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INFECTIOUS RISK FACTORS FOR IMMUNE IMBALANCE AND COAGULOPATHY IN PATIENTS WITH VENOUS THROMBOSIS AND PULMONARY EMBOLISM

Key words: coagulopathy, viral infection, immune disorders heteroneneity, risk of thrombosis.

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Clinical heterogeneity of a vascular pathology, including coagulopathies, is caused by virus persistence [1]. Primary disturbances of barrier phagocytic function and disturbance of immunoglobulins synthesis can be mediated by the mutations of genes influencing at stages of B-lymphocytes maturation. Viruses cause changes of many metabolic reactions that are not studied up to the end.

As viruses cause cells infection of vessels intima, it causes rising of an expression of molecules of adhesion and stimulation of proinflammatory cytokines synthesis [2].

Research objective: disturbances of immunoresistance and indicators of hemostasis in patients with various kinds of thrombophilias are the interest of our study.

MATERIALS AND METHODS

49 patients with deep vein thrombosis (DVT), 10 patients with inferior vena cava thrombosis (IVCT) and 13 patients with thromboembolism of pulmonary artery (PE) were examined. The frequency of occurrence of antibodies to CMV, EBV-1 and to *Mykoplasma hominis* (ELISA-method), changes of phagocytic function and activity of enzymatic systems of neutrocytes (NST-TEST), concentration of immunoglobulins A, M, G, concentration of peptides of average molecular mass, concentration of circulating immune complexes (CIC) and size of CIC constant, expression of differentiation clusters $CD3^+$, $CD4^+$, $CD8^+$, $CD19^+$, $CD31^+$, $CD50^+$, $CD54^+$, $CD89^+$, $CD162^+$, HLA-DR^+ were investigated.

RESULTS

In patients of all examined groups with venous pathology the increase of IgG — antibodies to a cytomegalovirus of the human CMV in 89% of patients, IgG antibodies to Epstein–Barr EBV-1 virus in 48% of patients, and in 40% competitive presence both pathogens was observed, and also presence of IgG antibodies to *M. hominis* in 69% of patients was revealed. In all groups of patients with venous thrombosis of various localization antibodies to a virus infection disturbance the increase of phagocytic activity of granulocytic neutrophils was revealed. These data which characterize function of enzymes and formation of superoxide-anions, are characterized by an indicator of average cytochemical factor (ACF) in spontaneous (ACF (SP) and induced by a zymosan restoration of nitroblue tetrazolium (ACF (BT) with definition of index stimulation (IS) are proved by true authentic depression of all indicators of the NST-TEST.

In group of patients with a deep vein thrombosis (DVT) rising of phagocytic index (quantity of cells which have entered a phagocytosis) to (82.3±2.34%) became perceptible. Therefore, larger in comparison with a reference group the quantity of neutrophils participates in a phagocytosis. The average cytochemical factor (ACF) in the spontaneous NST-TEST (ACF (SP), which reflects activity of cationic fibers in respiratory and enzymatic processes of phagocytes, is lowered to (0.44±0.18) arbitrary units at its control value (1.15±0.12) arbitrary units. Therefore, in this group the power processes providing an operating time of biooxidizers with bactericidal action are lowered. In stimulated NST-TEST, ACF made (0.94±0.3) arbitrary units and has been lowered. In its turn it has negatively affected stimulation reserve to that testifies the lowered index stimulation (IS) (1.61±0.19) which referential size is in limits from 5 to 10 units. Lowered IS value is regarded as consequence of attrition of phagocytic function against long viruses persistence. Incompleteness of a phagocytosis can lead to that infectious agents in a hematogenic way spread on the circulation system, and at specific features of an endothelium of vessels at chronic inflammatory processes with involving of mediators of an inflammation of cytokines there is a change of mural and platelet factors of coagulation with initiation of thrombogeneses. In comparison

with data of a reference group in this group lowered maintenance of A immunoglobulin, which carries out barrier function is revealed. Depression of barrier functions upsets the general physiological resistance of an organism. The expression of differentiation clusters of T-lymphocytes $CD3^+$ and $CD4^+$ in group of patients with DVT that testifies on secondary acquired immunodeficiency condition caused by a virus infection is lowered. There is revealed increase of expression of differentiation clusters of $CD8^+$ T-killers subpopulations T-lymphocytes (to (26.5±5.84)%) at value in control group (23.4±4.4)%), that could be possible to survey as depression of an autoimmune component because of autolytic function of T-cellular link. The appreciable role plays receptors of $CD31^+$ in adhesion regulation to an endothelium of separate subpopulations of T-lymphocytes. At linkage of $CD31^+$ with immune competent cells there is an activation of integrin with the subsequent stimulation of autolysis. The expression of the given receptor is authentically lowered in 20% at patients with DVT to (13.14±3.29)%, the expression of receptor of molecules of adhesion $CD50^+$ (51.17±4.00)% on leucocytes is authentically increased. Depression of expression of $CD54^+$ to (14.67±1.61%) can be a consequence of linkage of receptors with various virus ligands (effect of consumption), and it has been revealed in our researches at referential value of this indicator (18.5±3.6%). Reduction of expression of $CD89^+$ at DVT to (4.86±1.98%) at referential value (30.0±3.0%) also is connected with the presence of associated persistent infection. At the same patients appreciable depression of adhesion and endocytosis of englobing cells is revealed. In this group activity of native physiological anticoagulant of antithrombin III has made (78.09±4.06%), 40% of them — in the range of insufficiency, in the system of protein C insufficiency wasn`t revealed.

At patients with inferior vena cava thrombosis (IVCT) there was revealed appreciable depression of a phagocytic index that testifies neutrophils lose ability to absorb foreign antigens, for example, the cells affected by a virus, and also circulating immune complexes, that is proportional to concentration of CIC in this group of patients, where concentration of CIC with high molecular mass to (127.0±3.0) arbitrary units is increased. $CD8^+$ T-killer population of lymphocytes is lowered to (15.0±0.03%), and also the expression of markers of a differentiation of $CD3^+$ (to 34.5±1.5%) and $CD4^+$ is lowered in 2 times to (19.5±2.5%), that at considerably increased concentration of immunoglobulin G allows to characterize this condition as a chronic inflammation at virus infection. In this group of patients patholologically lowered activity of an antithrombin III (66.17±16.18%) and the greatest frequency of occurrence of insufficiency of this native physiological anticoagulant (67%), and also insufficiency in the system of other native physiological anticoagulant of protein C — in 14% of patients is observed.

In group of patients with thromboembolism of pulmonary artery (PE) the following disturbances of phagocytosis process of neutrophilic granulocytes: phagocytic number to (2.67±0.32) is considerably lowered, that testifies to depression of ability to absorption of foreign antigens, to disturbance of processes of a chemotaxis, adhesion, endocytosis that makes impossible realization of barrier function of a phagocytosis. ACF (SP) is below the control and also makes (0.92±0.11) in group of patients with PE, that can negatively influence completeness of a phagocytosis, i.e. there are not enough enzymes for realization of respiratory explosion. Minimum IS value is (1.43±0.15) and depression of reserve of sensitivity NG to stimulation, was observed at patients with PE. It can testify the attrition of phagocytic function against long virus persistence. In group of patients with PE the increased maintenance of M and G immunoglobulins in comparison with reference group data is revealed. The maintenance of CIC (125.2±10.6) arbitrary units that correlates with high concentration of immunoglobulins in serum of these groups of patients is increased. Patients with PE were characterized by high level of intoxication (PSMM are increased to (0.344±0.032) arbitrary units in comparison with (0.233±0.012) arbitrary units in a reference group), therefore, the massive thrombotic lesion, and the higher intoxication level. $CD31^+$ plays an appreciable role in adhesion regulation to an endothelium of separate subpopulation of T-lymphocytes. At linkage of $CD31^+$ with immune competent cells there is an activation of integrin to the subsequent stimulation of autolysis. The expression of the given receptor is authentically lowered in 31.25% in patients with PE to (11.0±0.02%) and it could lead to the increased risk of aggregation of thrombocytes. The depression of $CD54^+$ expression to (12.0±0.01%), connected with disturbance of the cascade of adhesion is revealed. Frequency of occurrence in the specified group of insufficiency of native physiological anticoagulant of an antithrombin III made 45%, thus in research of activity of protein C the insufficiency in this group has revealed in 50% of patients.

In patients with a venous pathology of various localization the multifactorial disturbances of a hemostasis arising against associated virus and bacteremia infection, namely different kinds of insufficiency of native physiological anticoagulants that can be bound to insufficiency of a complement system are revealed.

The multi-directed changes of immune resistance against disturbance of factors of coagulation and concentration of natural anticoagulants are also revealed. The examined groups have clinical heterogeneity in connection with gravity of a condition, prescription of disease, presence of an accompanying pathology.

Thus, in each group it is necessary to allocate the following leading risk factors:

In "deep vein thrombosis" group — disturbance of phagocytic barrier function against substantial increase of concentration of specific Ig-antibodies; decrease of killer function of T-lymphocytes, increase of expression of adhesion $CD50^+$ molecules, decrease of expression of differentiation $CD89^+$ clusters, insufficiency in system of antithrombin III (40%) were observed.

In "inferior vena cava thrombosis" group — disturbances of phagocytic barrier function with the high maintenance of circulating immune complexes, insufficiency in system of antithrombin III (67%) and protein C (70%) were investigated.

In "thromboembolism of pulmonary artery" group — substantial increase of concentration of immunoglobulins of all classes, increase of concentration of specific Ig-antibodies, especially to Epstein–Barr virus, an appreciable endogenous intoxication, insufficiency in system of antithrombin III (45%) and protein C (50%) were defined.

Thus, viruses impair immune resistance and cause disturbances in hemostasis system; the effect of their action is aggravated with presence of hereditary factors that increase heterogeneity of clinical forms when action of viruses strengthens insufficiency of other factors of protection.

Under the influence of virus persistence various epigenomic compensator-adaptive reactions which can lead to augmentation of quantity of risk factors, serious current of disease are formed.

Condition severity level depended on degree of a lesion of immune system and quantity of risk factors of general resistance, expression degree correlated with depression of a complex of immunologic factors and presence of virus mixed-infection unlike a mono infection.

Parameters of the adverse complicated current: general is a depression of general resistance of an organism, mixed-infection presence, increase of quantity of risk factors is combination of deficiencies of native anticoagulants and depression of general resistance of an organism against coagulopathies and severity level depends on frequency virus persistence.

REFERENCES

1. *Imbronito A. V. Detection of human cytomegalovirus and Epstein–Barr virus in coronary atherosclerotic tissue / A.V. Imbronito, S.L.Marcelino, S.R. Grande, F.D. Nunes, G.A. Romito // Brazilian J. Microbiology. — 2010. — Vol. 41. — P. 563–566.*
2. *Kimura H. Clinical and virologic characteristics of chronic active Epstein-Barr virus infection / H. Kimura, Y. Hoshino, H. Kanegane et al. // Blood. — 2001. — Vol. 98, № 215. — P. 280–286.*
3. *Kreutz R., Bliden K., Tantry U. et al. Viral respiratory tract infections increase platelet reactivity and activation: an explanation for the higher rates of myocardial infarction and stroke during viral illness // Journal of Thrombosis and Haemostasis. — 2005. — № 3. — P. 2108–2109.*

ІНФЕКЦІЙНІ ФАКТОРИ РИЗИКУ У РОЗВИТКУ ІМУННОГО ДИЗБАЛАНСУ І КОАГУЛОПАТІЙ У ХВОРИХ З ТРОМБОЗОМ ВЕН І ТРОМБОЕМБОЛІЄЮ ЛЕГЕНЕВОЇ АРТЕРІЇ

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З метою вивчити роль поєднаних вірусних і бактеріальних інфекцій інфекційних чинників ризику в розвитку імунного дизбалансу і коагулопатій обстежено 49 хворих тромбозами глибоких вен, 10 хворих тромбозом нижньої порожнистої вени і 13 хворих с тромбоемболією легеневої артерії. Досліджували частоту зустрічальності антитіл до CMV, EBV-1 і до *Mycoplasma hominis* (методом ІФА), фагоцитарну функцію і активність ферментативних систем нейтрофілів (НСТ-тест) (світлова мікроскопія), концентрацію імуноглобулінів A, M, G (імунотурбідіметрія), концентрацію пептидів середньої молекулярної маси (спектрофотометрія), концентрацію циркулюючих імунних комплексів (ЦІК) і величину константи ЦІК (спектрофотометрія), експресію кластерів диференціювання $CD3^+$, $CD4^+$, $CD8^+$, $CD19^+$, $CD31^+$, $CD50^+$, $CD54^+$, $CD89^+$ (флуоресцентна мікроскопія). У пацієнтів всіх обстежених груп з вказаною патологією виявлена вірусна персистенція CMV і EBV-1. Провідними факторами ризику в кожній групі були такі: у групі "тромбоз глибоких вен" — порушення фагоцитарної бар'єрної функції на тлі значного підвищення концентрації специфічних Ig-антитіл; зниження кілерної функції Т-лімфоцитів, підвищення експресії молекул адгезії $CD50^+$, зниження експресії кластерів диференціювання $CD89^+$, недостатність в системі антитромбіну III (40 %). У групі "тромбоз нижньої порожнистої вени" — порушення фагоцитарної бар'єрної функції з високим вмістом циркулюючих імунних комплексів, недостатність в системі антитромбіну III (67 %) і протеїну С (70%). У групі "тромбоемболія легеневої артерії" — значне під-

вищення концентрації імуноглобулінів всіх класів, підвищення концентрації специфічних Ig-антитіл, особливо до вірусу Епштейна–Барра, значна ендогенна інтоксикація, недостатність в системі антитромбіну III (45 %) і протеїну С (50 %). Таким чином, віруси порушують імунорезистентність, викликають порушення в системі гемостазу, ефект їх дії посилюється наявністю спадкових факторів, що підвищує гетерогенність клінічних форм, коли дія вірусів підсилює недостатність інших факторів захисту.

Ключові слова: коагулопатії, вірусна інфекція, гетерогенність імунних порушень, ризик тромбозів.