Investigation of skin vasoreactivity and blood flow oscillations in hypertensive patients: effect of short-term antihypertensive treatment

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In order to evaluate whether arterial hypertension (AH) affects skin microcirculation, 46 newly diagnosed, never-treated, hypertensive patients and 20 healthy normotensive controls underwent a forearm skin postocclusive reactive hyperaemia (PORH) test, using laser-Doppler flowmetry (LDF). Their resting skin blood flow oscillations (SBFOs) were also investigated using wavelet spectral analyses of skin LDF tracings within six frequency subintervals in the 0.005–2 Hz spectral range. To evaluate whether antihypertensive treatment affects skin microcirculation, the same measurements were repeated in 22 of the recruited hypertensive patients after 8 ± 2 weeks of antihypertensive treatment. Significantly reduced PORH, together with significantly higher spectral amplitudes within the majority of the investigated SBFO subintervals, was found in untreated hypertensive patients compared with controls. In the 22 hypertensive patients who completed the follow-up, there was a significant increase in PORH after antihypertensive treatment compared with before (357 ± 178 vs. 284 ± 214%, respectively, $P < 0.05$). Following antihypertensive treatment, the same 22 hypertensive patients did not differ significantly from controls either in PORH or in the majority of the investigated SBFO frequency subintervals. This study showed reduced skin vasoreactivity in the hypertensive patients, confirming that antihypertensive treatment negatively affects skin microcirculation. The short period of efficacious antihypertensive treatment resulted in normalization of skin vasoreactivity in hypertensive patients who completed the follow-up, suggesting that antihypertensive treatment affects positively skin microcirculation in AH. The SBFO increase in untreated hypertensive patients, and its almost complete normalization in treated hypertensive patients, suggests that SBFO enhancement in untreated hypertensive patients could be an adaptive reversible response to AH. *J Hypertens* 29:000–000 © 2011 Wolters Kluwer Health | Lippincott Williams & Wilkins.

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Abbreviations: LDF, laser-Doppler flowmetry; PORH, postocclusive reactive hyperaemia; SBFO, skin blood flow oscillations

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Introduction

During recent years a number of experimental and clinical studies have investigated peripheral microcirculation in human hypertension, providing evidence that microvascular abnormalities are important features of this pathological condition [1–6]. More recently, it has been hypothesized that microvascular abnormalities may initiate the pathogenesis sequence in primary hypertension [7], further stimulating the investigation of peripheral microcirculation in hypertensive patients. In this context, the skin microvascular bed can be a suitable model for study, in accord with the evidence that skin microcirculation may mirror the state of microcirculation in other vascular beds, including cardiac muscle [8–11]. Skin microcirculation is easily accessible to investigation by laser-Doppler flowmetry (LDF), a noninvasive technique that allows continuous, noninvasive, real-time assessment of skin blood perfusion [12]. LDF measurements have revealed a reduced skin blood postocclusive reactive hyperaemia (PORH) in hypertensive patients [4,13], consistent with there being skin microvascular dysfunction associated with arterial hypertension (AH). More recently, the LDF investigation of skin microcirculation has been implemented through the analysis of skin blood flow oscillations (SBFOs), using different methods of spectral analysis [14,15]. It has been demonstrated that some SBFO spectral intervals (ranges of frequency) are related to arteriolar diameter oscillations, that is vasomotion [16], related to endothelial, sympathetic and myogenic activities [14,17,18]. There are also two other SBFO spectral intervals, related to respiratory and cardiac activities, respectively [14]. Some findings suggest SBFO play an important role in optimizing the distribution of blood flow in the microvascular bed [19–21]. Individuals with masked hypertension were found to have abnormalities in their resting SBFO [22]. Abnormalities in postischemic SBFO were observed in newly diagnosed and long-standing hypertensive patients [23].

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To our knowledge, no previous study has investigated possible effects of the antihypertensive treatment on skin vasoreactivity and resting SBFO in hypertensive patients.

We assessed skin vasoreactivity and resting SBFO in newly diagnosed, untreated, hypertensive patients in order to establish whether AH affects skin blood flow parameters. The same tests were repeated on some of the hypertensive patients after 8 weeks of antihypertensive treatment in order to evaluate whether a stable normalization of blood pressure (BP) is, or is not, associated with changes in skin blood flow parameters. This period of treatment was chosen as the minimum time needed to obtain a steady antihypertensive effect.

Materials and methods

Design of the study and patients' selection

We planned to investigate newly diagnosed, hypertensive patients before the initiation of their antihypertensive treatment, and the same patients again after 8 weeks of antihypertensive treatment.

Individuals were recruited among the ambulatory patients who were visiting the hypertension center of our department for the first time. They were enrolled if they met all of the following inclusion criteria: to be affected by mild or moderate essential AH; to be 18–65 years old; and to have given in writing their consent to participate in the study. Exclusion criteria were any one of previous antihypertensive treatment; severe AH; confirmed secondary hypertension; diabetes mellitus; coronary, peripheral or cerebral artery disease; heart failure; renal failure; or smoking.

Apparently, healthy volunteers with normal levels of BP were recruited from among the visitors to our department as controls.

The protocol of this study was approved by the local Ethics Committee and was in accordance with the Helsinki Declaration of 1975, as revised in 2000.

Laser-Doppler flowmetry measurement

LDF measurements were performed with a Periflux PF4001 apparatus (Perimed, Jarfallan, Sweden). This instrument was equipped with a nonheated probe (PF408), which was fixed to the medial surface of the individual's forearm. LDF is based on a laser beam that penetrates the skin and is partially back scattered by moving blood cells. According to the Doppler principle, a frequency shift occurs, generating a signal that is linearly related to the flux of red blood cells [12]. The Periflux PF4001 had the following characteristics: wavelength 780 nm, bandwidth 10 Hz–19 kHz, time constant 0.1 s, sampling frequency 32 Hz. Before each measurement, an LDF calibration was performed using colloidal latex particles whose Brownian motion provides the standard value. The LDF output was recorded continuously by an interfaced computer (Acer, Travelmate 202 T; Acer Italy Srl, Milano, Italy) equipped with the Perisoft dedicated software (Perimed, Järfälla, Sweden), which quantified skin blood flow in the illuminated tissue in terms of conventional perfusion units.

Note that LDF measurements are reproducible and reliable provided, first, that the probes are appropriately positioned (see below) and, second, that proper account is taken of the physiological reality in which, for mammals, there are continuous oscillatory changes in the radii of the blood vessels. The latter effect gives rise to corresponding fluctuations in the recorded signal [14,24,25], and it is precisely the periodic components of these fluctuations that are a major point of interest in the present article.

Resting blood flow laser-Doppler flowmetry measurement

Individuals were asked to abstain from food, drugs, alcohol, coffee and tea for 8 h prior to the LDF measurement. Each individual had 20 min of acclimatization in the supine position at a room temperature of 22–24°C before measurements were initiated. After acclimatization, skin blood flow in his/her right forearm was recorded as described above, the precise measurement site being selected so as to avoid proximity to any of the larger blood vessels, hairs and blemishes. Measurements were always made in the morning, over a period of 30 min in each case.

Postocclusive reactive hyperaemia test

The PORH test was performed using the method reported previously [23,26]. Briefly, after the resting skin blood flow had been recorded for 30 min, right forearm skin ischemia was induced by inflating a pneumatic cuff (which was positioned on the right arm prior to the resting blood flow measurement) to 30 mmHg above the systolic BP of the individual, for 3 min. After this time, the pneumatic cuff was instantaneously deflated and skin blood flow was then recorded for 10 min. The choice of 3 min for the occlusion was based on a previous study [26] showing that 3 min ischemia caused a greater increase in skin perfusion compared with 1 or 2 min, whereas prolonging ischemia for more than 3 min produced patient discomfort.

Basal skin blood flow was taken as the mean value during the 5 min interval before the occlusion; maximal skin blood flow (peak flow) was taken as the higher blood flow value recorded after the 3 min of ischemia. The PORH was expressed as the percentage change in peak flow above baseline. The time taken to reach the maximum postischemic blood flow value (peak time) and the time taken for PORH to decrease to the postischemic 95% value (recoverytime) were also measured in hypertensive patients (both before and after treatment) as well as in controls.

Dynamical analysis of resting skin blood flow

The dynamics of skin blood flow was investigated by application of wavelet analysis [14] to the resting right

forearm skin LDF signal recorded prior to cuff inflation. A Morlet mother wavelet with a central frequency of 1 Hz was used and scaled such that a family of wavelets was generated capable of characterizing frequencies in the range 0.005–2 Hz. On the basis of previous studies [14,18,25], the frequency range 0.005–2 Hz was divided into six frequency subintervals: subinterval I (0.6–2 Hz), related to cardiac activity; subinterval II (0.145–0.6 Hz), related to respiratory activity; subinterval III (0.052–0.145 Hz), related to the spontaneous activity of smooth muscle cells of the microvascular wall (myogenic); subinterval IV (0.021–0.052 Hz), related to sympathetic activity; subinterval V (0.0095–0.021 Hz), related to endothelial (nitric oxide-mediated) activity; and subinterval VI (0.005–0.0095 Hz), related to endothelial (non nitric oxide-mediated) activity [18].

The wavelet analysis was carried out using in-house purpose-written code within the MATLAB software package (The MathWorks Inc., Natick, Massachusetts, USA). The spectral amplitude of each frequency subinterval was calculated as an absolute value. The relative value of each analyzed frequency subinterval was also calculated as the ratio between the absolute amplitude value for that subinterval and the sum of the amplitudes for all frequency subintervals.

Statistical analysis

Data are presented as mean ± standard deviation or as box plots. The three horizontal lines on each box are the 25th, 50th and 75th percentiles. The two lines above and below each box represent the highest and the lowest values, respectively.

A Wilcoxon rank sum test was applied for comparison of nonpaired data (hypertensive patients before treatment and controls, hypertensive patients after treatment and controls). A paired Wilcoxon signed-rank test was applied to pair-matched data (hypertensive patients before and after treatment). All statistical analyses were carried out within the Matlab Statistics Toolbox and, in all tests, $P < 0.05$ was considered statistically significant.

Results

Studied individuals

Forty-six hypertensive patients (aged 50 ± 8 years, range: 32–65 years) who fulfilled the inclusion and exclusion criteria were enrolled in the study and underwent LDF measurement before the beginning of their antihypertensive treatment. We also recruited 20 normotensive healthy individuals as a control group.

Twenty two of the enrolled patients also underwent the second LDF measurement after 8 ± 2 weeks from the beginning of the antihypertensive treatment. The main clinical characteristics of the hypertensive patients and controls are summarized in Table 1. Antihypertensive drugs administered to the 22 re-examined patients included calcium antagonists (10 patients), AT_1-antagonists (eight patients), renin inhibitors (one patient) and angiotensin-converting enzyme (ACE)-inhibitors (three patients). These different antihypertensive treatments were effective in normalizing BP in the 22 re-examined patients.

Results of laser-Doppler flowmetry measurements

The results of the LDF measurements are reported in Table 2.

Both the whole group of untreated hypertensive patients and the subgroup of 22 hypertensive patients who underwent the second LDF measurement after treatment had significantly higher basal skin blood flows, compared with controls.

The whole group of 46 untreated hypertensive patients had significantly lower skin PORH as compared with controls (320 ± 211 vs. 457 ± 154%, respectively; $P < 0.05$), whereas they did not differ from controls in the peak-time or in recovery-time values.

The 22 hypertensive patients who were re-examined after treatment showed significantly higher skin PORH after treatment compared with before treatment (357 ± 178 vs. 284 ± 214%, respectively, $P < 0.05$), whereas they did not differ in the peak time and in recovery-time values compared with before treatment. The same 22 patients after treatment did not differ from the controls in skin PORH, nor in peak time or recovery time.

Results of dynamical analysis of resting skin blood flow

The results of the dynamical analysis of resting skin blood flow obtained in the individuals studied are reported in Figs 1 and 2.

Table 1 Main clinical characteristics of the studied individuals

Clinical feature	Controls ($n = 20$)	Whole group HPs before AT ($n = 46$)	Subgroup HPs before AT ($n = 22$)	Subgroup HP after AT ($n = 22$)
Age (years)	49 ± 7	50 ± 8	50 ± 9	
Sex (M/F)	8/12	30/16	12/10	
BMI (kg/m^2)	25 ± 6	27 ± 4	26 ± 4	
SBP (mmHg)	124 ± 6	154 ± 12*	154 ± 11$^{\&}$	132 ± 16**
DBP (mmHg)	79 ± 6	101 ± 7*	100 ± 7$^{\&}$	84 ± 9**

Table shows the main clinical characteristics of the controls, of the 46 studied hypertensive patients before antihypertensive treatment, and of a subgroup of 22 hypertensive patients (who completed the follow-up) before and after antihypertensive treatment. Data are mean and standard deviation. Data were presented as mean ± standard deviation. One symbol indicates statistical differences between hypertensive patients and controls. Two symbols indicate statistical difference between before and after treatment in the subgroup of 22 patients. AT, antihypertensive treatment; F, females; HPs, hypertensive patients; M, males. $^*P < 0.0001$. $^{\&}P < 0.05$. $^{**}P < 0.0001$.

Table 2 Results of laser-Doppler measurements in the studied individuals

Parameter	Controls ($n = 20$)	Whole group HPs before AT ($n = 46$)	Subgroup HPs before AT (n = 22)	Subgroup HPs after AT (n = 22)
Basal BF (PU)	6.3 ± 2.5	$11.4 \pm 7.5^{*}$	$13.7 \pm 9.5^{*}$	$10.6 \pm 6.9^{*}$
PORH (%)	457 ± 154	$320 \pm 211^{*}$	$284 \pm 214^{*}$	$357 \pm 179^{**}$
Peak time (s)	10 ± 3.9	11 ± 3.6	11 ± 3.4	11 ± 3.8
Recovery time (s)	37 ± 15	46 ± 26	47 ± 33	45 ± 28

The table shows the results of laser-Doppler measurements obtained in controls, in the 46 studied hypertensive patients before antihypertensive treatment, and in a subgroup of 22 hypertensive patients (who completed the follow-up) examined before and after antihypertensive treatment. Data are mean and standard deviation. One asterisk indicates differences at the $P < 0.05$ level between hypertensive patients and controls. Two asterisks indicate difference at the $P < 0.05$ level between before and after treatment in the subgroup of 22 patients. AT, antihypertensive treatment; BF, basal flow; HPs, hypertensive patients; PORH, postocclusive reactive hyperaemia; PU, perfusion unit.

The whole group of 46 untreated hypertensive patients had a significantly higher absolute amplitude within subintervals VI ($P < 0.05$), V ($P < 0.01$), IV ($P < 0.05$), II ($P < 0.01$) and I ($P < 0.05$), compared with controls (Fig. 1). The same patients showed a significantly lower relative amplitude within subinterval III ($P < 0.01$) compared with controls (Fig. 1). No significant differences in relative amplitude within the other frequency subintervals (Fig. 1) were observed between the 46 untreated hypertensive patients and the controls.

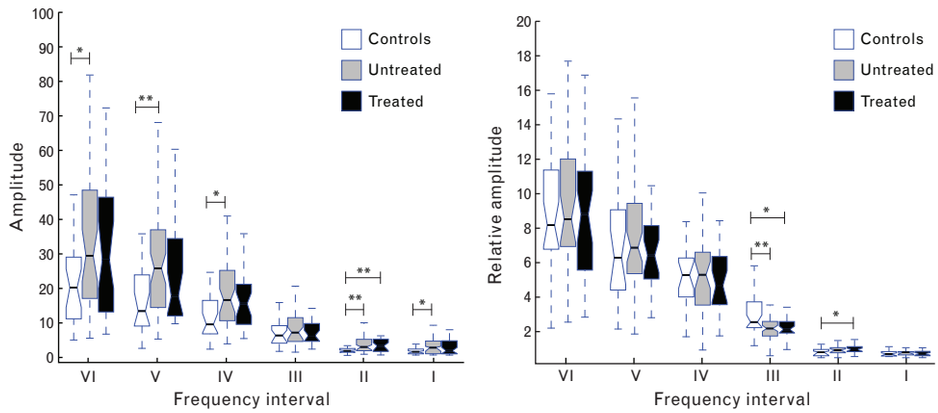
After treatment, the 22 hypertensive patients who completed the follow-up did not differ significantly from the controls in terms of absolute spectral amplitude within frequency subintervals VI, V, IV, III and I (Fig. 1). They showed significantly higher absolute ($P < 0.01$) and relative ($P < 0.05$) amplitudes within subinterval II, as well as a significantly lower relative amplitude within frequency interval III ($P < 0.05$) (Fig. 1) compared with controls. No significant differences in relative amplitude within the other subintervals were observed between the 22 hypertensive patients and the controls.

In these 22 hypertensive patients there was also, after treatment, a significant increase ($P < 0.01$) in the relative amplitude within frequency subinterval II, and a significant decrease ($P < 0.05$) within frequency subinterval I, compared with before treatment (Fig. 2). The absolute or relative values of amplitude in the other frequency intervals did not differ significantly between before and after treatment in these 22 patients (Fig. 2).

Discussion

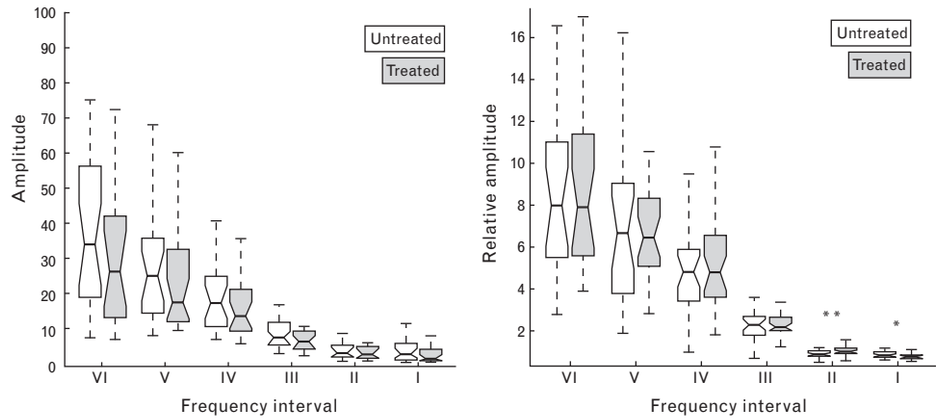
This study has shown that newly diagnosed, untreated, hypertensive patients had a reduced skin PORH and that a subgroup of hypertensive patients who underwent the same test after a short period of antihypertensive treatment had a significantly higher skin PORH, compared with before treatment, as well as no significant differences in skin PORH as compared with control normotensive inviduals. Other findings of this study were SBFO enhancement in untreated hypertensive patients, as well as almost complete SBFO normalisation in hypertensive patients after antihypertensive treatment.

Fig. 1



Results of dynamic analysis of skin blood flow in hypertensive patients and controls. The figure shows the values of the absolute (left panel) and relative (right panel) spectral amplitude obtained from the dynamic analysis of resting skin blood flow in 20 controls (controls), in 46 untreated hypertensive patients (untreated) and in a subgroup 22 hypertensive patients who underwent the same analysis after antihypertensive treatment (treated). Data are presented as box plots. The three horizontal lines at the boxes are the 25th, 50th and 75th percentiles. The two lines out of the boxes represent the highest and the lowest values, respectively. One asterisk indicates differences at the $P < 0.05$ level. Two asterisks indicate difference at the $P < 0.01$ level. I, heart activity, related frequency intervals; II, respiratory activity; III, myogenic activity; IV, sympathetic activity; V, endothelial activity (nitric oxide mediated); VI, endothelial activity (nonnitric oxide mediated).

Fig. 2



Results of dynamic analysis of skin blood flow in a subgroup of 22 hypertensive patients before and after treatment. The figure shows the values of absolute (left panel) and relative (right panel) spectral amplitude obtained from the dynamic analysis of resting skin blood flow in a subgroup of 22 hypertensive patients who completed the follow-up, before (untreated) and after (treated) antihypertensive treatment. Data are presented as box plots. The three horizontal lines at the boxes are the 25th, 50th and 75th percentiles. The two lines out of the boxes represent the highest and the lowest values, respectively. One asterisk indicates differences at the $P < 0.05$ level. Two asterisks indicate differences at the $P < 0.01$ level. I, heart activity, related frequency intervals; II, respiratory activity; III, myogenic activity; IV, sympathetic activity; V, endothelial activity (nitric oxide mediated); VI, endothelial activity (nonnitric oxide mediated).

Skin PORH results mainly from a cessation of the myogenic activity of the microvascular wall, as a consequence of reduced shear stress during the ischemic challenge [27]. A further mechanism of PORH is the build-up of various vasodilator substances, such as nitric oxide and prostaglandins, released in the affected tissue by vascular endothelium [27]. Once the occlusion is released, the reduced resistance offered by the dilated vessels facilitates a significant increase in blood flow. This increase is rapidly reduced by the activation of the myogenic vasoconstrictor response and by the removal of the vasodilator substances from the microvascular bed by the restored blood flow. However, in a previous study [28], it was observed that the build-up of vasodilator substances during an ischemic challenge of 3 min (which was also the period of skin ischemia in our own study) was minimal. Thus, the PORH test in our study was mostly a measure of the myogenic function of the microvascular wall and only to a lesser extent of the microvascular endothelial function.

Our finding of reduced skin PORH in untreated hypertensive patients confirms the results of previous studies [4,13]. Because our 3 min PORH test is mostly a measure of myogenic function, its abnormality in untreated hypertensive patients may reflect a reduced skin vasoreactivity due to myogenic dysfunction. On the contrary, the untreated hypertensive patients did not differ from controls in terms of their PORH peak time, or in their recovery time. This suggests that the vasoconstrictor myogenic response triggered by PORH was intact in our untreated hypertensive patients.

That skin PORH normalization occurs in hypertensive patients after a short period of efficacious antihypertensive treatment is a new finding. Only one previous study [29] has examined the effect of the discontinuation of antihypertensive treatment on skin PORH in hypertensive patients, showing a reduction in skin PORH after withdrawal of the antihypertensive drug. Skin PORH normalization in the hypertensive patients who were reexamined after antihypertensive treatment is consistent with normalization in skin vasoreactivity, as a result of restored myogenic function. Accordingly, because it is mainly the myogenic mechanism that is involved in the 3 min PORH test, the same finding does not allow us to argue that there are improvements in skin endothelial function associated with antihypertensive treatment in our patients. Antihypertensive drugs administered to the studied hypertensive patients included calcium antagonists (10 patients), T_1-antagonists (eight patients), renin inhibitors (one patient), ACE-inhibitors (three patients). Each of these classes of drugs acts through a different mechanism in reducing the amount of vasoconstriction occurring within the vascular system, the net result being a recovery in skin vasoreactivity to ischemia.

In accord with the suggestion that the skin microvasculature mirrors the vascular function of other parts of the body [8–11], we may suppose that antihypertensive treatment exerts a systemic beneficial effect on myogenic vascular function in hypertensive patients. Interestingly, a significant correlation was previously observed [30] between the impairment of skin PORH and the weight of cardiovascular risk in a healthy female population. This correlation gives additional relevance to our finding of

normalized skin PORH in hypertensive patients after antihypertensive treatment.

In dynamical analyses of resting SBFO, we found that untreated hypertensive patients showed a higher absolute spectral amplitude within subintervals VI, V, IV, II and I, compared with normotensive controls. In contrast, for the 22 hypertensive patients who completed the follow-up, there were no significant differences from controls in the same SBFO parameters, the only exception being in SBFO subinterval II in which the amplitude was enhanced in treated hypertensive patients as compared with controls. No significant changes in SBFO were observed before and after antihypertensive treatment in the same group of 22 hypertensive patients. The relatively low patient number of this subgroup weakens the statistical significance of the comparison.

Considering the suggested role of SBFO in optimizing blood flow distribution in the microvascular network [19–21], we may suppose that SBFO enhancement in untreated hypertensive patients could represent an adaptive response to increased BP, with beneficial effects on the blood flow distribution in the skin microvascular bed.

In relation to the possible mechanisms involved in the SBFO modifications observed in our hypertensive patients we advance the following hypotheses. In untreated hypertensive patients, the enhancement of SBFO subintervals VI and V, both of which are related to endothelial activity [17,18], could be due to endothelial activation being triggered by the increased shear stress on the microvessel wall caused by elevated BP. The lack of any enhancement of the same SBFO subintervals in hypertensive patients after an efficacious antihypertensive treatment reinforces this hypothesis.

The enhancement in untreated hypertensive patients of SBFO subinterval IV, related to sympathetic activity [14], might be attributable to sympathetic overactivity, which was found to be associated with AH in a previous study [31].

There was no significant difference in the absolute amplitude of SBFO frequency subinterval III, related to myogenic activity, between either untreated or treated hypertensive patients, and controls. In contrast, the relative amplitudes in this subinterval were significantly reduced in both the untreated and treated hypertensive patients. These findings suggest a relative reduction in the efficiency of the myogenic mechanism involved in the genesis of SBFO subinterval III in hypertensive patients, which remained unaffected by the antihypertensive treatment. We may suppose that the antihypertensive treatment was not carried out for long enough to improve this mechanism in our hypertensive patients.

The enhancement of SBFO sub-interval II, related to respiratory activity, both in untreated and treated hypertensive patients, could be indirectly related to capillary rarefaction, which is a well known abnormality associated with AH [2]. Capillary rarefaction can result in poor oxygen diffusion out of the microvessel lumen and, consequently, in higher oxygen levels in red blood cells in the venules exiting the capillary bed. It is pertinent to recall here that LDF measurements are partly dependent on the oxygen content of the red blood cells [12]. Indeed, for hypertensive patients, the LDF probe of skin blood flow may detect a greater proportion of red blood cells circulating in the venules, which are mostly under the influence of respiratory activity. This may account for the enhancement of the respiratory-dependent SBFO subinterval II being found in both untreated and treated hypertensive patients. Again, we may suppose that the antihypertensive treatment was not carried out for long enough to improve skin capillary rarefaction or for normalizing the amplitude of SBFO subinterval II.

The enhancement of SBFO subinterval I, related to heart activity, in untreated hypertensive patients was an unexpected finding. This SBFO subinterval is due to the transmission to the level of the skin microcirculation of central haemodynamic modifications synchronous with heart activity [14]. Then we could expect a reduction of this SBFO subinterval in untreated hypertensive patients, as a consequence of increased peripheral vascular resistance. However, previous findings showing increased BP at the level of skin capillaries in hypertensive patients [5,6], suggest that the skin microvascular network in hypertensive patients is not protected from the effects of central haemodynamic modifications. This may account for the enhancement of SBFO subinterval I in untreated hypertensive patients.

Our resting SBFO findings for untreated hypertensive patients disagree with the results of previous clinical studies showing no significant changes in resting SBFO in hypertensive patients [22,23]. On the contrary, in agreement with our SBFO findings in untreated hypertensive patients, arteriolar vasomotion (which is the mechanism responsible for SBFO subintervals VI, V, IV and III) was enhanced at the level of skeletal muscle in rats with chronic salt-induced hypertension [32]. The discrepancies between the results of our study and those of the earlier clinical are probably due to different spectral analysis methods being used in our SBFO investigations. We used wavelet analysis, whereas a modified version of Fourier analysis was applied in the previous studies [22,23]. Another important methodological difference between our study and previous clinical studies is the length of time used for the LDF recordings. The greater length of 30 min used in the present study allows for a more adequate analysis of the lowest SBFO frequency sub-intervals. These differences in data collection and analysis are quite sufficient to explain [25] the differences between the present results and the earlier ones [22,23].

Another unexpected finding of this study was the higher resting blood flow found in untreated and treated hypertensive patients, compared with normotensive controls. It is pertinent to recall here that microvessel tone in glabrous skin areas – such as the forearm skin in which flow measurements were performed in the present study – is predominantly mediated by sympathetic cholinergic vasodilator nerves [33]. Sympathetic vasodilator nerve overactivity could account for the higher resting SBFO that we found in hypertensive patients. On the contrary, differences in basal skin blood flow between hypertensive patients and controls did not influence the results of the PORH test in our study because the latter were expressed as a normalized value for resting skin blood flow.

In conclusion, this study showed reduced skin vasoreactivity in untreated hypertensive patients, confirming that AH negatively affects skin microcirculation. A completely new finding was that a relatively short period of efficacious antihypertensive treatment was associated with normalization of skin vasoreactivity in hypertensive patients, suggesting that antihypertensive treatment affects skin microcirculation positively in AH. We also observed an SBFO increase in untreated hypertensive patients and almost complete normalization of the SBFO dynamics in treated hypertensive patients, suggesting that SBFO enhancement in untreated hypertensive patients could be an adaptive reversible response to AH. A larger and longer term study is needed to separate the effects of different drugs, evaluate the effect after longer treatment periods and include a check for capillary rarefaction.

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