Evaluation of carotid artery stiffness and flow-mediated vasodilation in patients with resistant hypertension after renal sympathetic denervation

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Abstract. The aim of the study was to determine the effect of catheter-based renal sympathetic denervation (RDN) on office blood pressure (BP). Thereafter we evaluated the effect of BP lowering on carotid artery stiffness and flow-mediated vasodilation (FMD) using ultrasound and echo-tracking during 6 month follow-up. We found significantly improved office systolic BP changes at 1 month after RDN (p=0.023) together with pulse pressure (PP) changes at 6 months follow-up (systolic BP changes: p=0.041; PP changes: p=0.016). Carotid stiffness values stiffness parameter (beta) and pressure-strain elastic modulus (Ep) were significantly reduced (beta: p=0.04; Ep: p=0.03) and arterial compliance (AC) increased (p=0.03), especially 1 and 3 months after RDN. Mean FMD value in cardiac diastole was significantly improved (p=0.01) too. We observed significant relationships between office BP and carotid stiffness values at 6 months follow-up. Using linear regression model the fall in systolic BP>10 mmHg would be associated with decrease of stiffness parameters beta>1.8%, Ep>55.92 kPa and pulse wave velocity (PWV)>0.7 m/s and with an increase of AC>0.009 mm^2/kPa.
We conclude that the BP lowering effect of RDN was followed by the improvement of structural and functional parameters of artery wall in short-term follow-up study.
Key words: carotid artery stiffness – flow-mediated vasodilatation – renal sympathetic denervation – resistant hypertension

Škultétyová D, Filipová S, Maďarič J, Chňupa P, Fridrich V, Vulev I, Gočár M. **Hodnotenie karotickej tuhosti a prietokom navodenej dilatácie u pacientov s rezistentnou hypertenziou po renálnej denervácii sympatika.** Cardiology Lett. 2014;23(3):215–222

Abstrakt. Cieľom práce je posúdiť efekt renálnej denervácie sympatika (RDN) na ambulantný krvný tlak (TK). Pomocou ultrasonografie v kombinácii s echo-tracking sme hodnotili vplyv poklesu TK na karotickú tuhosť a prietokom navodenú vazodilatáciu (FMD) počas šiestich mesiacov sledovania. Zistili sme významné zlepšenie zmien ambulantného systolického TK v prvom mesiaci (p = 0,023) po RDN a spolu so zmenami pulzného tlaku (PP) v 6. mesiaci sledovania (systolický TK: p = 0,041; PP: p = 0,016). Ukazovatele karotickej tuhosti, parameter tuhosti (beta) a elastický modul (Ep), sa signifikantne znížili (beta: p = 0,04; Ep: p = 0,03) a artériová poddajnosť (AC) sa zvýšila, predovšetkým v prvom a treťom mesiaci po RDN. Stredná hodnota FMD v diastole srdca sa tiež významne zlepšila (p = 0,01). Zistili sme signifikantné korelácie medzi ambulantným TK a hodnotami karotickej tuhosti v 6. mesiaci sledovania. Pomocou lineárnej regresnej analýzy môže pokles systolického TK > 10mmHg viesť k zníženiu parametrov tuhosti beta >1,8 %, Ep > 55,92 kPa a rýchlosti šírenia pulznej vlny (PWV) > 0,7 m/s a k zvýšeniu AC > 0,009 mm^2/kPa.
Na základe výsledkov usudzujeme, že pokles TK po RDN viedol k zlepšeniu štrukturálnych a funkčných parametrov cievnej steny v krátkodobom sledovaní.
Kľúčové slová: karotická tuhosť – prietokom navodená vazodilatácia – renálna denervácia sympatika – rezistentná hypertenzia

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Resistant hypertension (RH) is defined when a therapeutic strategy that includes appropriate lifestyle measures plus a diuretic and two other antihypertensive drugs belonging to different classes at adequate doses fails to lower systolic and diastolic BP levels to <140/90 mmHg, respectively (1). The prevalence of RH has been reported to range from 5-30% of the overall hypertensive population, but the prevalence of true RH is rated at less than 10% (1). The elevated BP in RH may cause cardiovascular (CV) structural and functional alterations.

Arterial stiffness is a hallmark of hypertension and is recognized as an independent predictor of total and CV morbidity and mortality (2). The loss of arterial compliance is associated with the development and progression of endothelial dysfunction (3).

Treatment with catheter-based renal denervation (RDN) has been shown as a promising and a viable therapeutic approach for patients with RH. The endovascular procedure reduces sympathetic renal and central tonus and hence lowers arterial BP. Several studies have reported the BP lowering effect of RDN (4, 5). Until now only a few studies have been devoted to the possible favourable response of RDN on arterial stiffness and central hemodynamics (6, 7).

The aim of our study was to determine the effect of RDN on office BP in patients with RH. Thereafter we evaluated if the RDN also has an effect on carotid artery stiffness and endothelial function via flow-mediated vasodilation (FMD) of brachial artery in short-term follow-up (6 months).

Material and methods

Patients

From March 2012 to March 2013 we performed RDN in 17 patients (age 55±9 years, M:F=12:5) with true RH in the National Institute of Cardiovascular Disease (Bratislava). The procedure was approved by the Ethical Committee of the Institute. All physicians in the team for RDN have obtained the international certificate for RDN (University of Heidelberg, Germany).

The baseline characteristics of patients are shown in **Table 1**. All patients fulfilled the eligibility criteria for RDN (5, 8). All patients signed an informed consent. Patients were followed at 1, 3 and 6 months after the procedure. 3 patients failed the follow-up at 3 and 6 months (All three patients are alive. Causes of loss of follow-up are outside the RDN). Routine blood tests, focused mainly on kidney function, and renal duplex ultrasound examinations were estimated during each visit. Control computed tomography angiogram was performed 1 month after RDN. Echocardiography was performed before RDN and at 6 months follow-up. Besides office BP controls all patients underwent 24 hours ambulatory BP monitoring before RDN and during each follow-up visit.

Office BP measurement

The average office BP was measured in a supine position on the left brachial artery after at least 5 minutes of rest during ultrasoud examination. The office BP was calculated as the mean value of three consecutive measurements with an automatic oscillometric device Microlife BP100 PLUS (Microlife AG, Swiss).

High resolution echo-tracking of carotid arteries and of brachial artery

Measurement of carotid stiffness was performed with ALOKA Prosound Alpha 10 equipment (ALOKA Co., Ltd., Tokyo, Japan). This unit is equipped with an integrated and automated ultrasound, Doppler and echo-tracking system. Echo-tracking system allowed a real-time measurement of the diameter changes between the near and far wall of the common carotid artery (CCA). Accuracy of on-line measurement is one-sixteenth of ultrasound wavelength; the data were updated at a rate of 1 kHz (9). Experimental studies on animals demonstrated that arterial pressure waveforms and diameter-change waveforms were similar (10, 11). Sugawara and coworkers (12) reported in humans the similarity between carotid arterial pressure waveforms measured with a catheter-tipped micromanometer and carotid arterial diameter-change waveforms measured by echo-tracking. Brachial systolic and diastolic BP was used for calibration of the maximum and minimum values of diameter-change waveforms and was used as a surrogate for BP waveforms. We used 5 consecutive beats ensemble-averaged to obtain a representative waveform **(Figure 1)**.

The stiffness parameters were measured on both sides of the CCA after clear visualization of intima-media complex on anterior and posterior wall in its longitudinal axis and after applying echo-tracking to the adventitia of the wall. Following stiffness values were evaluated (13, 14):

- stiffness index (beta), calculated from changes in vessel diameter and BP:
  beta=In (Ps/Pd)/[(Ds-Dd)/Dd],
  where: In = the logarithm, Ps = systolic BP, Pd = diastolic BP,
  Ds = arterial systolic diameter, Dd = arterial diastolic diameter
- pressure-strain elasticity modulus (Ep), index of vessel elasticity, calculated from changes in vessel diameter and BP:
  Ep=(Ps-Pd)/[(Ds-Dd)/Dd]
- arterial compliance (AC), index of blood vessel compliance, calculated from the arterial cross area and BP:
  AC=Ad/R(Ps-Pd),

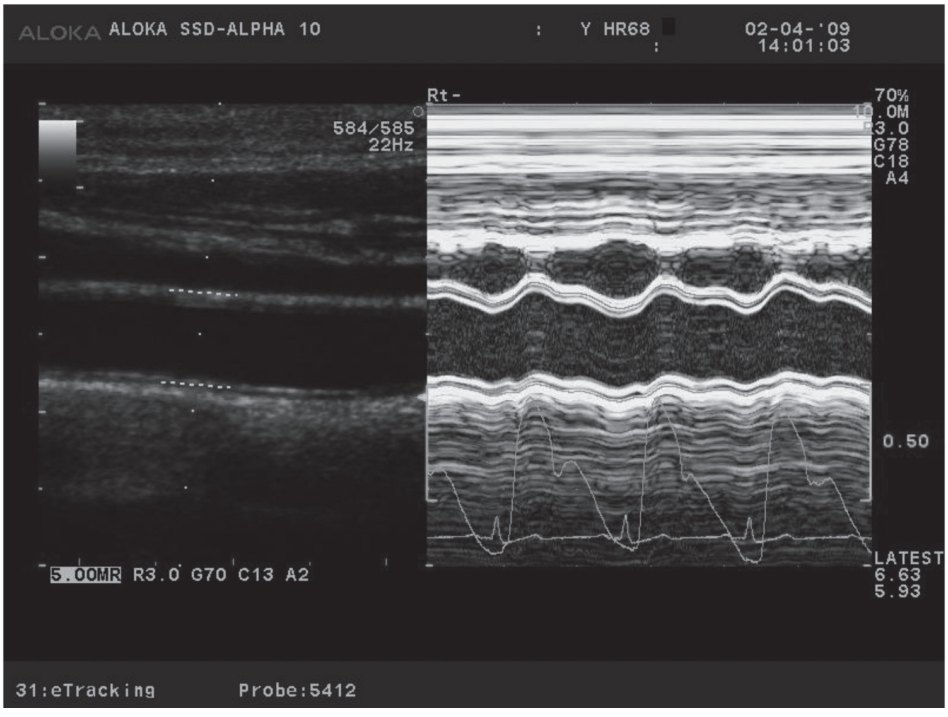

Figure 1 Ultrasound and echo-tracking of common carotid artery

where: Ad = the area under the BP diastolic decay curve from end-systole to end-diastole,
R = total peripheral resistance, Ps = the end systolic BP, Pd = the end-diastolic BP

- one-point pulse wave velocity (PWV), calculated from the time delay between two adjacent distension waveforms from water hammer equation for forward travelling waves with the usage of β-stiffness parameter
- AIx-augmentation index, calculated as the ratio between the augmentation pressure (AP) and PP (pulse pressure): **AIx=AP/PP**

Increased stiffness indexes as beta, EP and PWV and decreased parameter AC are related to stiffening of the vessel wall. Stiffness values were assessed at baseline and during each visit after RDN.

Ultrasound and echo-tracking system used for measurement of FMD is frequently used non-invasive ultrasound method for assessment of endothelial function. The measurement was performed on the right brachial artery. The sphygmomanometer BP cuff was placed on the forearm. Longitudinal image of brachial artery was obtained with a high resolution ultrasound probe and the interface between the near and far arterial wall was clearly defined. The cuff was inflated to ≥50mmHg above systolic pressure to occlude arterial flow for 5 minutes. After cuff deflation the maximal increase in diameter was measured continuously for 60 seconds during reactive hyperemia. The parameter FMD was reported as percentage change from baseline diameter (15). We undertook these ultrasound examinations before RDN and 6 months after the procedure.

Catheter-based renal denervation

A radiofrequency ablation catheter (Catheter Symplicity; Medtronic Ardian Inc., California, USA) was advanced into each renal artery via femoral access. Low-power radiofrequency treatments were applied along the length of both main renal arteries as describe elsewhere (4, 5). Patients were given heparin in i.v. bolus to achieve a prolonged activated clotting time. Intraprocedural diffuse visceral pain was managed with i.v. anxiolytics and narcotics (5). We did not observe serious intraprocedural or periprocedural complications. No important adverse events related to the procedure were noted in any of the treated patients during the follow-up.

Statistical analysis

Demographic characteristics, systolic BP, diastolic BP and all other analysed parameters were summarised with the descriptive statistics, including mean and SD for continuous variables, and frequency and percentage for categorical variables. Statistical significance of change in continuous

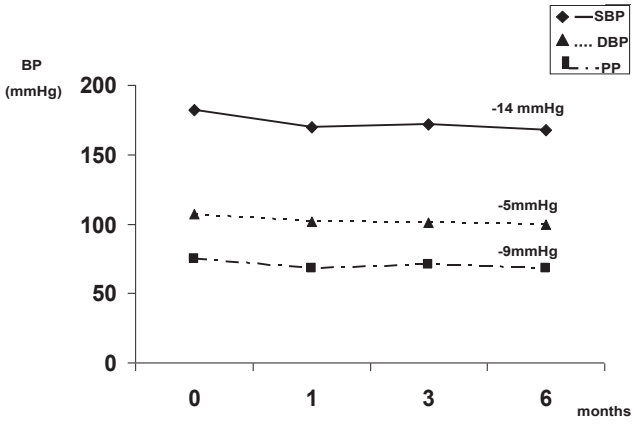

Figure 2 Changes in office systolic, diastolic and pulse pressure after renal sympathetic denervation
BP – blood pressure, SBP – systolic blood pressure, DBP – diastolic blood pressure, PP – pulse pressure

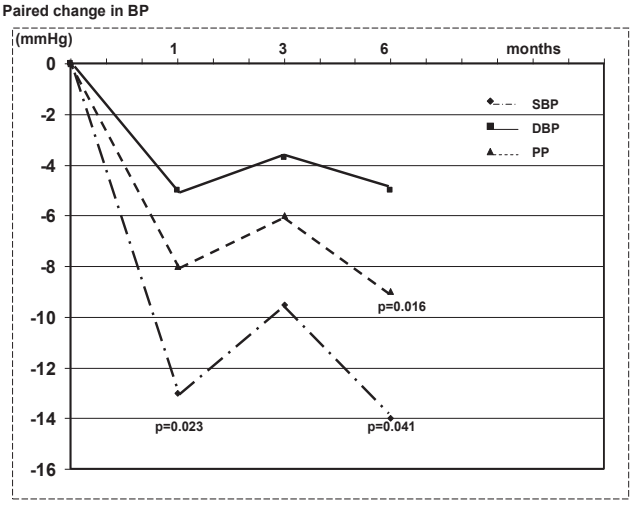

Figure 3 Paired changes in office systolic, diastolic and pulse pressure after renal sympathetic denervation
BP – blood pressure, SBP – systolic blood pressure, DBP – diastolic blood pressure, PP – pulse pressure

parameters in time was analyzed by mean of the Wilcoxon Signed Rank test for dependent data. The Spearman correlation coefficient was used to assess whether there was a relationship between variables. Using the linear regression model we examined the simultaneous effects of age, BMI, systolic BP, PP on the other analyzed parameters. All testing were two-sided tests with the criteria set at α=0.05. A p-value of <0.05 was considered statistically significant.

Results

Baseline characteristics

Table 1 presents baseline clinical characteristics of 17 patients with RH before RDN and at 6 months follow-up.

Table 1 Baseline characteristics and at 6 months follow up

Variables Renal denervation group	Baseline n = 17	6 month after RDN n = 14
Age ±SD, (years)	55 ± 9	
Men/Women	12/5	9/5
BMI kg/m^2	36 ± 6	
Type 2 diabetes mellitus	4 (23%)	
Hyperlipidemia	10 (59%)	
CAD	2 (12%)	
No. of antihypertensive drugs	6 (5 – 7)	6 (5 – 8)
Heart rate, bpm	67 ± 13	62 ± 10
eGFR mL/min per 1.73m^2	81.9 ± 19	78 ± 8
LVEF %	63 ± 9	62 ± 9

BMI – body mass index; CAD – coronary artery disease; eGFR – calculated glomerular filtration rate; LVEF – left ventricular ejection fraction; DD – diastolic dysfunction

RDN and BP

The office systolic and diastolic BP was reduced by 14/5±22/11 mmHg from 182/107±24/16 mmHg to 168/100±24/16mmHg at 6 months after the procedure **(Figure 2)**. The changes in office systolic BP were significantly improved at 1 month (-13±22 mmHg, p=0.023) and at 6 months after RDN (-14±25 mmHg, p=0.041). Significant changes in pulse pressure there were observed (PP) (9±11mmHg from 75±18mmHg at baseline to 68±17mmHg, p=0.016) at 6 months after RDN too **(Figure 3)**. The decrease in systolic BP of 10 mmHg was established in 6 patients (37.5%) after 1 month and in 7 patients (50%) after 6 months. We observed that in 4 patients (28.6%) after 6 months systolic BP did not fall.

RDN and carotid stiffness indexes

After RDN the parameters beta, EP and PWV decreased and AC increased on both sides of CCA, which is why we present the results of right CCA. Index Ep was significantly lower (p=0.039) and AC higher (p=0.030) 1 month after RDN. Values beta and Ep significantly decreased after 3 months too (beta: p=0.041; Ep: p=0.033). Levels of PWV showed slight decrease for 3 months after the procedure. We did not observe significant changes in AIx values **(Table 2)**.

Spearman's correlation coefficient was used to assess whether there are relationships between office BP and stiffness parameters at 6 months after RDN. We obtained positive univariant correlations between systolic BP and Ep (r=0.613, p=0.02; **Figure 4**), between systolic BP and PWV (r=0.559, p=0.03). Systolic BP negatively correlated with AC

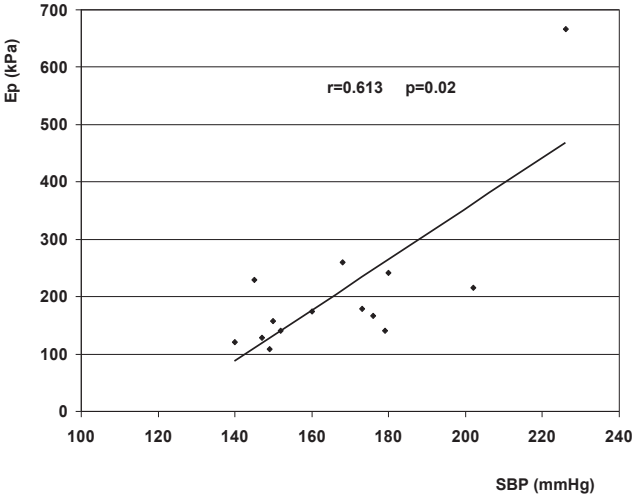

Figure 4 Relationships between systolic blood pressure and pressure-strain elasticity modulus
Ep – pressure-strain elasticity modulus, SBP – systolic blood pressure

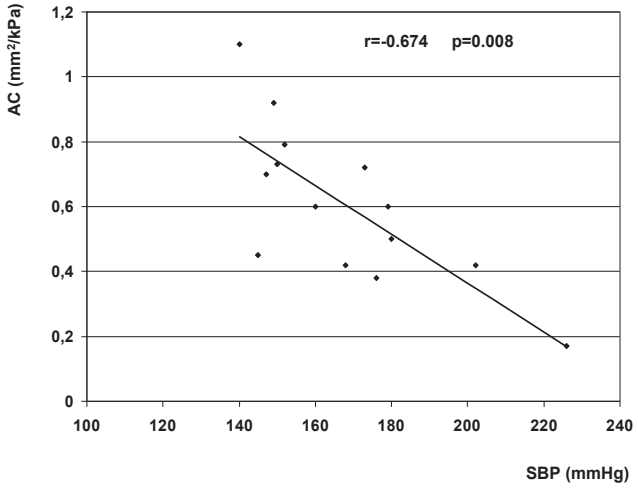

Figure 5 Relationship between systolic blood pressure and arterial compliance
AC – arterial compliance, SBP – systolic blood pressure

(r=-0.674, p=0.008; **Figure 5**). Similarly we found positive univariant correlations between diastolic BP and Ep (r=0.617, p=0.019), between diastolic BP and PWV (r=0.673, p=0.008) and negative correlations between diastolic BP and AC (r=-0.693, p=0.006). Changes of PP were related to Ep (r=0.737, p=0.003; **Figure 6**), and to PWV (r=0.701, p=0.005).

RDN and flow-mediated dilatation

Levels of FMD in diastole were significantly improved (3.87% versus 11.26%, p=0.01) at 6 months after RDN. We found slight improvement of FMD in systole (3.58% versus 10.52%, p=0.071). We did not obtain significant correlations between BP and FMD at 6 months follow-up.

Using linear regression model we examined the simultaneous effects of age, BMI, systolic BP, PP on stiffness values and FMD. Systolic BP was positively related to beta-stiffness and PWV. Systolic BP together with BMI were negatively related to AC. Systolic BP, BMI and age were positively correlated with Ep.

The fall in systolic BP >10mmHg would be associated with reduction of stiffness parameters beta >1.8%, Ep

Table 2 Changes and paired changes in carotid stiffness parameters after renal sympathetic denervation

					Paired change			P value		
Months	0	1	3	6	1	3	6	1	3	6
CCA right										
Beta	11.6	10.2	10.1	11.0	-1.4	-1.5	-0.6	0.162	0.041	0.530
(%)	±4.6	±3.1	±4.4	±6.6	±3.5	±3.3	±3.8			
Ep	216	181	180	209	-35	-36	-7	0.039	0.033	0.594
(kPa)	±80	±58	±90	±140	±58	±65	±83			
AC	0.56	0.67	0.68	0.61	0.11	0.12	0.05	0.030	0.069	0.167
(mm²/kPa)	±0.19	±0.20	±0.23	±0.24	±0.19	±0.23	±0.19			
Alx	-1.6	0.8	4.2	0.4	2.4	5.8	2.0	0.364	0.637	0.753
	±9.7	±0.8	±13.8	±0.4	±9.5	±16.8	±10.5			
PWV	8.5	8.0	7.9	8.4	-0.5	-0.6	-0.1	0.099	0.169	0.593
(m/s)	±0.9	±1.1	±1.5	±2.2	±1.1	±1.8	±2.2			

CCA – common carotid artery; Beta – beta stiffness; Ep – pressure-strain elasticity modulus; AC – arterial compliance; AIx – augmentation index; PWV – pulse wave velocity

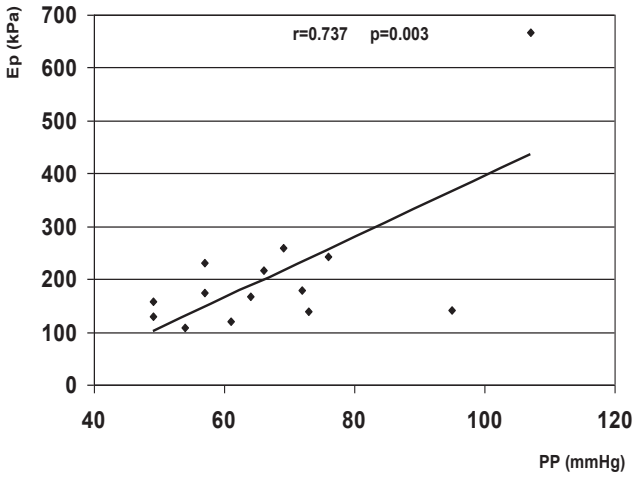

Figure 6 Relationship between pulse pressure and pressure-strain elasticity modulus
Ep – pressure-strain elasticity modulus, PP – pulse pressure

>55.92 kPa and PWV >0.7 m/s and with an increase of AC >0.009 mm²/kPa.

Discussion

In our group of patients with RH the catheter-based RDN was followed by an improvement of office BP, mainly of systolic BP and PP, one and six months after the procedure. Previous studies have reported the various BP lowering effect of RDN. The clinical efficacy of the procedure is mainly known from Symplicity Clinical Trial Program (8). To date Symplicity HTN-1 and Symplicity HTN-2 are the largest trials for this topic. A non-randomised Symplicity HTN-1 pilot trial involved 153 patients with mean office BP of 176/98±17/15 mmHg. The office BP values were reduced by 25/11 mmHg at 6 months and 32/14 mmHg 2 years after RDN. The Symplicity HTN-2 randomized controlled trial enrolled 106 patients randomized in 1:1 manner to RDN or control (without RDN) group. There was a demonstrated drop in BP 32/12 mmHg at 6 months follow-up. The BP reduction was significantly better in the RDN group than in the control group. The decrease in systolic BP >10 mmHg was achieved in 84% patients after 6 months (4, 5, 16). The Simplicity HTN-3 trial is a blind randomized controlled trial, but the results are still not available. Other single center studies mostly have included a small number of patients. Ott and co-workers (17) have demonstrated significant reduction in peripheral and central BP in 19 patients after RDN. There is evidence that this procedure is effective in some patients, but in other patients this type of treatment is without BP lowering effect (18). In contrast to this, in the other study (19) only a moderate effect on office BP was observed in 17 patients after RDN. It seems that the crucial question is the selection of patients. All patients need to fulfill the required criteria for RDN according to ESH/ESC guidelines 2013 (1).

The mechanisms of BP reduction after RDN are not completely known. Reduced sympathetic activity after RDN is the main recognized mechanism responsible for the BP lowering (20). It is known that the procedure is followed by reduced efferent sympathetic stimulation, by lowering of norepinephrine spillover, by reduced plasma renin activity, and by increased natriuresis and renal blood flow. The decrease in renal afferent signalling is associated with lowering of central sympathetic activity (18).

Human studies during the last years have shown that altered sympathetic activation may play a causal role in the development and progression of artery stiffening in RH. It was shown that aortic PWV and carotid distensibility are predictors of CV events in patients with hypertension (21, 22). Recent studies have reported that RDN had improved peripheral and central BP as well as arterial stiffness and CV outcome. Brandt and co-workers (6) found significantly reduced carotid-femoral PWV in 110 patients 6 months after RDN. Another study demonstrated in responders an improvement of two-point PWV by 13.7% 6 months after the procedure (7). Most of the studies have used the method of applanation tonometry for the measurement of arterial stiffness. Very few studies were done with the local carotid artery stiffness measurement using the ultrasound with echo-tracking method (23), but without an association with RDN. In our previous study (24) we found significantly elevated intima-media thickness and parameters of carotid artery stiffness in 50 patients with arterial hypertension. In this presented clinical work we have used ultrasound with echo-tracking for measurement of carotid stiffness parameters as well as for assessment of FMD values. The main advantage of the echo-tracking method is that carotid stiffness is determined directly from the change in pressure and the change in volume without the use modeling of the circulation (25), without indirect derivation as in other methods.

We have found significantly improved carotid stiffness parameters (beta, Ep, AC). We have affirmed the relationships between BP values and stiffness parameters at 6 months after RDN. According to these results we may suppose that the BP reduction together with modification of sympathetic tone after RDN may lead to attenuation of the carotid artery wall structural alterations.

Impaired endothelial function in hypertensive patients may be associated with other hypertensive manifestations such as thickening of the carotid artery wall (26). Experimental studies have reported that nitric oxide is a key signalling molecule involved in the sympathetic outflow from the brainstem (27).

Increased oxygen stress may directly activate or sensitize sympathetic neurones (28). Many clinical trials have shown the improved endothelial function assessed by FMD after pharmacological intervention (29, 30). In our study we have found significant improvement of FMD in diastole 6 months after RDN. We have not obtained correlations between BP and FMD. This fact is very interesting and worth further analysis. We may assume that the BP reduction was not the only factor responsible for improvement of FMD. The improvement of endothelial function probably represents a consequence of other factors, especially the decreased sympathetic activity caused by RDN.

Conclusion

We have evaluated the BP lowering effects of RDN on structural and functional parameters of artery wall in patients with RH. We have found significant changes in office systolic BP and PP, significantly decreased carotid stiffness values (beta, Ep and increased AC levels) during 6 months follow-up after RDN. We have detected the significant relationship between office BP and carotid stiffness values after 6 months follow-up. Endothelial function assessed on the basis of FMD in diastole was significantly improved also.

We conclude that in humans BP lowering effect of RDN (with a consequence of reduced sympathetic activity) is very likely to be followed by the improvement of structural and functional vessel wall parameters.

In order to confirm the effect of clinical RDN on vascular function changes in the hypertensive patients will require to be continued in follow-up, focusing on the duration of hypertensive disease and other variables.

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