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Association of *ACE* Gene Polymorphism with Hypertension and Metabolic Risk Factors among Indigenous People of the Northern Territory of Yakutia

Sargylana I. Sofronova, PhD*; Maria P. Kirillina, PhD; Vyacheslav M. Nikolaev, PhD; Sardaana K. Kononova; Oksana G. Sidorova; Anna N. Romanova

Yakut Science Center of Complex Medical Problems
Yakutsk, the Republic of Sakha (Yakutia), Russia

Abstract

The research objective was to study the association of the *ACE* gene I/D polymorphism with essential hypertension (EH) and metabolic risk factors among indigenous people of the northern territory of Yakutia. The obtained data show that representatives of indigenous people of the North of Yakutia with the *ACE* ID genotype are characterized with high levels of systolic blood pressure. The carriage of DD genotype in EH patients was associated with a high frequency of hypercholesterolemia and hyper-LDL cholesterolemia. The carriage of ID genotype in EH patients, compared to subjects without EH, was characterized by higher blood levels of TC, LDL-C, TG, and FPG and associated with a high frequency of obesity. Thus, the *ACE* I/D polymorphism was found to be associated with metabolic risk factors in indigenous EH patients of the North of Yakutia. (**International Journal of Biomedicine. 2019;9(2):102-105.**)

Key Words: essential hypertension • *ACE* gene • indigenous people • risk factors

Abbreviations

ACE, angiotensin-converting enzyme; **AO**, abdominal obesity; **EH**, essential hypertension; **FPG**, fasting plasma glucose; **HDL-C**, high-density lipoprotein cholesterol; **LDL-C**, low-density lipoprotein cholesterol; **OGTT**, oral glucose tolerance test; **RAS**, renin–angiotensin system; **SBP**, systolic blood pressure; **WS**, waist circumference.

Introduction

Hypertension is one of the main risk factors worldwide for cardiovascular disease and the main reason for a high mortality rate among the adult population.[1,2] Essential hypertension (EH), the most common form of hypertension,[3] is defined as an elevation in blood pressure of unknown cause, which increases the risks for cerebral, cardiac, and renal complications.[4] EH is considered a multifactorial disease.[5] From a genetic perspective, many single nucleotide polymorphisms (SNPs), genes and epigenetic factors are associated with EH.[6] Currently, increasing attention is being paid to RAS genes in the development of EH, and the value of *ACE* gene I/D polymorphism has been investigated in many studies.[7-9] A strong association between the DD genotype and the D allele with EH, abdominal obesity and coronary artery disease was revealed in a number of studies.[10-12] Grigor'eva found a significant association between *ACE* gene polymorphism and the risk of myocardial infarction in Yakut men.[13]

The research objective was to study the association of *ACE* gene polymorphism with hypertension and metabolic risk factors among indigenous people of the northern territory of Yakutia.

Materials and Methods

This study was done to determine the clinico-epidemiological aspects of EH in the remote districts in the North of Yakutia (Kolymskoye and Andryushkino rural

***Corresponding author:** Sargylana I. Sofronova, PhD. Yakut Science Center of Complex Medical Problems. Yakutsk, the Republic of Sakha (Yakutia), Russia. E-mail: sara2208@mail.ru*

localities of the Nizhnekolymsky District, Topolinoe rural locality of the Tomponsky District, Nelemnoye rural locality of the Verkhnekolymsky District). A total of 348 indigenous people of Yakutia (Evens, Chukchi, Yukaghirs, Yakuts) (225 women and 123 men) were examined. The patient sample consisted of adults aged between 20 and 70 years, with an average age of 45.71±0.67 years. All patients were divided into 2 groups. Group 1 consisted of 175 patients (mean age of 53.11±0.51 years) with EH; Group 2 (control group) included 173 people (mean age of 38.88±0.60 years) without elevated blood pressure.

The study was approved by the Ethics Committee of the Yakut Science Center of Complex Medical Problems. Written informed consent was obtained from each patient.

A comprehensive clinical examination and laboratory tests included the following procedures:

- Anthopometrical reference data: BMI was calculated using Quetelet's formula (in kg/cm^2). Measurement of WC was made at the uppermost lateral border of the ilium using a tape measure (in cm)
- Assessment of blood pressure by Korotkov's method.
- Assessment of FPG, OGTT, and blood levels of TG, HDL-C, LDL-C.

Glucose and lipid metabolism disorders were diagnosed according to the Russian national recommendations (the All-Russian Scientific Society of Cardiologists, 2009)[14] based on the IDF consensus criteria (2006)[15]: TG ≥1.7 mmol/l; HDL-C <1.0 mmol/l in males and <1.2 mmol/l in females; LDL-C >3.0 mmol/l; FPG >6.1 mmol/l; IGT 2Hr PG ≥7.8 mmol/l and ≤ 11 mmol/l . Abdominal obesity (AO) was confirmed at WC ≥ 94 cm in males and ≥ 80 cm in females.

The diagnosis of hypertension was based on 2017 ACC/AHA Guideline for or the Prevention, Detection, Evaluation, and Management of High Blood Pressure in Adults.[16]

The insertion/deletion (I/D) polymorphism of the *ACE* gene was examined by PCR in the laboratory of molecular genetics at Yakut Science Center of Complex Medical Problems. From each patient, 2ml of peripheral blood were drawn into an EDTA tube. Genomic DNA was isolated from the peripheral blood leukocytes using standard phenol–chloroform extraction technique (Maniatis et al., 1982) Genotyping was carried out with the allele specific primers method.

Reactions were performed with 10 pmol of each primer:

F: 5'-CTG GAG ACC ACT CCC ATC CTT TCT-3'
R: 5'-GAT GTG GCC ATC ACA TTC GTC AGA T-3'.

PCR products were analyzed on 2% agarose gels after staining with ethidium bromide and were visualized using a UV transilluminator. Two alleles were identified: a 490-bp fragment I (with the insertion) and a 190-bp fragment D (without the insertion). In heterozygous samples, two bands (490 and 190 bp) were detected. To avoid mistyping of heterozygotes (ID) DNA samples identified as a DD genotype were subsequently amplified with second set of primers designed for the insertion specific allele.

Statistical analysis was performed using SPSS (version 17.0). Baseline characteristics were summarized as frequencies and percentages for categorical variables and as mean± SEM for continuous variables. Student's unpaired and paired t-tests were used to compare two groups for data with normal distribution. Odds ratios (OR) and 95% confidence intervals (CI) were calculated. Multiple comparisons were performed with one-way ANOVA and post-hoc Tukey HSD test. Deviation from Hardy-Weinberg equilibrium and differences in allele distributions between the two groups were assessed by χ2- test with 1 degree of freedom (df). A probability value of P<0.05 was considered statistically significant.

Results and Discussion

In the general population, the *ACE* II, ID, and DD genotype frequencies were 19.5% (n=68), 65.2% (n=227), and 15.2% (n=227). We did not find statistically significant differences in the frequency distribution of *ACE* I/D alleles and genotypes between the group of patients and control group (Table 1). The distribution of the genotype frequency was not in HWE for patients (χ^2=19.17, p<0.05) and for controls (χ^2=13.95, p<0.05). The occurrence of the departure from HWE in controls is probably due to population substructure. We further used the three types of genetic models to test the association between the *ACE* I/D polymorphism and EH; the results are shown in Tables 2a-2c. We found no association between the *ACE* I/D polymorphism and EH in our case–control study.

Table 2a.
General model of inheritance (df = 2)

Genotype	Genotype frequencies		χ^2	*P*	OR	95% CI
	Patients	Control				
II	0.200	0.191	0.63	0.73	1.06	0.62-1.80
ID	0.663	0.642			1.10	0.71-1.71
DD	0.137	0.168			0.79	0.44-1.42

Table 1.
Frequencies of the genotypes and alleles of the ACE I/D polymorphism and deviations from HWE (df = 1)

Genotype	Patients	HWE	χ^2	*P*	Control	HWE	χ^2	*P*	Allele	Allele frequencies	
										Patients	Control
II	0.200	0.282	19.17	1.0E-5	0.191	0.262	13.95	0.0002	I	0.531	0.512
ID	0.663	0.498			0.642	0.500			D	0.469	0.488
DD	0.137	0.220			0.169	0.239					

Table 2b.

Dominant model of inheritance (df = 1)

Genotype	Genotype frequencies		χ^2	*P*	OR	95% CI
	Patients	Control				
II+ID	0.863	0.832	0.63	0.43	1.27	0.70-2.28
DD	0.137	0.168			0.79	0.44-1.42

Table 2c.

Recessive model of inheritance (df = 1)

Genotype	Genotype frequencies		χ^2	*P*	OR	95% CI
	Patients	Control				
II	0.200	0.191	0.05	0.83	1.06	0.62-1.80
ID+DD	0.800	0.809			0.94	0.55-1.60

In the general population (n=348), we did not find significant differences in average values of TG, LDL-C, and HDL-C depending on the carriage of genotypes of the *ACE* I/D polymorphism. The frequency of hypercholesterolemia was 51.5% in carriers of II homozygous genotype, 42.7% in carriers of ID heterozygous genotype, and 49.1% in carriers of DD heterozygous genotype (*P*>0.05). Hyper-LDL cholesterolemia was found in 64.2% of DD homozygotes, 54.4% of II homozygotes, and 54.6% of ID heterozygotes (*P*>0.05). Hypo-HDL cholesterolemia was found in 33.8% of II homozygotes, 34.8% of ID heterozygotes, and 35.8% of DD homozygotes (*P*>0.05).The frequency of hyperglycemia was as follows: ID carriers - 6.6%, II carriers - 2.9%, and DD carriers - 3.8%.

In Croup 1 (patients with EH), the average level of SBP in ID carriers, II carriers and DD carriers was 144.2±1.2mmHg, 136.6±2.8mmHg and 138.8±2.1mmHg, respectively, *P*=0.0072. Table 3 presents the relationship between *ACE* genotype carriage and parameters of lipid and glucose metabolism in two groups. In ID carriers, the blood levels of TC, LDL-C, TG, and FPG were significantly higher in Group 1 than in Group 2. In II carriers, the blood level of TG was significantly higher in Group 1 than in Group 2.

We found a high frequency of hyper-LDL cholesterolemia in DD carriers of Group 1 (70.8%) compared to Group 2 (58.4%) (*P*=0.015). In DD carriers, the frequency of hypercholesterolemia was also significantly higher in Group 1 than in Group 2 (66.8% versus 34.7%, *P*=0.000)

In the general population, the frequency of AO in ID carriers, II carriers and DD carriers was as follows: 59.9%, 55.9%, and 50.9%, respectively, *P*>0.05. The average level of WC in ID carriers, II carriers and DD carriers was 89.61±0.65 cm, 86.46±1.49 cm, and 86.75±1.52 cm, respectively, *P*=0.0391.

In Group 1, the average level of WC in ID carriers, II carriers and DD carriers was 96.23±0.89 cm, 91.43±1.99 cm, and 92.63±1.27cm (*P*=0.0205), respectively. The frequency of AO was as follows: ID carriers – 84.5%, II carriers – 71.4%, and DD carriers – 66.7% (*P*>0.05). AO frequency in ID carriers vs. DD carriers was significantly higher (*P*<0.05).

In Group 2 the average level of WC in ID carriers, II carriers and DD carriers was 82.68±0.68 cm, 81.18±1.85 cm, and 81.90±1.21 cm (*P*>0.05). The frequency of AO was as follows: ID carriers – 34.2%, II carriers – 39.4%, and DD carriers – 37.9% (*P*>0.05)

Conclusion

The obtained data show that representatives of indigenous people of the North of Yakutia with the *ACE* ID genotype are characterized with high levels of SBD. The carriage of DD genotype in EH patients is associated with a high frequency of hypercholesterolemia and hyper-LDL cholesterolemia. The carriage of ID genotype in EH patients, compared to subjects without EH, is characterized by higher blood levels of TC, LDL-C, TG, and FPG and associated with a high frequency of AO. A number of studies have also found a high frequency of metabolic syndrome in ID carriers.[17-20] Thus, the *ACE* I/D polymorphism was found to be associated with metabolic risk factors in indigenous EH patients of the North of Yakutia.

Competing Interests

The authors declare that they have no competing interests.

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Table 3.

Mean concentrations of lipid spectrum and glucose among hypertensive patients and persons without hypertension

Blood parameters	Genotype II			Genotype ID			Genotype DD		
	Group 1	*P*	Group 2	Group 1	*P*	Group 2	Group 1	*P*	Group 2
TC	5.05±0.14	>0.05	4.68 ±0.16	5.15±0.09	<0.01	4.79±0.07	5.13±0.16	>0.05	4.71±0.16
LDL-C	3.24±0.13	>0.05	2.88±0.13	3.32±0.08	<0.01	3.05±0.06	3.32±0.14	>0.05	3.07±0.12
HDL-C	1.26±0.05	>0.05	1.37±0.06	1.26±0.03	>0.05	1.33±0.03	1.28±0.06	>0.05	1.14±0.05
TG	1.21±0.09	<0.02	0.92±0.07	1.21±0.05	0.000	0.91±0.03	1.16±0.10	>0.05	1.07±0.08
FPG	4.48±0.18	>0.05	4.41±0.14	5.02±0.17	0.000	4.19±0.08	4.48±0.27	>0.05	4.25±0.14

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