ENGLISH VERSION: INHERITANCE OF POLYMORPHIC GENOTYPES OF THE ENDOTHELIN-1 GENE AND INDICATORS OF CARDIAC AND SYSTEMIC HEMODYNAMICS IN MEN WITH ESSENTIAL HYPERTENSION, RESIDENTS OF PODILLYA*

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The parameters of intracardiac and systemic hemodynamics in male citizens of Podillia region in Ukraine aged 40-60 with different genotypes of the ET-1 gene were studied. The study involved 141 male citizens of Podillia region in Ukraine aged 40-60. Among them 62 men were diagnosed with uncomplicated EH with LVH, whose average age was 49,19±0,66 and 79 healthy men whose age (49.01±0.73) did not differ from patients with uncomplicated EH with LVH and made the control group (p>0.05). Genotyping of the ET-1 gene was conducted using polymerase chain reaction. The ET-1 concentration in plasma was determined by using ELISA. It was found that Lys/Lys genotype and the Lys allele of the ET-1 gene dominate among control group and patients with uncomplicated EH and LVH. In patients with uncomplicated EH and LVH all indicators of structural-functional state of the myocardium, except the size of LA, SI, DT was significantly greater in carriers of the allele Asn, than in homozygote carriers of Lys genotype of the ET-1 gene, indicating a possible negative prognosis in these patients for the development of chronic heart failure. CHLV occurs significantly more often than EHLV in men with uncomplicated EH and LVH without dependence on the genotypes of the ET-1 gene.

Key words: essential hypertension, gene polymorphism of endothelin-1, cardiac and systemic hemodynamics, plasma concentration of endothelin-1

Introduction.

It is well known that essential hypertension (EH) is a multifactorial disease, and genetic conditioning plays one of the key roles in it. According to the Global human genome, 30 chromosomal loci are already known to be involved in regulation of blood pressure (BP), and this number may increase in the future. One of the genes that can influence BP is the endothelin-1 gene (ET-1). It is established that level of vasoconstriction, myocardial contractility, the value of preload, afterload depend on concentration of ET-1. Important property of ET-1 is its ability to "run" intracellular mechanisms that lead to increased protein synthesis and the development of left ventricular hypertrophy (LVH). In addition, ET-1 promotes collagen synthesis in the heart muscle and the development of cardiac fibrosis [13-15]. This study provokes investigation of Single nucleotide polymorphism (SNP) of the ET-1 gene, because this factor influences the expression of the peptide. In Ukraine and in the world there are many research works about the structural and functional parameters of the heart in patients with EH but the studies about these changes in patients with gene polymorphisms of ET-1 have not been performed yet.

The aim of the research. To study the parameters of intracardiac and systemic hemodynamics in male citizens of Podillia region in Ukraine aged 40-60 with different genotypes of the ET-1 gene.

Materials and methods

The study involved 141 middle-aged male residents of Podillia region in Ukraine. Among them 62 men from the main group were diagnosed with uncomplicated EH with LVH with preserved systolic function and chronic heart failure (CHF) I classes according to NYHA Classification, whose average age was 49.19±0.66 and 79 healthy men whose age (49.01±0.73) did not differ from patients with uncomplicated EH with LVH and made the control group (p>0.05). The diagnosis of EH was established on the basis of patients' complaints, anamnesis, physical examination, laboratory and instrumental methods of investigation according to the guideline of the European Society of Hypertension (ESH) and the European Society of Cardiology (ESC) in 2013 in accordance with Unified clinical protocol of medical care of hypertension, approved by the Ministry of Health of Ukraine from 24.05.2012 year №384 [5,12]. All patients during the examination were treated at Vinnytsia regional specialized dispensary of radiation protection of the Ministry of Public Health of Ukraine, Military Medical Center of the Central Region of Air Force of Ukraine and were observed from December 2013 to June 2015.

Exclusion criteria of the study were as follows: secondary hypertension, renal and liver dysfunction, coronary heart disease the onset of which was before EH, endocrine, hematological, neoplastic and autoimmune disorders, patients with EH complications: myocardial infarction, acute cerebrovascular accident. Genotyping of the ET-1 gene was conducted using polymerase chain reaction (PCR). This study was carried out jointly with the Research Institute of the genetic and immunological bases of pathology and pharmacokinetics "Ukrainian Medical Stomatological Academy" (headed by Prof. I.P. Kaidashev, Poltava). The ET-1 concentration in plasma was determined by using ELISA method on enzyme-linked immunosorbent analyzer "Humareader single» (Germany). To determine the ET-1 plasma concentration a standard set of the firm «DRG» (USA) was used. We used ultrasound of the heart for assessing the hemodynamic parameters, which was carried out on «RADMIR ULTIMARA». The mathematical processing was performed on a personal computer using a standard statistical package STATISTICA 6.0. The frequency distribution of gene polymorphisms in the population was undergone a checking according to Hardy-Weinberg equilibrium law using a calculator gene expert to calculate the number of

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statistical parameters in the study "case-control" which using SNP.

Results and discussions

It was established that Lys/Lys genotype of the ET-1 gene was in 65.82% men from the control group (n=52), Lys/Asn genotype – 27.85% (n=22) and Asn/Asn genotype – 6.33% (n=5) ($p_{Lys/Asn-Lys/Lys}$<0.00001; $p_{Asn/Asn-Lys/Lys}$<0.00001; $p_{Asn/Asn-Lys/Asn}$<0.001). The frequency of the Lys allele in male from the control group was 79.75%, the Asn allele – 20.25% ($p_{Lys-Asn}$<0.00001). It was investigated that Lys/Lys genotype of ET-1 dominates in patients from the main group – 56.45% (n=35), Lys/Asn genotype was in 33.87% (n=21) patients and Asn/Asn genotype – 9.68% (n=6) ($p_{Lys/Asn-Lys/Lys}$<0.01; $p_{Asn/Asn-Lys/Lys}$<0.00001; $p_{Asn/Asn-Lys/Asn}$<0.001). The frequency of the Lys allele in male from the main group was 73.39%, the Asn allele – 26.61% ($p_{Lys-Asn}$<0.00001). Because of the low frequency of carriers of Asn/Asn genotype men with Lys/Asn and Asn/Asn genotypes in each group were united as carriers of the Asn allele. The frequency of the Asn allele in the male control group is 34.18% (n=27) ($p_{Asn\ allele-Lys/Lys}$<0.00001), in the main group is 43.55% (n=27) ($p_{Asn\ allele\ -Lys/Lys}$>0.05).

Comparing the frequencies of genotypes and alleles of the ET-1 gene in the control group and among patients with uncomplicated EH and LVH it was found that no significant differences were defined between these groups (p>0.05).

According to the results obtained in a Japanese study in healthy individuals of white and Japanese race, it was demonstrated that men and women (mixed group on gender) had Lys/Lys genotype of the ET-1 gene significantly more often [10]. In healthy American Negro and white races with burdened heredity by cardiovascular diseases dominates the Lys allele [16]. L.O. Minushkina [2008] showed that among patients of both sexes with both complicated and uncomplicated EH, residents of Yakutia, there was a high frequency of Lys/Lys genotype of the ET-1 gene carriers than among patients with EH, Moscow residents [3]. However, the analysis of frequency distribution of genotypes polymorphism of the ET-1 gene in men residents of Kazakhstan has shown that Lys/Lys genotype in patients with EH occurs 1,3 times less than in healthy individuals. Heterozygous Lys/Asn variant is equally common in patients with EH and in control group. Asn/Asn genotype was identified only in patients with EH [11]. Mutation of the ET-1 gene (Lys198Asn) leads to increased blood pressure in young people of both sexes especially overweight living in Samara (Russia) [2].

Then we have analyzed the state of intracardiac and systemic hemodynamics in the control group and in patients with uncomplicated EH and LVH. Patients with uncomplicated EH and LVH have significantly higher size cavities of the heart – the end diastolic diameter (EDD), end systolic diameter (ESD) and the volume of the heart - end diastolic volume index (EDVI) and end systolic volume index (ESVI) than in the control group (p<0.001). Left ventricular mass index (LVMI) was significantly higher in patients with uncomplicated EH and LVH than in men without cardiovascular diseases (138.30±4.07 g/m^2 and 79.73 ± 1.73 g/m^2, respectively, p<0.001).

The parameters of transmitral flow (TF) in patients with uncomplicated EH with LVH were significantly different from those in men from the control group. The E/A ratio was higher and the deceleration time (DT) was lower in men without cardiovascular diseases (p<.001). The isovolumic relaxation time (IVRT) was longer in patients with uncomplicated EH and LVH than in men from the control group (p□0.001). The next step was to determine the types TF in men with uncomplicated EH and LVH. It was investigated that men with uncomplicated EH and LVH have significantly higher level of the normal type of TF - 67,74% (n=42) than the hypertrophic type (29.03% (n=18) and pseudonormal type (3.23% (n=2) of TF (p<0.001).

Assessment of central hemodynamics has shown that systolic blood pressure (SBP), diastolic blood pressure (DBP) and heart rate (HR) significantly higher in men with uncomplicated EH and LVH than in the control group. It should be noted that patients with uncomplicated EH and LVH have significantly higher cardiac index (CI) and systemic vascular resistance (SVR) than in patients from the control group (p<0,001).

The abovementioned data is similar to literature results [1,6,8,9].

Concentric LVH (CLVH) occurs significantly more often (70.97% (n=44) than eccentric LVH (ELVH) (29.03% (n=18), with p<.001. Geometric changes of the myocardium in patients with essential uncomplicated EH and LVH correspond to the data of other researchers [1,4,6,7,9].

As mentioned above, ET-1 has an effect on the structural and functional performance of the heart. Therefore, the next stage of research was to determine parameters of intracardiac and systemic hemodynamics in individuals from the control group and patients with uncomplicated EH and LVH carriers of different variants of the ET-1 gene (Fig.1,2,3).

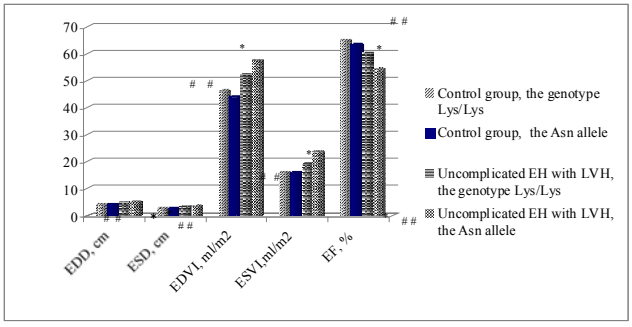

Fig. 1. Indicators of structural and functional changes in myocardium in male control group and patients with uncomplicated EH and LVH, the carriers of different variants of the ET-1 gene

Notes: The difference is statistically significant when compared with:
**- genotype Lys/Lys within each group (p<0.001)*
 # - patients with uncomplicated EH and LVH within each genotype (p<0.001)

It was found that men from the control group, carriers of the Asn allele have significantly higher rates of SVR, DBP and HR than carriers of genotype Lys/Lys. No differences in other indicators were found. All indicators of intracardiac and systemic hemodynamics were significantly greater in men with uncomplicated EH and LVH – carriers of the Asn allele, than in homozygote carriers of Lys genotype of the ET-1 gene.

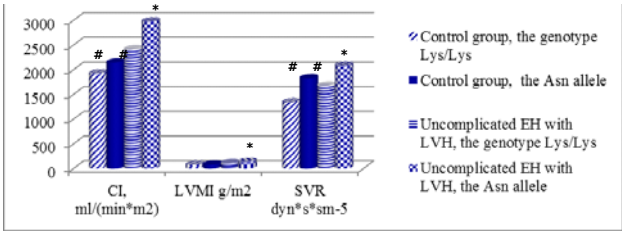

Fig. 2. Indicators of structural and functional changes in myocardium in the control group and patients with uncomplicated EH and LVH, the carriers of different variants of the ET-1 gene

Notes: The difference is statistically significant when compared with:
** - genotype Lys/Lys within each group,*
 # - from patients with uncomplicated EH and LVH within each genotype (p<0.001)

It was found that the hemodynamic parameters are higher in patients with uncomplicated EH and LVH than in patients without cardiovascular diseases, carriers of all genotypes of the ET-1 gene. The results demonstrated that patients with uncomplicated EH and LVH, carriers of the Asn allele are associated with worse hemodynamic performance than carriers of the genotype Lys/Lys.

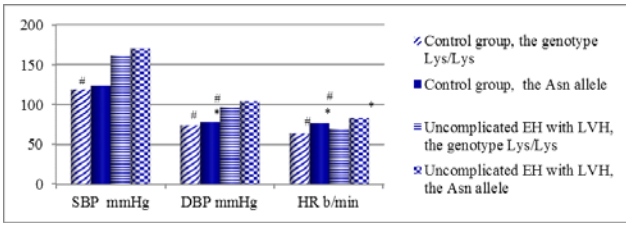

Fig. 3. Indicators of SBP, DBP, HR in male control group and patients with uncomplicated EH and LVH, (%)

Notes: The difference is statistically significant when compared with:
** - genotype Lys/Lys within each group (p<0.001)*
 # - patients with uncomplicated EH and LVH within each genotype (p<0.001)

Investigation of the frequency of TF in patients with uncomplicated EH and LVH, carriers of different genotypes of the ET-1 gene, revealed that carriers of genotype Lys/Lys and the Asn allele significantly more common normal type of TF. It was found that no significant differences were defined between carriers of different genotypes of the ET-1 gene (Table 1).

Table 1

Types of transmitral blood flow in men with uncomplicated EH and LVH, carriers of different variants of genotypes of the ET-1 gene, (%)

Groups	Men with a normal type of TF		Men with a hypertrophic type of TF		Persons with a pseudonormal type of TF		p
	1. Carriers of genotype Lys/Lys	2. Carriers of the Asn allele	3. Carriers of genotype Lys/Lys	4. Carriers of the Asn allele	5. Carriers of genotype Lys/Lys	6. Carriers of the Asn allele	
Men with uncomplicated EH and LVH (n=62)	62.86% (n=22)	74.07% (n=20)	34.29% (n=12)	22.22% (n=6)	2.86% (n=1)	3.7% (n=1)	p_{2-1}<0.05 p_{4-3} <0.05 p_{6-5}<0.05 p_{3-1}<0.01 p_{5-1}<0.01 p_{4-2}<0.01 p_{5-2}<0.01

Analysis of LV geometry in patients with uncomplicated EH and LVH, carriers with different genotypes of the ET-1 gene showed the following. In men with uncomplicated EH and LVH, carriers of all genotypes of the ET-1 gene CHLV occurs significantly more often than EHLV (p□0,01). No significant differences in the frequency of CHLV and EHLV between carriers of different genotypes of the ET-1 gene were detected (Fig. 4).

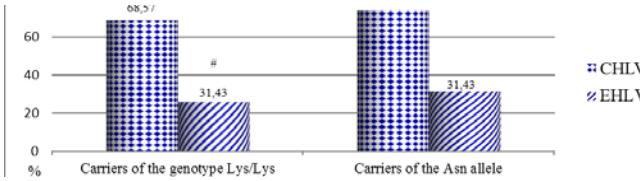

Fig. 4. Variants of LVH in men with uncomplicated EH and LVH, carriers of different genotypes of the ET-1 gene (%)

Notes: The difference is statistically significant when compared with: # - CHLV

Similar studies of structural and functional changes in the heart in carriers of different genotypes of the ET-1 gene in the world and in Ukraine have not been performed yet.

Conclusions.

1. Lys/Lys genotype and the Lys allele of the ET-1 gene dominate among men without cardiovascular diseases and patients with uncomplicated EH and LVH, residents of Podillia region in Ukraine, aged 40-60 years.

2. In patients with uncomplicated EH and LVH all parameters of structural-functional state of the myocardium, except the size of LA, SI, DT was significantly greater in carriers of the allele Asn, than in homozygote carriers of Lys genotype of the ET-1 gene, indicating a possible negative prognosis in these patients for the development of chronic heart failure.

3. CHLV occurs significantly more often than EHLV in men with uncomplicated EH and LVH without dependence on genotypes of the ET-1 gene.

References:

1. Bagriy V.V. Uncomplicated hypertension in women, clinical - diagnostic value of gene polymorphism of PPAR-γ and plasma concentrations of vasomotor peptides: Abstract of Dis. .... Cand. of Med. Sciences: 14.01.11 / V.V. Bagriy. - Ivano-Frankivsk, 2015. - 20 p.
2. Zarubina E. G. The role of genetic predisposition in the development of cardiovascular disease in young adults with a violation of work and rest / E.G. Zarubina, E.V. Aseeva // Basic Research. - 2013. – Vol. 11. - P. 51-55.
3. Minushkina L.O. Genetic aspects of the regulation of endothelial function in essential hypertension / L.O. Minushkina, D.A. Zateyschikov, B.A. Sidorenko // Cardiology. - 2000. - Vol 3. - P. 68-76.
4. Mitchenko O.I. Leptynoresistance, blood pressure and structural and functional characteristics of myocardium in patients with hypertension and metabolic syndrome / J.I.

Mitchenko, V. I. Romanov, O.I. Kulyk et.al. // Ukrainian Medical Journal. – 2015. – Vol. 4 (108). - P. 91-94.
5. Netyazhenko V.Z. et. al. Unified clinical protocol of medical care in hypertension, approved by Order Ministry of Health of Ukraine from 24.05.2012 year №384.
6. Sakovych O.O. Gene polymorphism of the first type receptor of angiotensin II and levels of natriuretic peptides in postmenopausal women with hypertension, uncomplicated and complicated chronic heart failure: Abstract of Dis. .... Cand. of Med. Sciences: 14.01.11 / O.O. Sakovych. - Kyiv, 2012. - 20 p.
7. Syvolap V.D. Left ventricular remodeling in patients with hypertension and coronary heart disease, depending on the blood pressure variability / V.D. Syvolap, Y.V. Zemlianoy / Zaporozhye Medical Journal. - 2011. – Vol. 6 (13). - S. 61-64.
8. Tatarkin A.A. Structural and functional changes in the heart in young patients with hypertension / A.A. Tatarkin / Bulletin of the Far Eastern Branch of the Russian Academy of Sciences. - 2007. – Vol. 6. - P. 99-104.
9. Franchuk S.V. Inheritance of the first type receptor of angiotensin II gene and plasma levels of natriuretic peptides in women with hypertension, myocardial infarction or stroke, a role in predicting and diagnosing: Abstract of Dis. .... Cand. of Med. Sciences.: 14.00.11 / Svetlana V. Franchuk. - Zaporozhye, 2013. - 22 p.
10. Asai T. Endothelin-1 Gene Variant Associates With Blood Pressure in Obese Japanese Subjects / T. Asai, T. Ohkubo, T. Katsuya [et al.] // Hypertension. – 2001. - Vol. 38. - P. 1321-1324.
11. Dzholdasbekova A. U. The Association Between Polymorphism of Lys198Asn of Endothelin-1 Gene and Arterial Hypertension Risk in Kazakh People / A. U. Dzholdasbekova, A. E. Gaipov // Eur J Gen Med. – 2010. - Vol. 7(2). – P. 187-191.
12. Chairperson G.M. 2013 ESH/ESC Guidelines for the management of arterial hypertension / G.M. Chairperson, R.F. Chairperson, K. Narkiewicz [et al.] // Почки. – 2013. - №4 (06). – С. 1-10.
13. Jambric Z. Periferal vascular endothelial function testing for the diagnosis of coronary artery disease / Z. Jambric, L. Venneri, A. Varga [et al.] // Amer. Heart J. – 2004. – Vol. 41. – P. 684-689.
14. Khare A. Evaluation of markers of endothelial damage in case of young myocardial infarction / A. Khare, S. Shetty, K. Ghosh // Atherosclerosis. – 2005. – Vol. 18. – P. 375-380.
15. Nystrom T., Nygren A., Sjoholm A. Persistent endothelial dysfunction is related to elevated C-reactive protein levels in type II diabetic patients after acute myocardial infarction // Clin. Science. – 2005. – Vol. 12. – P. 121-128.
16. Treiber F. A. Endothelin-1 Gene LYS198ASN Polymorphism and Blood Pressure Reactivity / F. A. Treiber, P. Barbeau, G. Harshfield [et al.] // Hypertension. – 2003. – Vol. 42. - P. 494-499

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