chemiluminescent analyze which is very sensitive to the blood free radicals level.

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MOLECULAR-GENETIC PRE-CONDITIONS FOR THROMBOSIS DEVELOPMENT

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One of the most important tasks of medical practice in modern society is an early detection of thrombophilic states, as various forms of thrombophilia are the origin causes of such severe diseases as infarctions, insults, and also the cause of operative intervention, pregnancy and inflammation complications resulting in disability and lethal outcomes.

Arterial and venous thromboses development risk can both be connected with the effect of acquired risk factors (operative therapy, oncologic and cardiovascular diseases, atherosclerosis, inflammation, pregnancy, stresses, etc.) and be of hereditary nature. According to modern ideas the thrombotic complications at cardiovascular diseases in 50-65% of the cases (on various authors' evidence) are connected with the defects of the genes controlling the hemostasis system components.

The **research purpose** is to find out the hemostasis system genetic mutations occurrence frequency, endothelial factors and define the significance of analysis for the diagnosis, purposeful pathogenetic therapy choice in the patients with thromboses of various localization vessels.

Materials and methods: The examination on the mutations in the hemostasis system, folate cycle, the genes controlling the vascular wall state and drug metabolism in the liver, warfarin (CYP2C9) in particular, was carried out for the purpose to estimate the predilection to the thrombophilia development and to define individual sensibility to warfarin.

The material for the molecular-genetic analysis was venous blood taken into plastic tubes with EDTA. For the hemostasis system investigation the tubes with citrate were used. The definition of allelic variants of the genes investigated was carried out by the polymerase chain reaction method with the following analysis of restriction fragment length polymorphism (PCR/RFLP).

48 patients aged from 24 to 59 years old with the diagnoses myocardial infarction, ischemic insult, and lower limbs deep venous thrombosis were examined. The first group (29 patients) was formed by the persons, who the treatment was prescribed after the investigation of the hemostasis system functional state and simultaneous molecular-genetic analysis; the 2nd group (19 persons) was made up of the patients appealed for the consultation from other medical and prophylactic institutions with non-effective therapy of the present disease and difficulties in drug dosage of the indirect anticoagulant – warfarin. For the warfarin sensibility estimation two quantity indexes were used: induction phase duration – terms of the INR therapeutic level achievement (number of days) and weekly warfarin dosage (mg), which was required for maintaining of the achieved effect.

Results: The heterozygous mutation of the folate cycle enzyme methylenetetrahydrofolate reductase (MTHFR), the polymophous substitution 677C->T (A223V) was registered by us in 56% of the cases; the homozygous mutation was registered in 5 patients. In 21% of the examinees the methionine synthase (MTR) gene mutation, the polymorphous substitution 2756A->G(D919G) was detected. These mutations lead to the substitution of amino acid residues in the polypeptide chain of enzymes, that decreases their specific activity. One of the manifestations of the MTHFR and MTR deficit is an abundant accumulation of homocysteine in blood that results in the negative influence on the endothelium, disturbing the cell permeability and decreasing the nitrogen oxide production. The hyperhomocysteinemia was detected in 21,6% of the cases, in three patients among them the homocysteine level exceeding the normal one 1,8 times.

From the number of the examinees with repeated infarctions and/or ischemic insult, deep venous thrombosis there were polymorphous variants of the genes controlling the thrombocytic hemostasis registered. In 24% of the cases the polymorphous locus containing variable number of tandem repeats (VNTR) of the thrombocytic glycoprotein gene 1b(GP1ba). In 32% of the cases the polymorphous substitution 1565T→C (Leu33Pro) of the IIIa thrombocytic glycoprotein gene (integrin beta 3), and in 18% - the polymorphous substitution 807C→T of the Ia thrombocytic glycoprotein gene (Gp1a-integrin - alpha -2).

The FII prothrombin gene mutation connected with the substitution of G with A (20210 G→A) and leading to the protein and prothrombin level increase in blood, was registered in our patients in 26% of the cases.

The most severe thrombophilia cases were characterized by the **combined** polymorphous variants of the genes of the thrombocyitc hemostasis, folate cycle, coagulative part of hemostasis and polymorphism of the genes of the endothelial part – the plas-

minogen activator inhibitor (PAI-1), the polymorphous substitution 675 5G→4G (9% in our research) and the gene of the endothelial NO-synthase (NOS3(e)), VNTR-polymorphism and the polymorphous substitution C→T(Glu298Asp) (11% of the cases). It is known that NO (nitrogen oxide) is a powerful vasodepressor, possesses antithrombotic action, inhibiting adhesion and thrombocyte aggregation, activating the tissue-plasminogen activator and other important antithrombotic functions of blood. The NOS3(e) expression or transcription disturbance at the gene mutation results in the NO synthesis decrease, the consequence of which is the vasoconstriction increase, vasodilatation decrease and the tendency to blood clot organization. The polymorphous variant 4G of the PAI-1 gene is attended by the gene's overexpression and, consequently, results in the PAI-1 increase in blood, therefore the fibrinolytic system activity decreases significantly.

All the patients taking part in this investigation needed a long or a life-long application of the indirect anticoagulant – warfarin, that required an adjustment of the preparation dosage to avoid the overdosage. When defining the warfarin sensibility 29 persons from the number of the examined patients had the most popular genotype CYP2C9*1/*1, 39,6% - turned out to bear the alleles CYP2C9*2 and CYP2C9*3 (11 and 8 persons accordingly). The CYP2C9*2 and CYP2C9*3 alleles bearers had unstable INR indexes and required special attention at the warfarin optimal dosage adjustment.

Conclusion: The evidence of the role of genic disturbances in the development of thrombophilias has been obtained. It is shown that the patients with homozygous variants of mutation alleles of the MTHFR genes (C677T), thrombocytic glycoproteins, endothelial NO-synthase and PAI-1 are subjected to the most severe course of thromboses and are hardly treatable. The plasminogen activation inhibitor and endothelial NO-synthase can be used as markers characterizing the endothelium state and for the definition of endothelial dysfunction.

The data obtained point out to a real possibility to influence this or that part of the hemostasis system and/or the system of fibrinolysis, correct the endothelial dysfunction at early stages of the disease depending on the genes' polymorphous variants form. The detection of thrombophilia predisposition genes gives an opportunity to take the preventive measures timely. A purposeful action on the thrombocytic part, coagulative hemostasis or vascular wall in the persons with genic disturbances allows avoiding severe complications at the influence of acquired risk factors on the body. At the necessity of the indirect anticoagulant warfarin adequate dosage adjustment in conditions of its long or life-long application the genetic testing allows forecasting the response to the given preparation intake with due consideration of the patient's sensibility.

The introduction of molecular-biological methods into the laboratory practice, the mutations of the genes coding tissue factors, proteins and glycoproteins of the vascular, thrombocytic, coagulative parts of the hemostasis system and the system of fibrinolysis in particular, can promote the definition of fine mechanisms of clotting and anticlotting blood systems' disturbances. An integrated research including the comparison of various hemostasis system parts' genes' mutations presence with the system's functional state is perspective in both practical and scientific relations, as it allows defining meaningful factors having an effect on the pathological process development and performing a search of pathgenetically relevant methods of treatment. The genetic typing of the hemostasis system parameters and the factors characterizing the endothelium function as the thrombophilia predisposition criteria should be included into the examination record of the patients subjected to either acquired or inherited risk factors.

At the present development stage of the biological and medical sciences the role of molecular-biological mechanisms in the formation of thromboses should be paid considerably much attention to in the programs of biological and medical departments' students training.

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HUMORAL-METABOLIC IMBALANCE IN MEN AND WOMEN SUFFERING FROM CARDIO-VASCULAR METABOLIC SYNDROME

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The purpose of the paper was the analysis of sex differences of basal insulinemia (BI) and malondialdehyde (MDA) – low density lipoprotein oxidation value (LDL), interrelation. 143 men aged 47,6±0,5 and 83 women aged 48,3±0,7 with metabolic syndrome (MS). Normotensive men and women without abdominal adiposis (AA) and dyslypoproteidemia formed control groups. The anthropometric characteristics were defined: lipidogram parameters - by the enzymatic calorimetric method using chemical agents "Vital Diagnostics", insulin basal parameters – by the radioimmunoassay technique with the help of Immunotech Insulin Irma sets, glycemia – by the glucose oxidase test. The LDL oxidation resistance was defined on the MDA levels (nmol/1mg of β-lipoproteins albumin) by fluorometry.