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ENDOTHELIAL DYSFUNCTION AND 24-HOUR AMBULATORY BLOOD PRESSURE MONITORING IN PATIENTS WITH DIABETES

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Abstract

Arterial hypertension in patients with diabetes plays a main role in the earlier development of diabetic kidney disease (DKD), and endothelial dysfunction is considered to be a process involved in the development of diabetic complications. It is important to study the markers which gives the opportunity to identify DKD in early stage. In this article is presented a study of 24-hour Ambulatory Blood Pressure Monitoring (24hABPM) data in patients with diabetes and its interconnection with estimated glomerular filtration rate and endothelial dysfunction. The endothelial function was determined using the levels of TGF-b1(transforming growth factor-beta1) and VCAM-1 (vascular cell adhesion molecule-1). In our study, we obtained convincing results that complex including endothelial dysfunction and results of 24hABPM can be considered as early signs of DKD progression in patients with diabetes.

key words: *diabetic kidney disease, endothelial dysfunction, 24hABPM, estimated glomerular filtration rate.*

Background and aims

Diabetes mellitus (DM) is considered to be a disease accompanied by oxidative stress, chronic nonspecific inflammation, damage of the vessel wall, endothelial dysfunction (ED). DM is most often combined with arterial hypertension (AH). Insulin resistance (IR) is a common pathway for both – DM and AH. Endothelial dysfunction is the link between IR and cardiovascular disease [1]. AH in diabetic patients plays a main role in the earlier formation of diabetic kidney disease (DKD) - as an integral component of the cardiorenal continuum [2].

Albuminuria (AU) is the "gold standard" for screening of DKD. But, the structural and functional changes in kidneys are developing long before we can see AU. It is known, that 20-25% of nephrons are sclerosis at the stage of AU and from 50 to 70 % of nephrons - at the stage of proteinuria [3]. These changes point to irreversible kidney damage. It means the progressive reduction of kidney filtration and no therapy effect. Thus, the preclinical screening of DKD is highly important. It will give the opportunity to prescribe therapy earlier and slowing the progression of DKD. Today, there are many studies looking for markers of ED which allow to determine DKD in the early

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stages, until changes in the kidneys are reversible.

TGF-ß1 (transforming growth factor-beta1) is considered to be one of the main renal damage mediator. Studies have shown that TGF-β1 is an important mediator in development of chronic kidney disease (CKD). TGF-β1 helps the accumulation of extracellular matrix (ECM) in glomeruli (glomerular fibrosis, glomerulosclerosis) and in tubular interstitial (tubulointerstitial fibrosis) [4,5]. Glomerulosclerosis is the main cause of estimated glomerular filtration rate (eGFR) reducing. Three major types of glomerular cells (podocytes or visceral epithelial cells, mesangial and endothelial cells) are involved in the fibrotic process. The effect of TGF-ß1 caused by: apoptosis of podocytes (podocytopeniya), detachment by the basis of the glomerular membrane, hypertrophy of mesangial cells, epithelial-mesenchymal transformation (the formation of glomerular myofibroblast - the main source of ECM).

VCAM-1 (vascular cell adhesion molecule-1) is a mediator of the adhesion of circulating leukocytes (not related to neutrophils), monocytes, eosinophils, basophils to the endothelium. Also it is a mediator of transendotelial migration of these cells. ED is one of the first stages of atherogenesis, and VCAM-1 is considered as an early marker of atherosclerosis development [6]. It has proven that VCAM-1 is a reliable indicator of high risk of mortality from cardiovascular disease [7]. The elevated level of VCAM-1 is associated not only with coronary atherosclerosis but also with widespread atherosclerotic vascular damage. It has been shown, that elevated level of VCAM-1 is a predictor of CKD in patients with type 1 diabetes (T1DM) [8]. The development of CKD suggests the presence and progression of AH. Studies have shown that successful control of blood pressure prevents the rapid progression of vascular complications of diabetes and prolongs patient's life.

Thus, it is necessary to study the changes in 24-hour Ambulatory Blood Pressure Monitoring (24hABPM) in patients with DM. It is important to estimate the link between endothelial dysfunction and 24hABPM data as a predictor of DKD progression.

The aim of this study is to evaluate the link study between 24hABPM data and concentration of endothelial function markers in patients with DM and different eGFR.

Material and method

The study included 124 patients with diabetes, 66 patients with T1DM, 58 with T2DM. Patients were involved into the study in the endocrinology department of the "Clinic of the Medical Academy", Dnipro, 2016-2017. Among the patients - 68 women and 56 men, age from 18 to 64 (33 ± 5.6) years. Duration of DM - 3,5-25 (12 ± 6,3) years. The patients were divided into three groups depending on the eGFR: 1st group - eGFR 90 ml / min / 1.73 m² and above; 2nd group - eGFR 89-60 ml / min / 1.73m²; 3rd group - eGFR 59-45 ml / min / 1.73m The control group consisted of 20 healthy persons. All patients were comparable for age and sex.

Clinical and laboratory studies included: determination of serum creatinine level using the «SpinLab» (Ukraine), eGFR calculated by CKD-EPI (Chronic Kidney Disease Epidemiology Collaboration). The endothelial function was determined by TGF-b1 and VCAM-1 using the «Bender Medsystems» (Austria) and «BCM Diagnostics» (Ukraine). Study was conducted in Diagnostic Center of «Pharmacy of Medical Academy», Dnipro. 24hABPM was carried out by a portable automatic monitor "VAT41-2" (Ukraine). Blood pressure measurements were

taken every 15 minutes during the day and every 30 minutes during the night. The night interval was set between 10 PM to 7 AM. The following parameters, in 24hABPM readings, were taken into consideration to this study. Maximum of systolic blood pressure (max SBP), maximum of diastolic blood pressure (max DBP), total average systolic blood pressure (total average SBP); total average diastolic blood pressure (total average DBP), day systolic blood pressure > 140 mmHg in % (day SBP> 140 mmHg,%), day diastolic blood pressure > 90 mmHg in % (day DBP> 90 mmHg, %), night systolic blood pressure>120 mmHg in % (night SBP>120 mmHg, %), night diastolic blood pressure > 80 mmHg in % (night DBP> 80 mmHg, %), dipper status (DS) (extreme dippers are defined by a reduction in BP by more than 20% from day to night, dippers - reduction in BP of 10-20 % from day to night , non-dippers - reduction in BP by less than 10% from day to night, reverse dippers - decline in the nocturnal BP of <0 %. The day-time BP less than or equal to 140/90 mmHg and the night-time BP less than or equal to 120/80 mmHg were considered to be normal.

The study was conducted according to the ethical principles of the Helsinki Declaration with the permission of the Bioethical Commission of the SE "Dnipropetrovsk Medical Academy of the Ministry of Health of Ukraine".

Data were analyzed using Microsoft Excel, add-on program AtteStat and software STATISTICA 6.1 (StatSoftInc). The median (Me) and the interquartile range (25%, 75%) were used to describe the selective abnormal distribution of quantitative signs. To describe of the interconnections between different features was used a correlation analysis with the calculation of the Spearman rank correlation coefficients (ρ). Coefficient of correlation in the range $0.7 \leq |\rho| < 1$ indicated a strong connection of correlation; $0.3 \leq |\rho| < 0.7$ - for an average connection; $0 < |\rho| < 0.3$ - for a weak connection [10].

Results

Patients of all study groups had significantly higher ($p<0.05$) levels of TGF-b1 and VCAM-1 (Table 1) in comparison with the control group. It's demonstrate that patients with DM have endothelial dysfunction.

In all study groups, in T2DM levels of TGF-b1 and VCAM-1 were higher average by 1.3 and 1.21 times, respectively. It happens because of longer duration of T2DM, less strong control of DM and presents of arterial hypertension and microangiopathy in all patients. Levels of TGF-b1 and VCAM-1were increased with decreasing of eGFR regardless of type of diabetes. Thus, in T1DM level of TGF-b1 was higher for 57.0% and 78.7% in 2nd and 3rd group, respectively, than in 1st group. In T2DM level of TGF-b1 was higher for 30% and 64.2% in 2nd and 3rd group, respectively, than in 1st group. In T1DM level of VCAM-1 was higher for 16.2% and 46.2% in 2nd and 3rd group, respectively, than in 1st group. In T2DM level of VCAM-1 was higher for 18.3% and 47.4% in 2nd and 3rd group, respectively, than in 1st group.

It was shown that differences between levels of TGF-b1 ($p1\text{-}2 = 0.001$; $p1\text{-}3 < 0.001$) and VCAM-1 ($p1\text{-}2 = 0.001$; $p1\text{-}3 < 0.001$) in the study groups were the statistically significant. It was determined that levels of TGF-b1 and VCAM-1 have by reverse strong correlation with eGFR ($\rho = -0.56$, $p < 0.001$) and ($\rho = -0.51$, $p < 0.001$) respectively. The less eGFR, the higher rates of TGF-b1 and VCAM-1, which indicates the progression of endothelial dysfunction.

The results of 24hABPM of patients of study groups are presented in Table 2.

In the 1st group it was determine the following features of 24hABPM (Table 2). There were no patients with hypertension in the

T1DM. But, according to the deeper status, it was determined that 28.0% of patients were "extreme-deeper"; 9.5% - "non-deeper", 62.5% - "deeper". There were 38.8% of patients with hypertension in the T2DM. According to the deeper status it was determined that 33.0% of patients were "non-deeper"; 16.0% - "reverse-deeper", 51.0% - "deeper". The significant differences between T1DM and T2DM of 1st group were determined by the: max. SBP (p <0.001); max DBP (p <0.001); day SBP> 140 mmHg (p <0.001). These results show that T2DM patients with normal or elevated eGFR have AH and more expressed changes in circadian rhythm. This can be explained by a combination of insulin resistance, hyperinsulinemia and atherosclerotic vascular damage in patient with T2DM. All these factors potentiate each other's and lead to more earlier appearance of hypertension versus T1DM.

Table 1. Differences in TGF-b1 and VCAM-1 in patients of study groups and control

Indicators Me (25 %;75 %)	T1DM	T2DM	*p*	Control group	*p*
1st Group (eGFR ≥ 90 ml / min / 1.73 m²)					
TGF-b1, ng/ml	16332,34 (13422,76; 18015,52)	28934,6 (17118,26; 39332,34)	*0,003*	9526,572 (5726,05; 11557,6)	p_{1-2}= *0,001;* p_{1-3}< *0,001*
VCAM-1, pg/ml	674,54 (539,97; 764,75)	811,75 (740,45; 901,92)	*0,004*	551,2272 (493,914 717,593)	p_{1-2}= *0,001;* p_{1-3}< *0,001*
2nd Group (eGFR 89-60 90 ml / min / 1.73 m²)					
TGF-b1, ng/ml	37998,89 (29927,94; 39147,93)	41633,59 (39664,41; 48309,88)	*<0,001*	9526,572 (5726,05; 11557,6)	p_{1-2}= *0,001;* p_{1-3}< *0,001*
VCAM-1, pg/ml	805,04 (698,73; 982,51)	993,6223 (796,69; 1213,26)	*<0,001*	551,2272 (493,914 717,593)	p_{1-2}= *0,001;* p_{1-3}< *0,001*
3rd Group (eGFR 59-45 ml / min / 1.73 m²)					
TGF-b1, ng/ml	76838,11 (61207,08; 82426,8)	80840,4 (71987,8; 87414,63)	*<0,001*	9526,572 (5726,05; 11557,6)	p_{1-2}= *0,001;* p_{1-3}< *0,001*
VCAM-1, pg/ml	1255,708 (966,53; 1520,31)	1544,27 (1201,13; 1865,97)	*<0,001*	551,2272 (493,914 717,593)	p_{1-2}= *0,001;* p_{1-3}< *0,001*

Note: *p* – is the difference between subgroups of different types of DM according to Mann-Whitney criterion;
p_{1-2} - is the difference between a subgroup of T1DM and a control group;
p_{1-3} - is the difference between a subgroup of T2DM and a control group.

The multivariate logistic correlations were determined with the level of TGF-b1, VCAM-1 and 24hABPM data. In 1st group level of TGF - b1 has reverse average correlation with deeper status (ρ = -0,32; p = 0.047); direct average correlation with total average SBP (ρ = 0,4; p = 0,013), max SBP (ρ = 0,48; p = 0,002), max DBP (ρ = 0,32; p = 0,045), day SBP > 140 mmHg (ρ = 0,5; p = 0,001); (ρ = 0,39; p = 0,013), night SBP > 120 mmHg (ρ = 0,38; p = 0,018), night DBP > 80 mmHg (ρ = 0.36; p = 0.023). Correlation analysis demonstrate the differences in the level of TGF-b1 between T1DM and T2DM (ρ = 0.48; p = 0.002).

It was shown that level of VCAM-1 has reverse average correlation with deeper status (ρ=-0,48; *p=0,002*), direct average correlation with total average SBP (ρ=0,39; *p=0,015*), max

SBP (ρ=0,45; *p=0,004*), max DBP (ρ=0,44; *p=0,004*), night SBP > 120 mmHg (ρ=0,4; *p=0,005*), day SBP > 140 mmHg (ρ=0,46; *p=0,011*).

Table 2. Results of 24hABPM of study groups depending on estimated glomerular filtration rate (eGFR)

Indicators Me (25 %;75 %)	1st Group (eGFR ≥ 90 ml / min / 1.73 m²)			2nd Group (eGFR 89-60 90 ml / min / 1.73 m²)			3rd Group (eGFR 59-45 ml / min / 1.73 m²)			Comparasion between groups
	T1DM	T2DM	*p*	T1DM	T2DM	*p*	T1DM	T2DM	*p*	
Total average SBP	124 (122; 130)	134 (124; 144)	*0,005*	126 (122; 136)	137,5 (128; 147)	*0,001*	143 (137; 147)	146 (140; 154)	*0,15*	p_{1-2}*>0,05;* p_{1-3}*<0,001;* p_{2-3}*<0,001*
Total average DBP	78 (75; 84)	80,5 (72; 86)	*0,693*	82 (78; 86)	87,5 (83; 93)	*0,003*	89,5 (86; 95)	92,5 (88; 97)	*0,192*	p_{1-2}*=0,015;* p_{1-3}*<0,001;* p_{2-3}*=0,001*
Max SBP	135 (133; 141)	159,5 (151; 168)	*<0,001*	142 (137; 146)	162 (154; 169)	*<0,001*	165,5 (157; 182)	171 (158; 183)	*0,634*	p_{1-2}*>0,05;* p_{1-3}*<0,001;* p_{2-3}*<0,001*
Max DBP	90 (87; 92)	96 (94; 99)	*<0,001*	92 (88; 96)	101 (97; 104)	*<0,001*	102 (98; 111)	102 (98; 112)	*0,86*	p_{1-2}*>0,05;* p_{1-3}*<0,001;* p_{2-3}*<0,001*
Day SBP> 140 mmHg,%	0 (0; 4)	14,55 (2,7; 22,69)	*<0,001*	5 (0; 12,4)	33,7 (13,48; 52,7)	*<0,001*	33,45 (24; 38,4)	36,9 (33,2; 54)	*0,112*	p_{1-2}*=0,009;* p_{1-3}*<0,001;* p_{2-3}*<0,001*
Day DBP> 90 mmHg,%	1,2 (0; 5)	4,93 (2,3; 16)	*0,022*	0 (0; 4,9)	24,25 (6,4; 39,1)	*<0,001*	27,75 (19,4; 36,4)	36,8 (27,6; 47,4)	*0,084*	p_{1-2}*>0,05;* p_{1-3}*<0,001;* p_{2-3}*<0,001*
Night SBP>120 mmHg, %	0 (0; 2)	2,86 (0; 27)	*0,099*	3,8 (0; 15,4)	51,4 (34,2; 87)	*<0,001*	31,36 (22,5; 42)	33,85 (27,1; 54,2)	*0,289*	p_{1-2}*<0,001;* p_{1-3}*<0,001;* p_{2-3}*>0,05*
Night DBP>80 mmHg, %	0 (0; 1,51)	3,13 (0; 18)	*0,032*	1,8 (0; 19,3)	38,5 (9,64; 60,32)	*<0,001*	31,05 (23,8; 48)	38,05 (25,4; 49,1)	*0,308*	p_{1-2}*=0,001;* p_{1-3}*<0,001;* p_{2-3}*=0,004*

Note. *p* - differences between T1DM and T2DM subgroups according to the Mann-Whitney criterion; comparison using the nonparametric dispersion analysis of Kruskal-Wallis

In the 2nd group there were established the following features of the 24hABPM. In the T1DM 26% of patients had hypertension. 8,6% patient of this patients were "extreme-deeper", 34,7% - «non-deeper», 52,0% - «deeper», 4,7% - «reverse- deeper». There were demonstrated the significant differences (*p <0,001*) between T1DM and T2DM of 2nd group by the: total average SBP, total average BBP, max SBP, max DBP, day SBP > 140 mmHg, day DBP > 90 mmHg, night SBP > 120 mmHg, night DBP > 80 mmHg. These rates were higher in patients with T2DM.

In 2nd group we also estimated that level of TGF -b1 has: reverse correlation with deeper status (ρ=-0,44; *p=0,003),* direct average correlation with max SBP (ρ=0,38; *p=0,010*), max DBP(ρ=0,4; *p=0,007*), day SBP > 140 mmHg (ρ=0,34; *p=0,022*), day DBP > 90 mmHg (ρ=0,34; *p=0,024*), night SBP > 120 mmHg (ρ=0,47; *p=0,001*). As a result, we have an evidence of the differences in the level of TGF-b1 between T1DM and T2DM (ρ=0,59; *p<0,001*).

It was determined that level of VCAM-1 has reverse average correlation with DS (ρ=-0,48; *p=0,002*), direct average correlation with total average SBP (ρ=0,39; *p=0,015*), max SBP (ρ=0,45; *p=0,004*), max DBP (ρ=0,44; *p=0,005*), day SBP > 140 mmHg (ρ=0,46; *p=0,004*), night SBP > 120 mmHg (ρ=0,4; *p=0,011*).

In the 3rd group there were showed the following features of the 24hABPM. In the T1DM 63,6% of patients had hypertension. 59% patient of this patients were «non-deeper», 22,7 % - «reverse- deeper», 18,3% - «deeper». In the T2DM 94,44% of patients had hypertension. 66,6% patient of this subgroup were «non-deeper», 33,3% - «reverse- deeper», 0,01% - «deeper». There were no evident differences of 24hABPM indicators between T1DM and T2DM patients. Results of 24hABPM significantly worsened with the decreased of eGFR. In T1DM, in 3rd group, the number of patients with AH was 63.6%, while in 1st group there no patients with hypertension. In 3rd group, the number of "non-deeper" was more for 83.8%, and the number of "deeper" was less for 70% than in 1st group. The activation of renin-angiotensin-aldosterone system (RAAS) leads to increased reviling of AH and changes in circadian rhythm in patients with T1DM. All this conducted to DKD progression.

The multivariate logistic correlation was determined with TGF -b1, VCAM-1 and 24hABPM data in the 3rd group. The level of TGF -b1 has: reverse average correlation with deeper status (ρ=-0,43; *p=0,006*), direct average correlation with total average SBP (ρ=0,47; *p=0,002*) and total average DBP (ρ=0,51; *p=0,001*).

It was evaluated that level of VCAM-1 has reverse average correlation with DS (ρ=-0,48; *p=0,002*), direct average correlation with max SBP (ρ=0,36; *p=0,024*), night SBP > 120 mmHg (ρ=0,37; *p=0,020*), night DBP>80 mmHg (ρ=0,32; *p=0,045*).

Discussion

Most authors consider changes of circadian rhythm of BP as a marker of target organs being damaged [11]. In our study, it was shown that the results of 24hABPM of patients with DM were expressed in progression changes of circadian rhythm according to the decline of eGFR. The results of the study have demonstrated that the more decline of eGFR, the more number of patients having insufficient decrease of nocturnal BP. This process didn't depend on the type of diabetes but was more obvious in patients with T2DM (T1DM - 43.8% vs T2DM - 74.1%).

Today most researchers consider the factors of endothelial function as possible early markers of DKD progression [12-14]. Important, that we determined the elevated levels of TGF-b1 and VCAM-1 in patients with DM without clinical signs of DKD. And there were significant differences between patients with DM and healthy ($p < 0,005$). In addition to, it was found the significant differences of levels of TGF-b1 and VCAM-1 between study groups ($p_{1-2}= 0,001$; $p_{1-3}< 0,001$). Thus, we have shown that the increase TGF-b1 and VCAM-1 concentration were an independent factor of ED aggravation the likelihood of DKD.

Conclusion

The results of the study show that significant (p <0.005) changes of circadian rhythm in patients with diabetes in the early stages of DKD (stage of hyperperfusion and normal or elevated eGFR) determined by 24hABPM. In patients with T1DM and T2DM it was shown an increase number of «non-dipper» from 7.5% to 27.5% and «reverse-dipper» from 20% to 60.5% depending on the eGFR decline: in group of patients with eGFR ≥90 ml / min / 1, 73m³ and with GFR 59-45 ml / min / 1.73m³, respectively. Lowering of blood pressure at night in patients with T1DM and T2DM was insufficient: deeper status (p = 0.018). That confirm the progression of DKD in these patients. In the study, we obtained convincing results that the levels of markers of endothelial function - TGF-b1 and

VCAM-1 in patients with T1DM and T2DM were quantitatively dependent on eGFR. The levels of TGF-b1 and VCAM-1 in patient with DM and DKD (stage of hyperperfusion and elevated levels of eGFR) significantly differed ($p < 0.005$) from the control. The strong and average correlations were detected between levels of TGF-b1, VCAM-1 and results of 24hABPM. We can conclude that complex including results of 24hABPM and endothelial dysfunction can be considered as early signs of DKD progression.

REFERENCES

1. **Tushar PP, Komal R, Ashim KB et al.** Insulin resistance: an additional risk factor in the pathogenesis of cardiovascular disease in type 2 diabetes. *Heart Fail Rev* 21(1): 11-23, 2016.

2. **Statsenko ME, Derevyanchenko MV.** Cardiorenal pathology in patients with arterial hypertension and type 2 diabetes mellitus. *Russian days of the heart. Materials of the Fifth International Educational Forum*: 98, 2017 (abstract)

3. **Ribal'chenko BM.** Early diagnosis of diabetic kidney disease in patients with type 2 diabetes mellitus. *Endocrinology*: 344, 2014 (abstract)

4. **López-Hernández FJ, López-Novoa JM.** Role of TGF-β in chronic kidney disease: an integration of tubular, glomerular and vascular effects. *Cell Tissue Res* 347(1): 141-54, 2012

5. **Meng XM, Nikolic-Paterson DJ, Lan HY.** TGF-β: the master regulator of fibrosis. *Nat Rev Nephrol* 12(6): 325–338, 2016

6. **Gimbrone MA, García-Cardeña G.** Endothelial Cell Dysfunction and the Pathobiology of Atherosclerosis. *Circ Res* 118(4): 620–636, 2016.

7. **Blankenberg S, Rupprecht HJ, Bickel C et al.** Circulating cell adnesion molecules and death in patients with coronary artery disease. *Circulation* 104(12): 1336-1342, 2001.

8. **Triñanes J, Salido E, Fernández J et al.** 1 Diabetes Increases the Expression of Proinflammatory Cytokines and Adhesion Molecules in the Artery Wall of Candidate Patients for Kidney Transplantation. *Diabetes Care* 35(2): 427–433, 2012.

9. **Medvedeva M. S., Mukhina D. D., Tarasov A. N.** Diagnostic significance of daily monitoring of blood pressure in type 2 diabetes mellitus. *ACADEMY* 6: 89–92, 2017.

10. **Rebrova OY.** Statistical analysis of medical data. Application package STATISTICA. Media-Sfera, Moskow 312, 2006.

11. **Aksit E, Gursul E, Aydin F, Samsa M, Ozcelik F.** Non-dipper hypertension is associated with slow coronary flow among hypertensives with normal coronary angiogram. *Cardiovasc Journal Afr* 28(1): 14–18, 2017.

12. **MacIsaac RJ, Ekinci EI, Jerums G.** Markers of and Risk Factors for the Development and Progression of Diabetic Kidney Disease. *Am J Kidney Dis* 63(2): 39-62 2014.

13. **Futrakul N, Chanakul A, Futrakul P, Deekajorndech T.** Early stage of vascular disease and diabetic kidney disease: an underrecognized entity. *Ren Fail* 37(8): 1243-1246 2015.

14. **Rossi GP, Seccia TM, Barton M, Danser AHJ et al**. Endothelial factors in the pathogenesis and treatment of chronic kidney disease Part II: Role in disease conditions a joint consensus statement from the European Society of Hypertension Working Group on Endothelin and Endothelial Factors and the Japanese Society of Hypertension. *J Hypertens* 36(3): 462-471, 2018.