685.)

The impaired normal segmental dilation behavior due to the biomechanical alterations of the arterial wall status like vessel narrowing or increase in vessel wall rigidity may represent an alternative reason for our obtained results on smoothing of longitudinal arterial profiles in systemic hypertension during vessel dilation (Fig. 8). A completely relaxed or a completely constricted normal retinal vessel would presumably represent a tube with almost straight parallel inner walls. A normal baseline longitudinal vessel configuration represents a configuration between maximum vessel dilation and its maximum constriction. Smooth muscles of the vessel wall are partially relaxed, partially constricted at this stage, resulting in a wavy vessel profile. A normal vessel has large reserves for reactions in either direction. It never reaches its maximum dilation or constriction during physiological blood flow regulation. Its walls, when considering their shape in a longitudinal section, shift up (dilation) or down (constriction) almost parallel during vessel reaction (Fig. 8, top) in contrast to a diseased vessel with exhausted dilation reserves because of vessel narrowing or alterations in the vessel collagen structure. Such a vessel can reach its maximum dilation ability during its response to flicker stimulus. Consequently, the wavy baseline longitudinal vessel profile becomes flatter (Fig. 8, bottom).

Using methods of computational fluid dynamics (CFD) simulation, we showed in a prior study that an irregular rough structure of the internal vessel wall leads to increased resistance to flow (Kotliar KE, et al. IOVS 2006; 47:ARVO E-Abstract 469): the rougher the inner wall of a retinal vessel, the higher its resistance to blood flow. Consequently, since the arteries in systemic hypertension become smoother during dilation, our morphologic finding may represent a natural mechanism of retinal blood flow regulation in systemic hypertension. Thus, the retinal blood flow in systemic hypertension can be increased during vessel dilation, not only because of internal diameter increase but also through the smoothing of internal
arterial walls at the dilation. This sophisticated mechanism would allow the blood flow to increase in dilated hypertensive vessels to a certain extent, despite the vessel’s reduced dilatory ability. A reduced reactive retinal arterial dilation in systemic hypertension has been reported.7,8 Our CFD results show that a smoothened, low-frequency, longitudinal arterial profile provides more blood flow in comparison to a more wavy midfrequency profile, even if the inflow vessel diameter remains constant (Kotliar KE, et al. IOVS 2006;47:ARVO E-Abstract 469).

Characterization of microirregularities of the arterial inner wall by mathematical methods including frequency analysis is now widely performed in clinical medicine. Several groups have analyzed intima media configuration in the carotid artery.27-29 However, those reported attempts have been limited mostly to large vessels. Vascular characteristics of microcirculation so far are difficult to examine noninvasively in vivo. Retinal vasculature resembles that of the central nervous system and is easily accessible by noninvasive optical methods. Our findings in retinal arteries represent part of the central microcirculation and complement the knowledge about microirregularities in large vessels.

Alstrom et al.7 and Jacobsen et al.8 reported that a uniform increase in vessel wall tension induces a pattern characterized by periodic constrictions and dilations along the vessel. Such patterns have been observed in large and small blood vessels. In macrocirculation, the phenomenon is called “corrugated arteries” and has been described in detail elsewhere.10,11 We want to emphasize that we could see periodic constrictions and dilations along the vessel both in healthy volunteers and in patients with systemic hypertension and were able to differentiate this pattern by using functional provocation.

In severe hypertension, a so-called sausage-string pattern may occur under certain conditions during vessel constriction, which is explained through the instability of the vessel wall rather than by a mechanical failure of the vessel wall due to high blood pressure or to standing pressure waves caused by the heart beat.7,9 The phenomenon was shown in rats mostly in large arterioles of 25 to 50 μm in diameter. The probability of observing the phenomenon in larger or smaller vessels was low.8 The vessels in our study ranged from 90 to 210 μm. According to Jacobsen et al.8, the length of a “sausage” can be estimated by multiplication of the diameter to a coefficient of 6.4. In our case, it would range from 576 to 1344 μm and would presumably occur during vessel constriction. The reasons that we did not observe such a pattern in our systemic hypertension group are: large vessel diameters, insufficiency of flicker-induced vasoconstriction, and no severe form of systemic hypertension in our patient group. However, we cannot exclude mechanical instability of the vessel wall as a possible reason for the main phenomenon observed in the present study: although a relaxed dilated artery in systemic hypertension has a relatively smooth profile, it may switch to the uneven form with periodic constrictions and dilations along the vessel when vessel wall tension increases uniformly (Fig. 6, right) because of the mentioned instability phenomenon.

It could be questioned whether the higher blood pressure in the hypertensive group produces a structural alteration, per se, in the arterioles by increased intraluminal pressure. We think that it does not, since the blood pressure does not reach very high levels that would necessitate a pressure-related passive distension of the vessel. Such a vessel would also not be able to dilate after a stimulus. The vessels we examined did dilate after flicker light application.

Before the conclusion, some considerations of the technical limitations must be elucidated to enable better interpretation of our reported results.

 Frequencies in all power-spectrum charts were calculated in reciprocal local measurement units (MU⁻¹). The term frequency, measured in hertz, is commonly related to the temporal changes. For our purposes in the present work, we introduced the term spatial frequency, which shows that we are dealing with spatial, not with temporal, curves. The measure of spatial frequency is calculated in hertz. Like the temporal meaning of hertz (one oscillation per second) the latter means one oscillation per local measurement unit. For example, the spatial frequency of 0.1 Hz corresponds to one whole oscillation of a longitudinal vessel profile in 10 MU or in 125 μm in a Gullstrand eye. Thus, in the present work, the following remains valid: spatial frequency = MU⁻¹ = Hz. Consequently, since analyzed spatial curves of longitudinal arterial profiles are represented as a percentage of the baseline mean (Fig. 4), the area under a reduced power spectrum and the area under ARPS are dimensionless values: ARPS area = Hz · Hz⁻¹ = 1.

Because of the definition of ARPS, statistical parameters (median, quartiles) of peak or band frequencies calculated from individual power spectra do not always coincide with the peak frequencies seen in ARPS (e.g., Fig. 6, top left). During the averaging of power spectra, individual primary peaks compensate for each other, building a peak that is not necessarily at the site of the median peak frequency.

Although for each curve, one can build a reduced power spectrum, there is an infinite set of curves corresponding to the certain power spectrum chart. They all possess the frequency spectrum with different phases shifted to each other. It is impossible to build an average representative profile of the group from individual ARPS. A simple mean (median, quartiles) of peak or band frequencies calculated from individual profiles do not always coincide with the peak frequencies seen in ARPS (e.g., Fig. 6, top left). During the averaging of power spectra, individual primary peaks compensate for each other, building a peak that is not necessarily at the site of the median peak frequency.

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CONCLUSIONS

Longitudinal sections of the retinal artery blood column do not change for different phases of the vessel reaction in healthy volunteers and undergo microstructural alterations in systemic hypertension, although less irregularly during vessel dilation. These microstructural changes in longitudinal profiles of retinal arteries in systemic hypertension may be an indication for alterations in the vessel wall rigidity, vascular endothelium, and smooth muscle cells in this disease, leading to impaired perfusion and regulation. Changes in vessel microstructure can alter its hemodynamic parameters. Thus, our morphologic finding may represent a natural mechanism that regulates retinal blood flow in systemic hypertension.

References