ORIGINAL ARTICLE

Genetic Determinants of Complicated Pregnancy

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Abstract

Objectives: The connections of polymorphic variants of the gene of vasoactive hormones with the level of arterial pressure in pregnant women, depending on the development of preeclampsia (PE), have been studied. **Materials and Methods:** The study group included 382 pregnant women diagnosed with PE and 205 women with normal pregnancy. Polymorphisms of the endothelin-1 gene rs5370 (G>T *EDN1*) and guanine-binding protein β_3 subunit gene rs2301339 (G>A *GNB3*) were studied by real-time polymerase chain reaction (PCR) of DNA synthesis (real-time PCR). **Results:** Women with PE with genotype TT *EDN1* have lower values of systolic, diastolic, and mean arterial pressure at the end of pregnancy compared to those with G *EDN1* allele (genotypes GG and GT, $P = 0.01–0.04$). **Conclusions:** Thus, as a result of this study, significant associations of genetic polymorphisms with blood pressure indicators in pregnant women with PE were established.

Key words: Blood pressure, Genetic polymorphism, Preeclampsia, Pregnancy

INTRODUCTION

Recently, there has been a tendency to increase in the frequency of the pathological course of pregnancy, childbirth, the postpartum period in women, and perinatal morbidity and mortality in newborns.[1]

In this regard, the number of research devoted to molecular genetic studies of the complications of pregnancy has been constantly increasing.[2-4]

Preeclampsia (PE) is a complication of pregnancy, characterized by the development of endothelial dysfunction, multiple organ failure, disruption of coagulation and anticoagulation systems, microcirculation, metabolic processes, and immune response.[5]

According to the world literature and the WHO, the incidence of PE is 2–8%.[6,7] PE remains an important cause of maternal, perinatal, and neonatal morbidity and mortality.[7]

Severe PE and eclampsia cause the risk of complications such as hemorrhages and cerebral edema, placental abruption, disseminated intravascular coagulation syndrome, massive obstetric hemorrhages, HELLP syndrome, hemorrhage and rupture of the liver capsule, pulmonary edema, adult respiratory distress syndrome, and acute renal and hepatic insufficiency.[4,7]

According to a number of studies, this complication of pregnancy has a multifactorial nature.[3,8] Local gene networks of PE include endothelial dysfunction genes, vascular reaction genes, growth factor and cytokine genes, and major histocompatibility genes.[9-15]

MATERIALS AND METHODS

The study involved 587 women living in the territory of Central Russia (Belgorod region). The age of women ranged from 20 to 43 years (mean age 27.98 ± 4.50 years). Clinical and laboratory studies of women were conducted in the Perinatal Center of the Belgorod Regional Clinical Hospital (Department of Pathological Pregnancy). Criteria

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for inclusion in the study group were (a) Russian nationality, (b) gestational age 37–40 weeks, and (c) informed consent for the study. Pregnant women with diabetes and liver or kidney failure, as well as pregnant women with diabetes with a gestation period of <37 weeks and over 40 weeks, were excluded from the study. Blood pressure was measured 3 times according to the recommendations of the American Heart Association.[16] Data on blood pressure before pregnancy were obtained from case records of every woman. Mean blood pressure (MBP, mmHg) was calculated using the Hickam formula: MBP = (SBP + 2DBP)/3, where SBP is the systolic blood pressure and DBP is the diastolic blood pressure. Among 587 pregnant, 205 patients were with a physiological course of gestation and 382 women with a pregnancy complicated by PE.

PE was defined as the presence of hypertension, accompanied by proteinuria, as defined by a 24 h urine protein excretion more 300 mg.[17]

Molecular genetic methods

Venous blood (8–9 ml) was drawn from the ulnar vein of each woman. Genomic DNA was isolated using the method proposed Miller *et al*.[18] All women underwent typing of molecular genetic markers of endothelin-1 (G>T *EDN1* [rs5370]) and β_3 subunits of guanine-binding protein (G>A *GNB3* [rs2301339]).

The selection of polymorphisms was made in accordance with the criteria set forth in the paper by Ponomarenko.[19]

Loci genotyping was produced using real-time polymerase chain reaction (PCR) by the method of TaqMan probes detection according to relative fluorescence unit values of each probe on the thermocycler IQ5 with detecting system in real time. "Bio-Rad IQ5-Standart Edition" program was used for the alleles discrimination.

Statistical analysis

Allele frequencies of the genes polymorphism were estimated by the gene counting method, and the Chi-square test was used to identify significant departure from Hardy–Weinberg equilibrium. The distribution of allele and genotype frequencies between the study groups was compared by the Chi-square test for 2 × 2 contingency tables. The distribution of the quantitative traits such as SBP, DBP, MAP, and pulse pressure (the pressure difference between the systolic and diastolic pressures, PBP) before and at the end of pregnancy was analyzed by the Shapiro–Wilks test. Since the values of the quantitative traits did not follow the normal distribution, median (Me) and interquartile range (Q25-Q75) were used for their description and intergroup comparisons were done using the Mann–Whitney U-test. All statistical analyses were performed using STATISTICA for Windows v. 6.0 (StatSoft, USA).

RESULTS

The biomedical and clinical characteristics of the study women are shown in Table 1.

As a result of studying the relationship of polymorphism of endothelin gene 1 G>T EDN1 rs5370 with BP levels in women at the end of pregnancy, significant associations were established only for pregnant women with PE [Table 2]. Women with PE with genotype TT *EDN1* have lower values of systolic, diastolic, and mean arterial pressure at the end of pregnancy compared to those with G *EDN1* allele (genotypes GG and GT, $P = 0.01–0.04$).

The study of the relationship between the polymorphism of the guanine-binding protein β3 subunit of rs2301339 and the blood pressure levels in pregnant women revealed no significant associations.

DISCUSSION

The results of this study suggest that G>T *EDN1* rs5370 polymorphism is associated with the level of arterial pressure in pregnant women with PE (37–40 weeks). The genotype TT *EDN1* is associated with lower rates of systolic, diastolic, and MBP.

Gene *EDN1* is located on the chromosome 06p24. The polymorphism studied is due to the replacement of guanine with thymine at position 5665 in exon 5 and leads to the replacement of the amino acid lysine with asparagine in the 198th codon of

Table 1: The biomedical and clinical characteristics of the study women

Variables	Value variables, Me (Q25-Q75)	
	Pregnant women without preeclampsia	Pregnant women with preeclampsia
Number	205	382
Age, year	26.7 (24.0–30.0)	27.2 (25.0–31.0)
Height, m	1.57 (1.54–1.60)	1.65 (1.62–1.68)
Weight, kg	70.0 (59.0–81.7)	79.3 (71.6–90.4)
BMI, kg/m^2	23.0 (21.2–26.9)	29.2 (26.4–33.8)
SBP, mmHg	110.0 (110.0–120.0)	140.0 (135.0–150.0)
DBP, mmHg	70,0 (70.0–75.0)	90.0 (85.0–100.0)
PBP, mmHg	40.0 (40.0–40.0)	50.0 (50.0–60.0)
MAP, mmHg	85.0 (83.3–90.0)	106.7 (103.3–116.7)
ΔMAP, mmHg	5.0 (0.0–10.0)	25.0 (20.0–35.0)
ΔSBP, mmHg	0.0 (0.0–10.0)	20.0 (10.0–25.0)
ΔDBP, mmHg	3.3 (0.0–8.3)	20.0 (13.3–28.3)

BMI: Body mass index, SBP: Systolic blood pressure, DBP: Diastolic blood pressure, PBP: Pulse blood pressure, MAP: Mean arterial pressure, Δ MAP: Change of mean arterial pressure, ΔSBP: Change of systolic blood pressure, ΔDBP: Change of diastolic blood pressure

Table 2: Association rs5370 polymorphism of the endothelin 1 gene with indicators of blood pressure in women at the end of pregnancy, Me (Q25-Q75)

Values of blood pressure	Genotypes		*P*
	GG+GT	TT	
Pregnant women without preeclampsia (PE) (*n*=205)			
N	195	10	
SBP, mmHg	110.0 (110.0–120.0)	110.0 (110.0–115.0)	0.3
DBP, mmHg	70.0 (70.0–80.0)	70.0 (70.0–75.0)	0.8
PBP, mmHg	40.0 (40.0–45.0)	40.0 (30.0–40.0)	0.3
MAP, mmHg	85.0 (83.3–90.0)	83.3 (83.3–83.3)	0.4
ΔMAP, mmHg	3.3 (0.0–8.3)	0.8 (0.0–6.6)	0.4
ΔSBP, mmHg	5.0 (0.0–10.0)	0.0 (0.0–10.0)	0.3
ΔDBP, mmHg	0.0 (0.0–10.0)	0.0 (0.0–5.0)	0.7
Pregnant women with PE (***n***=382)			
N	377	5	
SBP, mmHg	140.0 (135.0–150.0)	130.0 (130.0–140.0)	0.04
DBP, mmHg	90.0 (85.0–100.0)	80.0 (80.0–80.0)	0.02
PBP, mmHg	50.0 (50.0–60.0)	50.0 (50.0–55.0)	0.8
MAP, mmHg	106.7 (103.3–116.7)	100.0 (96.7–103.3)	0.01
ΔMAP, mmHg	20.0 (13.3–28.3)	20.0 (6.7–20.0)	0.2
ΔSBP, mmHg	25.0 (20.0–40.0)	20.0 (20.0–20.0)	0.1
ΔDBP, mmHg	20.0 (10.0–25.0)	15.0 (5.0–20.0)	0.2

BMI: Body mass index, SBP: Systolic blood pressure, DBP: Diastolic blood pressure, PBP: Pulse blood pressure, MAP: Mean arterial pressure, Δ MAP: Change of mean arterial pressure, ΔSBP: Change of systolic blood pressure, ΔDBP: Change of diastolic blood pressure

the polypeptide encoded by the gene (rs5370).[20] As some studies show, this polymorphism is associated with the plasma content of endothelin 1 in women during pregnancy.[21,22] It has been established that carriers of the allele T *EDN1* have a higher level of endothelin 1 in the blood plasma compared to subjects with the genotype GG EDN1. In contrast, the study by Tanaka *et al*.[23] showed that plasma amounts of endothelin-1 in patients with essential hypertension do not differ in individuals with the allele T *EDN1* and the genotype GG *EDN1*.[23]

Perhaps, the polymorphism of the gene *EDN1* rs5370 is in close adherence to another SNP that provides an impact on blood pressure values.

The study conducted on the Mexican population has established the protective effect of this polymorphism on the development of PE.[24] Similar results were obtained in a study on the polish population.[25]

The observed inconsistency in the above results may be related to interpopulation and interethnic differences in the frequencies of alleles of polymorphism *EDN1* rs5370.[26,27]

CONCLUSIONS

Thus, as a result of the study, the relationships of the genetic polymorphisms of vasoactive hormones with the level of blood pressure in women with PE were revealed. Women with genotypes GG and GT EDN1 (rs5370) have higher values of systolic, diastolic, and mean arterial pressure at the end of pregnancy.

Data obtained as a result of the research broaden the understanding of the mechanisms of preeclampsia development, and also allows prediction the nature of the clinical course of the disease, which will ensure the optimization of the treatment and diagnostic process for each patient.

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