

## **ABSTRACT**

of the dissertation «Molecular and genetic predictors to intracranial subarachnoid hemorrhage in Kazakh ethnic group» of Akbota M. Aitkulova for the degree Doctor of Philosophy (PhD) in «Biology - 6D060700»

### **General description of the study**

The dissertation is devoted to the study of associative links of allelic polymorphisms of the genes with the risk of intracranial subarachnoid hemorrhage.

### **Relevance of the research topic**

Stroke is the second leading cause of death worldwide. The most common subtype of hemorrhagic stroke is intracranial subarachnoid hemorrhage (SAH). The main cause of SAH is rupture of intracranial aneurysms (IA). Asymptomatic carriers of intracranial aneurysms are 2-5% of the population. About 30% of patients with ruptured aneurysms die during the acute period of hemorrhage, 50% die within the first month. Patients survived at the first bleeding have risk of developing recurrent intracranial hemorrhages and a high risk of death. Since the process of aneurysm formation is asymptomatic, timely diagnosis of IA is a major medical problem. Often, aneurysms are detected accidentally during screening or immediately upon rupture and SAH. Today, IA diagnostics is carried out only by instrumental methods, including magnetic resonance angiography (MRA), computed tomography (CTA) and selective cerebral angiography (SCA). However, despite the effectiveness, the methods are invasive and have a number of limitations.

Timely IA detection, surgery and intensive care can reduce the risk of SAH by up to 50%. Therefore, it is important to know the mechanisms of formation and rupture of intracranial aneurysms, which are still not fully understood. Many studies of the IA showed that the rupture was preceded by many complex processes leading to a change in the structure of the cerebral vascular wall. These processes included dysfunction of the endothelium, the inflammatory and pro-inflammatory reaction, the effect of hemodynamic pressure on blood vessels, and a number of other endogenous and exogenous factors. The size and location of IA played an important role in the risk of SAH. It is known that most often the formation and rupture of aneurysms occurs in the area of bifurcation of the vessels, since these areas are most strongly affected by hemodynamic pressure. Studies carried out in various populations have shown that patients with arterial hypertension have a higher risk of rupture of aneurysms and SAH than those without hypertension. At the same time, hypertension was an independent risk factor for SAH. Several

studies showed that smoking, alcohol consumption, gender and age are the risk of IA rupture and SAH.

The important role of heredity in the risk of SAH has been proven in studies in various populations. A history of subarachnoid hemorrhages in the first relatives increased the risk of SAH in patients by 7-10 times. In addition, patients at risk of intracranial aneurysm rupture and SAH had a younger age among patients with familial SAH compared with sporadic cases. In this regard, genetic factors that regulate the processes that determine the risk of aneurysm rupture and SAH are being studied [24-35]. Genome-wide association analysis (GWAS) and exome sequence analysis have identified more than 200 significant polymorphic loci potentially involved in the molecular mechanisms of disease development. However, not all polymorphic loci support association with SAH in replication studies due to genetic differences among ethnic groups. Therefore, it is necessary to conduct a replicative study of the association of genetic variants with the risk of SAH in the Kazakh ethnic group.

**The object of the study** is DNA samples isolated from the blood of patients with intracranial aneurysms and people of the control group.

**The subject of the study** is allelic polymorphisms, as well as genes associated with intracranial subarachnoid hemorrhage.

#### **The aim and objectives of the study**

The aim of the study is investigation of associations of polymorphic genetic variants with the risk of spontaneous intracranial subarachnoid hemorrhages in Kazakhs ethnic group.

To achieve the goal, the following objectives were set:

1. Identification of the genetic similarity or difference between the Kazakh ethnic group and world populations in terms of significant polymorphisms of genes associated with the risk of SAH.

2. Identification of the association of the studied polymorphic genetic markers with the risk of sporadic forms of intracranial subarachnoid hemorrhages in Kazakhs ethnic group.

3. Identification of candidate genes involved in the risk of familial SAH in Kazakhs ethnic group.

4. Identification an association of the clinical predictors with potential molecular genetic markers determine of the risk of subarachnoid hemorrhage in Kazakhs ethnic group.

#### **Research methods**

The study of genetic variants was carried out by molecular biological and statistical methods.

#### **The scientific novelty of the study**

- For the first time, molecular genetic markers associated with sporadic and familial forms of subarachnoid hemorrhages in the Kazakh ethnic group were identified.

- Associations of genetic variants c.202-4463 C>T (rs175646) of the gene *JDP2* (rs175646) and c.2092 G>A (rs1800255) of the gene *COL3A1* with sporadic forms of SAH in the Kazakh ethnic group were revealed for the first time.

- Candidate genes involved in the development of SAH in the hereditary form in Kazakhs ethnic group were identified for the first time.

- A targeted panel of molecular genetic markers for early diagnosis of SAH by exome sequencing and Open Array genotyping was developed.

- For the first time, associations of genetic and clinical factors that cause SAH were established. It was revealed that arterial hypertension and age are independent risk factors for sporadic forms of SAH in Kazakhs ethnic group.

### **The theoretical project significance**

The results obtained during the research will help to expand knowledge about the molecular basis of the pathophysiology of sporadic and hereditary forms of subarachnoid hemorrhage and can be used in personalized medicine for the development of therapeutic approaches for the diagnosis and treatment of SAH, considering an important factor as ethnicity.

An interdisciplinary nature of the obtained results provides valuable information for specialists in related sciences, finding wide application in the educational process of students of biological and medical specialties. As part of the dissertation thesis, methodical manual, approved by the Ministry of Health of the Republic of Kazakhstan were developed for students of the specialties of general medicine and medical genetics, researchers, and doctors (No. 31 / 17.10.2017, ISBN 978-601-305. -248-9).

### **The practical project significance**

Based on the results of the dissertation thesis the patent for the invention of the Republic of Kazakhstan No. 33654 / 05/29/2019 "A method for diagnosing the risk of developing intracranial aneurysms in the Kazakh population" was obtained. Ethnic specific diagnostic panel determines the potential risk of intracranial subarachnoid hemorrhage based on molecular genetic markers identified during the study was created. Applying of genetic testing in practical medicine will identify patients with a high risk of subarachnoid hemorrhage and provide timely prevention and treatment of the disease.

### **Main conclusions based on research results**

1. The Kazakh ethnic group has differences in the frequency of occurrence of allelic variants of genes associated with subarachnoid hemorrhages, and occupies an intermediate position between Latin American, European, and Asian populations according to cluster analysis of genetic distances.

2. Polymorphisms c.202-4463 C>T (rs175646) of the gene *JDP2* (rs175646) and c.2092 G>A (rs1800255) of the gene *COL3A1* are potential genetic markers that contribute to the risk of sporadic SAH cases in the Kazakh ethnic group.

3. Genetic variant c.1382 G>A of the *ALCAM* gene is potential candidate gene that cause the risk of familial subarachnoid hemorrhages in the Kazakh ethnic group.

4. Clinical factors such as hypertension and age determine the risk of SAH in Kazakhs ethnic group.

### **Connection of work with scientific research programs**

The dissertation thesis was carried out within the framework of the project No. 4751 / GF4 "Research of genetic markers to assess the prognosis of the disease in cerebrovascular aneurysms in the Kazakh population" (GR 0115RK00539, 2015-2017). GF4 "Study of candidate genes involved in the development of familial subarachnoid hemorrhage based on whole exome sequencing» (AP08955996, 2020-2021).

### **Work approbation**

The main research results were presented at national and international conferences:

- 3rd International Scientific Conference "Genetics and biotechnology of the 21st century: problems, achievements, prospects" dedicated to the 115th anniversary of the birth of Academician A.R. Zhebrak (Minsk, Belarus, 2016);

- IX All-Russian Scientific and Practical Conference "Molecular Diagnostics 2017" (Moscow, Russian Federation, 2017);

- XVI World Congress of Neurosurgery, WFNS 2017 (Istanbul, Turkey, 2017);

- International conference of students and young scientists "al Farabi Alemi" (Almaty, Kazakhstan, 2018);

- XXV International Scientific Conference of Students, Postgraduates and Young Scientists "Lomonosov" (Moscow, Russian Federation, 2018);

- International Symposium "Astana Biotech 2018" (Astana, Kazakhstan, 2018);

- International Symposium of Human Phenomics (Shanghai, China, 2018).

### **Publications**

16 publications, including 2 papers in the journals abroad, indexed by Web of Knowledge (Thomson Reuters, USA) and Scopus (Elsevier, Netherlands), 5 papers in the journals of the Republic of Kazakhstan, included in the list of recommended by Ministry of education and science of RK, 5 abstracts in proceedings of conferences of foreign countries, and 2 abstracts in proceedings of international conferences of the Republic of Kazakhstan, 1 patent for an invention of the Republic of Kazakhstan, 1 methodical manual, approved by the Ministry of Health of the Republic of Kazakhstan were published.

### **Volume and structure of the dissertation**

The volume of the dissertation thesis 122 pages and consists of introduction, literature review, materials and methods, results and discussion of results, conclusions, list of references consisting of 262 titles and 13 supplement materials. There are 22 tables and 20 figures in the dissertation.