«MEDICAL UNIVERSITY KARAGANDA»

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# ANNOTATION

Dissertation work for the PhD degree specialty 8D10100 "Medicine"

# Topic: “ Molecular genetic prediction of the risk of developing new cardiovascular events after coronary artery stenting”

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# Relevance of the research topic:

# The problem of treating patients with coronary heart disease (CHD) remains one of the most urgent and priority tasks of world and domestic health care. Despite significant progress in the prevention and treatment of coronary artery disease over the past decade, it still occupies a leading position in the structure of morbidity and mortality in the population of developed industrial countries.

# Despite the progress and widespread use of percutaneous coronary intervention, the development of new cardiovascular events are decisive factors that limit its long-term effectiveness.

# To date, there are a number of diagnostic concepts aimed at studying the factors influencing the development of new cardiovascular events after coronary artery revascularization. The development of these events depends on the procedure of intracoronary intervention, the age of the patient, the presence of concomitant diseases, the degree and extent of the lesion, the length of the lesion, the diameter of the vessel lumen after implantation, and other factors.

# One recent trend of interest is the study of the role of gene polymorphisms in the development of fatal cardiovascular events.

# Due to the fact that there are scientific discussions about the significance of gene polymorphism in the risk of developing cardiovascular complications after interventional interventions according to scientific databases, it is of interest to evaluate molecular and genetic predictors of the development of cardiovascular events.

# The purpose of the study:

To evaluate the relationship of clinical and polymorphic genetic markers with the risk of developing new cardiovascular events in coronary artery disease in the early and late periods after stent placement..

# Research objectives:

1. To assess the significance of clinical and laboratory parameters in the risk of developing cardiovascular events in patients with coronary artery disease after percutaneous coronary intervention.

2. To determine the association of polymorphic genetic markers with the risk of developing cardiovascular events after percutaneous coronary intervention in the ethnic group of Kazakhs, depending on the pathogenetic factors in the development of the disease.

3. To assess the prognostic significance of gene polymorphisms in the risk of complications and outcomes after percutaneous coronary intervention.

4. To develop predictors of the development of cardiovascular events in the early and late periods after percutaneous coronary intervention

# Scientific novelty:

-Identified clinical markers associated with the risk of new cardiovascular events in the early period after stent placement in the ethnic group of Kazakhs.

- The association of genetic variants of polymorphism with cardiovascular risks in patients belonging to the Kazakh ethnic group was determined: rs1234313-TNFSF4; rs3184504 - SH2B3; rs2340690 - HSPE1; rs6725887-ICA1L; rs1799963 - CKAP5; rs1799983-CKAP5; rs688034-SEZ6L; rs268 - LPL; rs2229616 - MC4R; rs2943634; rs599839 - PSRC1; rs2774279 - USF1; rs1800783 - NOS3; rs1051730 - NOS3; rs10116277 - CDKN2B-AS1; rs2383207 - CDKN2B-AS1; rs2713604 - DNAJB8-AS1; rs9536314 - KL.

- Genetic polymorphism was determined in the risk of developing cardiovascular events after endovascular intervention in patients with coronary artery disease in the long term.

 - Associations of clinical and genetic predictors that increase the risk of new cardiovascular events after percutaneous coronary intervention have been established.

# Practical significance: Predictors for predicting the risk of developing new cardiovascular events in the early and late periods after stent placement were evaluated.

# Relationship of the thesis with other research papers

#  The dissertation work was carried out at the Department of Internal Diseases and at the Collective Use Laboratory of the NJSC "MUK".

#  The implementation of the dissertation work was carried out within the framework of program-targeted funding of the Ministry of Education and Science of the Republic of Kazakhstan for 2018-2020. on the topic: “Personalized approach in solving a number of significant diseases”, on task 3: “Search and evaluation of the main genetic markers of resistance to antiplatelet therapy in patients with coronary heart disease among representatives of the main ethnic group of Kazakhstan”. Registration number No. 0118RKO1034.

**The main provisions for defense:**

1. The risk of new adverse cardiovascular events in the early period after percutaneous coronary intervention increases in the presence of risk factors in the form of overweight, obesity of 1 and 2 degrees, high levels of low-density lipoproteins, as well as the elderly age of the patient: respectively in 3.9 -12.6 -8.0 -1.6 -1.07 times. Cardiac risk increases significantly in the presence of arrhythmia and chronic heart failure: 9.0 - 25.0 times, respectively.

2. Identification of intergenic polymorphism of the rs2943634 gene (OR-4.0 times, p=0.006) after percutaneous coronary intervention predicts a high risk of developing cardiovascular events in the long-term period.

3. Identification of polymorphisms of the inflammatory biomarker gene (Rs 1234313 of the TNFSF4 gene), the immune response gene (Rs 3184504 of the SH2D3 gene), the lipid metabolism biomarker gene (Rs 2943634), and the vascular endothelial biomarker gene (Rs 2713604, DNAJB8-AS1) increase the risk of developing new cardiovascular events.

4. Genetic polymorphisms that increase cardiovascular risks in patients with Kazakh ethnicity include:

- genotypes of genes for biomarkers of inflammation and immune response: rs1234313 TNFSF4, p=0.000; rs3184504 - SH2B3, p=0.000; rs2340690 - HSPE1, p=0.002; rs6725887 - ICA1L, p=0.002.

- genotypes of biomarker genes of the hemostasis system: rs1799963 - CKAP5, p=0.000; rs1799983 -CKAP5, p=0.008; rs688034 -SEZ6L, p=0.045.

- genotypes of lipid metabolism biomarker genes: rs268 - LPL, p=0.022; rs2229616 - MC4R, p=0.000; rs2943634, p=0.030; rs599839 - PSRC1, p=0.030; rs2774279 - USF1, p=0.006.

- genotypes of endothelial biomarker genes: rs1800783 - NOS3, p=0.000; rs1051730 - NOS3, p=0.001; rs10116277 - CDKN2B-AS1, p=0.001; rs2383207 - CDKN2B-AS1, p=0.004; rs2713604 - DNAJB8-AS1, p=0.000; rs9536314 - KL, p=0.000.

5. The predictors of the development of new cardiovascular events in the early and late periods after endovascular interventions include a combination of clinical and genetic factors: the patient's advanced age, overweight, obesity of 1-2 degrees, an increase in low-density lipoproteins, cardiac arrhythmias, chronic heart disease. insufficiency and polymorphism of genes.

# Implementation into practice

# Based on the materials of the dissertation, 3 certificates of registration of rights to the object of copyright were obtained:

#  - KZ №13260. Certificate of entering information into the state register of rights to objects protected by copyright dated November 17, 2020. "Questionnaire for assessing the risk of complications during dual antiplatelet therapy in cardiac patients", authors Bodaubai R., Taizhanova D.Zh. Visternichan O.A., Kalimbetova A.B.

# - KZ №13249. Certificate of entering information into the state register of rights to objects protected by copyright dated November 16, 2020. "Questionnaire for assessing risk factors in atrial fibrillation", authors Taizhanova D.Zh. Bazarova N.K., Bodaubay R., Kalimbetova A.B.

# - KZ No. 31084. Certificate of entering information into the state register of rights to objects protected by copyright. December 14, 2022 “An algorithm for predicting the development of new cardiovascular events after PCI. Teri arkyly coronarlyk aralasudan keyin zhan zhurek-kan tamyrlary oқigalarynyn damuyn bolzhau algorithmi”, authors Taizhanova D.Zh., Kalimbetova A.B.

#  The results of the study have been introduced into cardiology practice to predict complications after percutaneous coronary intervention in the early and late periods. Information on predictors of predicting cardiovascular events after PCI has been introduced into the educational process of the Department of Internal Diseases of the NJSC "MUK" under the educational program: 7R01108 "Adult and Pediatric Cardiology" for residents as part of practical classes and seminars.

# Approbation of work

# The main provisions of the dissertation are presented:

# - The International Conference of the European Society of Cardiology "Heart & Stroke", 24.01.-25.01.2020, Barcelona, Spain in the form of a poster presentation;

# - Russian National Congress of Cardiology, 29.09. -01.10. 2020, Kazan, Russia;

# - Russian National Congress of Cardiology, 21.10. -23.10. 2021, St. Petersburg, Russia;

# - Online conference “Gene polymorphism and oncogenesis”, 25.05. 2022, which was held jointly by NJSC "MUK" and "Life Science Leadership School" as part of a series of seminars "Permanent International Conference On Health Innovative Solutions", 2021-2022, Karaganda, Kazakhstan in the form of oral presentations;

# - I Interuniversity Conference "Actual Issues of Somatic Diseases", 09.06.-10.06.2022, Moscow, Russia.

# - At an expanded meeting of the Department of Internal Diseases (minutes No. 10 / A dated 06/29/2022) NJSC "MUK".

# Publications

Published 19 scientific papers, including 7 articles, 12 theses. Of these, 3 works in publications of the Scopus rating agency, 3 articles in journals recommended by the Committee for Control in Education and Science of the Ministry of Education and Science of the Republic of Kazakhstan. The results of the study on clinical data were reported at the 5th conference in the form of 4 poster presentations and 2 oral presentations. 3 certificates were received on entering information into the state register of rights to objects protected by copyright (KZ No. 13260, KZ No. 13249, KZ No. 31084).

# Materials and methods of research

**Study design: Cohort prospective study**

Assessment of clinical factors and determination of gene polymorphisms

Assessment of the development of new cardiovascular events during the year

Recurrent MI, restenosis of the coronary arteries, early postinfarction angina pectoris

Life-threatening arrhythmias

Total mortality

AHF and decompensated CHF

Evaluation of the nature of the relationship between polymorphisms in the genes of the cardiological panel and cardiovascular events

Control group (n=91)

Main group (patients with ACS, n=163)

Stroke or TIA

 The scientific study was approved by the ethical committee of NJSC "Medical University of Karaganda" (protocol No. 32 dated December 23, 2019). This study is a clinical molecular genetic study, study design: cohort prospective study.

The examinations were carried out with the voluntary consent of the studied patients. Clinical examinations were carried out in cardiology, including the department of interventional cardiology at the Multidisciplinary Hospital No. 1 and Multidisciplinary Hospital No. 2 in Karaganda. The molecular genetic part of the study was carried out at the Collective Use Laboratory of NAO "Medical University Karaganda

 Statistical processing of the obtained data was carried out using the SPSS software package. The nature of the distribution for the normality of quantitative data was assessed by the Kolmogorov-Smirnov test, the number of observations was more than 50 determinations: the main group - 163 patients, the control group - 91 patients.

The type of distribution of genetic data was determined in both groups. With a normal distribution of quantitative data, mean values (M) and standard deviation (SD) were used for description. In case of non-normal distribution, quantitative data were described based on the median (Me), upper and lower quartiles (Q25, Q75).

To describe the qualitative data, the proportion of individuals with the trait of interest and the 95% confidence interval of the proportion calculated using Pearson's χ2 method was calculated.

The Mann-Whitney U-test was used to compare two independent samples on a quantitative basis. Logistic regression analysis was performed to determine the predictive value of adverse cardiovascular events.

 Statistical analysis of genotyping data was carried out using the SNPStat program.

For each polymorphism included in the genotyping panel, major and minor alleles, the minor allele frequency index (MAF – minor allele frequency), relative values for alleles and genotypes, as well as the p value index when calculating the Hardy-Weinberg law (HWE - Hardy– Weinberg equilibrium).

The assessment of the association of genetic polymorphisms with disease/condition, clinical factors was carried out in accordance with the case-control design based on the generalized linear model (GLM), assuming the main models of inheritance (recessive, dominant and log-additive).

# Conclusions:

1. The risk of new adverse cardiovascular events after PCI increases in elderly patients (OR = 1.07; 95% CI (1.03: 1.12)), as well as in initial overweight (OR = 3, 9; 95% CI (1.2:12.4)), obesity 1 (OR=12.6; -95% CI (2.04:77.8) and grade 2 (OR=7.9; -95 % CI (1.2-54.9)).The risk increases with cardiac arrhythmia (OR=8.9; 95% CI (1.17:69.0) and increases significantly with the development of chronic heart failure (respectively: OR =25; 95% CI (2.8:214.0)).Of the laboratory markers, the most significant predictor of the risk of developing cardiovascular complications was an increase in the level of low-density lipoproteins (OR=1.3; 95% CI (1.001:1.8) ).

2. Cardiovascular events increase after interventional interventions on the coronary arteries in carriers of mutant alleles of the gene polymorphism: Rs 1234313 -TNFSF4; OR=4.5 (p<0.0001); Rs 3184504 - SH2D3. T/T; OR=2.5 (p<0.0001); Rs 2943634; OR=2.5 (p=0.013); Rs 2713604 - DNAJB8-AS1; OR = 4 (p<0.0001).

3. Targeted panels of genetic markers associated with the risk of developing cardiovascular events after percutaneous coronary intervention in the ethnic group of Kazakhs include:

- genotypes of inflammation and immune response biomarker genes: rs1234313 (TNFSF4, p=0.000), rs3184504 (SH2B3, p=0.000), rs2340690 (HSPE1, p=0.002), rs6725887 (ICA1L, p=0.002);

- genotypes of biomarker genes of the hemostatic system: rs1799963 (CKAP5, p=0.000), rs1799983 (CKAP5, p=0.008), rs688034 (SEZ6L, p=0.045);

- genotypes of lipid metabolism biomarker genes: rs268 (LPL, p=0.022), rs2229616 (MC4R, p=0.000), rs2943634 (p=0.030), rs599839 (PSRC1, p=0.030), rs2774279 (USF1, p=0.006);

-genotypes of vascular endothelial biomarker genes: rs1800783 (NOS3, p=0.000), rs1051730 (NOS3, p=0.001), rs10116277 (CDKN2B-AS1, p=0.001), rs2383207 (CDKN2B-AS1, p=0.004), rs2713604 (DNAJB -AS1, p=0.000), rs9536314 (KL, p=0.000).

4. Predicting a high risk of developing cardiovascular events in the long-term period after percutaneous coronary intervention is associated with the identification of: intergenic polymorphism of the rs2943634 gene (OR- 4.0 times, p=0.006).

5. Predictors of the development of new cardiovascular events in the early period after percutaneous coronary intervention were determined: old age, overweight, obesity of 1-2 degrees, an increase in low-density lipoproteins, as well as the presence of cardiac arrhythmias and chronic heart failure. In the long-term period after percutaneous coronary intervention, the prognostic criterion for the development of cardiovascular complications was the heterozygous AS genotype of the intergenic variant rs2943634 (OR=4.0; 95% CI (1.502:10.692), p=0.006)), which is a biomarker of lipid metabolism disorders.

**Practical recommendations:**

1. Determination of clinical and laboratory markers in patients after percutaneous coronary intervention in the form of advanced age, overweight, obesity of 1-2 degrees, increased levels of low-density lipoproteins, cardiac arrhythmias, and chronic heart failure allow predicting the risk of developing new cardiovascular events that determine the indications for genetic testing to identify gene polymorphisms responsible for the risk of developing cardiac complications.

2. The late period after percutaneous coronary intervention determines the indications for genetic testing to identify the rs2943634 gene polymorphism associated with late cardiac complications.

An algorithm for predicting the risk of developing new cardiovascular events after percutaneous coronary intervention has been developed (Certificate for objects protected by copyright No. 31084 dated 12/14/2022).

**Algorithm for predicting the risk of developing new cardiovascular events after percutaneous coronary intervention (PCI)**

New cardiovascular events

Lifestyle modification (correction of risk factors)

High risk of early cardiovascular events

Gene polymorphism

rs2943634

High risk of late cardiovascular complications

Monitoring patient adherence to lifestyle modification and treatment

Genetic screening of first-line relatives

Genetic testing of rs2943634 polymorphism

 Risk factors for cardiovascular events:

 Elderly age

 Overweight

 Obesity 1-2 degrees

 Increasing the level of low density lipoproteins

 Heart rhythm disorder

 Chronic heart failure

Patient with acute coronary syndrome after PCI

yes

no

Gene polymorphism

rs2943634