

Influence of exogenous, physiological and factors of chronic inflammatory process on indicators of disturbances of adaptive processes of the person

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Changes in laboratory parameters can be caused by physical, chemical, biological and other external factors, depending on physiological conditions, in most cases they have an adaptive nature, and it is not always possible to distinguish them from physiological fluctuations. At the same time, obvious shifts in laboratory parameters may indicate a violation of adaptation processes and damage of tissues and organs, despite the absence of clinical manifestations of a disease.

Introduction

This article systematizes the evidence on the influence of environmental, nutritional, medicinal and other exogenous factors and physiological conditions on laboratory parameters of the hemostasis system, as well as violations of platelet hemostasis in

inflammation. The material of the article will help to detect pre - pathology in clinically healthy individuals, address issues of early diagnosis, prognosis, evaluation of efficacy and safety of therapy, determine the degree of positive and negative effects of external and physiological factors, prevent misinterpretation

of laboratory parameters, diagnostic errors, adverse treatment consequences.

All etiological factors of inflammation are divided into exogenous and endogenous ones.

Exogenous (external) inflammatory factors include: amicroorganisms (viruses, bacteria, fungi); chemicals (acids, alkalis); animal organisms (worms, insects, protozoa); thermal factors (heat, cold); ionizing and ultraviolet rays; mechanical factors (foreign bodies, rupture, pressure).

Endogenous (internal) inflammatory factors include those that occur in the body as a result of another disease: inflammation can develop as a reaction to the tumor; inflammation can occur as a reaction to gallstones and urinary stones; inflammation can occur as a reaction to a blood clot that has formed in the blood vessels. antigen + antibody complexes, which are fixed in a specific organ.

The main causes of inflammation are: necrobiotic changes in tissues and cells that occur under the influence of exogenous physicochemical factors; malignancy of the body's own cells; invasion of foreign microorganisms or antigens, immunogens into the internal environment; loss of immunological tolerance to antigens of the body

Experimental part

Material and methods

Study of the functional state of platelets

To research the state of platelet hemostasis, adhesion, adenosine diphosphate (ADP) - aggregation of peripheral blood platelets in platelet-rich plasma were studied. Platelet counting was performed using a Goryaev counting chamber. The time of ADP aggregation was also determined. The degree of maximum platelet aggregation (**Figure 1**). was investigated (expressed as a percentage difference between the initial number of platelets, which was taken as 100%, and the number of platelets 10 min after the addition of ADP solution to the sample); the degree of platelet adhesion (the difference between the initial number of platelets and the number of platelets after contact with glass under conditions of rotation on an electromagnetic stirrer for 5 min).

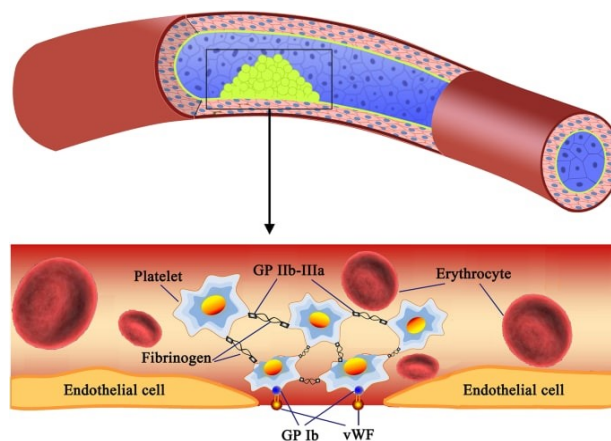


Figure 1. Platelet adhesion

In patients with chronic inflammatory processes, thallium genes were also determined by the content of PV contained in poor platelet plasma using formalized donor platelets by the method of Evans et Osten in the modification of O.A. Tsyguleva.

The principle of the method is that the influence of the investigated PV on the aggregation of washed and formalin-fixed platelets of healthy individuals under the influence of ristocetin is determined. Quantitative determination is performed on the dilution curve of normal mixed platelet-free plasma. The method is based on the fact that platelets treated with a weak solution of formalin retain the ability to ristocetin - aggregation (in the presence of PV), but are not subject to other types of aggregation (spontaneous, under the influence of ADP, adrenaline, thrombin, etc.).

Reagents.

1. 3.8% solution of sodium citrate;
2. Buffer solution (pH 7.6; $2 = \text{KH}_2\text{PO}_4$; 8.02 NaCl , $8.8 = \text{Na}_2\text{HPO}_4$ dissolved in 1 liter of distilled water);
3. Buffered EDTA (ethylenediaminetetraacetic acid) - formalin solution: 3 ml of 0.007 M (0.483%) EDTA, 5 ml of 4% formalin solution, 2 ml of buffer, 10 ml of distilled water;
4. EDTA - buffer solution: 0.77 M EDTA (1 part) with buffer solution (49 parts);
5. Ristocetin solution (10 mg per 1 ml of buffer).

Preparation of suspension of washed fixed platelets of healthy people: 9 parts of venous blood of donors are mixed with 1 part of EDTA - formalin solution; in which blood is injected directly from a puncture needle.

Centrifuge for 5 min at 1500 rpm to obtain platelet-rich plasma, in which hemolysis can be expressed. From this plasma, platelet-poor plasma is obtained by centrifugation for 20 min at 4000 rpm. Platelet-poor plasma is removed, and the platelet pellet is washed twice with EDTA buffer solution (4 times the volume by centrifugation each time for 10 min at 4000 rpm). The buffer containing EDTA is removed by suction, and the platelet precipitate is diluted in a buffer solution without EDTA so that 1 μl contained about 200,000 Tr.

Concentration of normal platelets: Normal formalized platelets can be preserved so as not to prepared them again each time. To do this, after washing with EDTA - buffer solution (twice) they are placed in phosphate buffer (pH 7.4) with 0.01% sodium azide solution.

The suspension is packaged in 3 ml sterile vials, rolled up and stored in a freezer (at -10 -12 °C) for 2-3 months. If necessary, the contents of the vials are thawed; platelets are washed twice from the preservative solution with buffer without EDTO and used in the test.

Preparation of platelet-free plasma of the subject. Blood is taken from a vein under siliconization conditions and mixed with 3.8% sodium citrate solution (9 :1). Centrifuge for 7 minutes at 1500 rpm. Remove platelet-rich plasma, which is centrifuged for 20 min at 4000 rpm. Platelet-poor plasma is transferred to another silicone or plastic tube and used in the test.

The course of the study.

In 2 cuvettes for PEC with a working face of 5.65 and a volume of 2.5 ml, 1 ml of the suspension of washed normal platelets, 0.4 ml of platelet-free plasma of the subject and 0.4 ml of buffer without EDTA are injected.

0.6 ml of buffer is injected into the control cuvette. Both cuvettes are placed in the PEC and at $\lambda = 630$ nm set the arrow of the PEC on "O" in the cuvette with the test sample, enter 0.2 ml of ristocetin solution, mix, turn on the stopwatch. After 2 minutes, changes in the optical density of the test sample, which are associated with platelet aggregation under the influence of ristocetin, are recorded.

Construction of the dilution curve.

To quantify the presence of Willebrand factor in the studied plasma use a standard dilution curve obtained from a large group of healthy young people with normal platelet-free plasma. This plasma is diluted with buffer without EDTA from 1: 2 to 1:32, after which the washed normal platelets are introduced into each sample according to the above method is determined by ristocetin - platelet aggregation. The calculation point is the dilution of plasma at which ristocetin no longer causes platelet aggregation. This point corresponds to 2 - 3% presence in the environment of the Willebrand factor. The obtained data are plotted on logarithmic paper. With the correct construction of the curve in the white-logarithmic coordinate system has the form of a straight line. Using this

curve, determine the presence of Willebrand factor in the test plasma. For greater accuracy, the latter can also be studied in 2 - 3 dilutions.

1. The influence of exogenous and physiological
2. factors on human adaptation processes.
1. Hemostasis (**Figure 2**) system research indicators.

1.1 Bleeding time. Bleeding time is measured after the formation of a wound on the skin of the earlobe, the flesh of the nail phalanx of the finger, or the upper third of the palmar surface of the forearm. Normal values: 2-5 minutes.

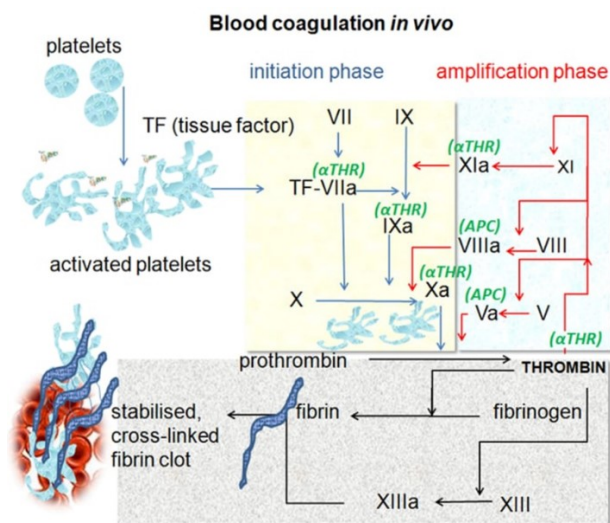


Figure 2. Vascular platelet hemostasis

The increase in bleeding time is due to the influence of nutritional factors: vitamin C deficiency in food; hapten thrombocytopenia when taking certain products, physical factors: thrombocytopenia after exposure to ionizing radiation, chemical factors, thrombocytopenia after exposure to myelotoxic industrial, agricultural and household chemicals - fluorine

and its organic compounds, carbamines, chlorocysts; drugs: thrombocytopenia after exposure to myelotoxic drugs (cytostatics, nonsteroidal anti-inflammatory drugs; thrombocytopenia of immunoallergic (hapten) genesis after taking quinine, quinidine, digitoxin, PAS, sulfanilatide, zilfanilamites, zifanilamides, zifanilamides, zifanilamides, zifanilamides, zifanilamides, metronidazole, dipyridamole, penicillin, indomethacin, delagil, furosemide, in newborns whose mothers in the second and third trimesters took salicylates, sulfonamides, dipyridamole, metronidazole [1].

Reduction of bleeding time occurs under the influence of physiological factors: - pregnancy, III trimester, drugs of influence: - cancellation of anticoagulants. *Capillary resistance.*

A "cuff" test is usually used to study it.

Normal values: the number of petechiae is not more than 10, with a diameter of not more than 1 mm.

Increasing the number and diameter of petechiae:

1. Nutritional factors: deficiency of vitamins C and D in food.

Platelet count

1.2. Platelet ADP aggregation (**Figure 3**) characterizes the ability of platelets to aggregate in the presence of aggregating factors (ADP).

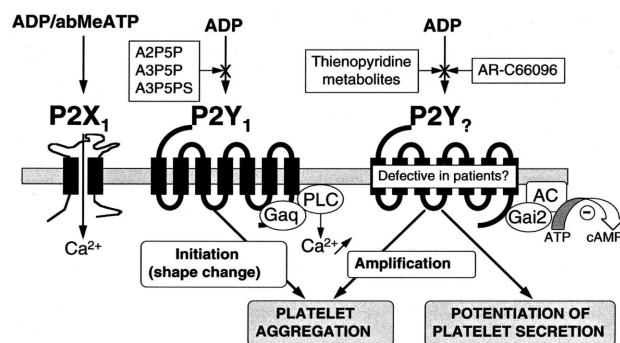


Figure 3. ADP platelet aggregation

Normal values: (according to the method of Baluda, 1996)

Aggregation after 1 minute - $50 \pm 3\%$, maximum aggregation - $66 \pm 3\%$, maximum aggregation time - 3-6 minutes, disaggregation start time - 10 minutes, the degree of disaggregation after 25 minutes - 53%.

Decreased platelet aggregation:

1. Alimentary factors of influence : deficiency of vitamin B12 in food; eating a bite.
2. Physical factors of influence: validity of ionizing radiation.
3. Drugs: thrombocytopeny with the use of nonsteroidal anti-inflammatory drugs (salicylates, pyrazolone derivatives, butadolidine, brufen, ibuprofen, indomethacin, naproxen); antispasmodics (papaverine, eufhylline); antibiotics (carbenicillin, penicillin, prostacyclin); psychotropic drugs (neuroleptics aminazine, dipyridamole; tranquilizers imipramine, amitriptyline); barbiturates, anesthetics (ether, cyclopropane, nitrous oxide); diuretics (phenothiazine derivatives, furosemide); derivatives of nitrofurans, antihistamines (diazoline, diphenhydramine); cytostatic agents

(vinblastine, vincristine); massive blood transfusions and blood substitutes (reopolyglucin); hormonal drugs (steroid hormones, insulin, anabolic steroids, vasopressin); anti-calcium drugs (isoptin, corinfar.); coronary dilators (curantil, sodium nitroprusside); adrenoblockers (phentolamine, yohimbine); fibrinolytics (streptokinase, urokinase); anticoagulants ticlopidine); derivatives of quinine (quinine, quinidine, quinacrine); vitamin preparations (nicotinic acid, tocopherol, cyanocobalamin, ascorbic acid, folic, pantothenic acids, pyridoxine); other drugs (intensaine, anthurane, imidazole and its derivatives, prostaglandin E, forskolin, anagremide, cilostamide, naphthazar, miscleron, anginine, suloctidil).

1.3. Ristomycin-platelet aggregation

The method allows to detect the activity of Willebrand factor (**Figure 4**). A linear relationship has been established between the degree of ristomycin aggregation and the amount of Willebrand factor [2].

Gp IIb-IIIa complex on platelet surface

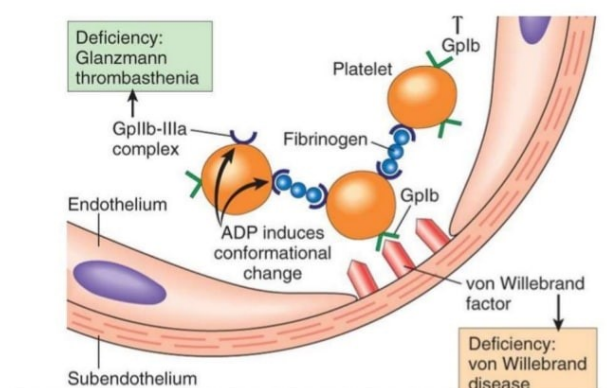


Figure 4. Platelet aggregation with Willebrand factor

Normal values : the minimum aggregating dose of ristomycin is 0.6 ± 0.05 mg / ml

Increased aggregation (reduced minimum aggregation dose):

1. Drugs: taking oral contraceptives, arteparon.

Decrease in aggregation (increase in the minimum aggregation dose):

1. Drugs: taking salicylates, penicillin, indomethacin, delagil, furosemide, in newborns whose mothers took metronidazole, dipyridamole, salicylates during pregnancy (II and III trimesters).

2.Coagulation hemostasis – acrocirculatory, secondary (**Figure 5**).

2.1. The clotting time of unstabilized whole venous blood at 37°C.

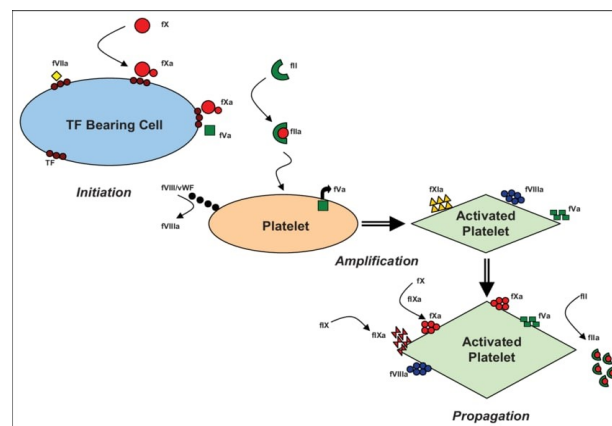


Figure 5. Coagulation hemostasis

Normal values: 4-10 minutes

Increased blood clotting time:

1. Nutritional factors: deficiency of vitamin K in food.

2. Physical factors: validity of ionizing radiation.

3. Drugs: treatment with heparin, direct-acting anticoagulants (phenylene, dicoumarin.).

Reduction of blood clotting time:

1. Physiological factors: pregnancy, III trimester.
2. Chemical factors: validity of dioxins.
3. Drugs: use of oral contraceptives (infecundin, bisecurin, rigevidon); taking psychotropic drugs (thioridazine, chlorprothixene, mepazin, thioproperazine, reserpine, raunatin); large doses of ascorbic acid; in newborns whose mothers took anticoagulants and salicylates during pregnancy (II and III trimesters).

2.2. Cephalin time (partial thromboplastin time)
 Cephalin time is an indicator of the internal system of prothrombin activation (Figure 6). Determine the time of recalcification of platelet plasma in the presence of cephalin (phospholipid activator) [3]. Normal values: 65-80 sec. *Increase in cephalin time:*

1. Chemical factors: dioxin intoxication.
2. Drugs: use of large doses of heparin; in newborns whose mothers took anticoagulants during pregnancy (II-III trimester).

Kaolin-cepahalin time (activated partial thromboplastin time) (APTT)

Kaolin-cepahalin time is an indicator of the internal system of prothrombin activation after the maximum activation of factors XI and XII. Determine the time of recalcification of platelet-free plasma in the presence of kaolin (contact factor) and cephalin (phospholipid activator)[4].

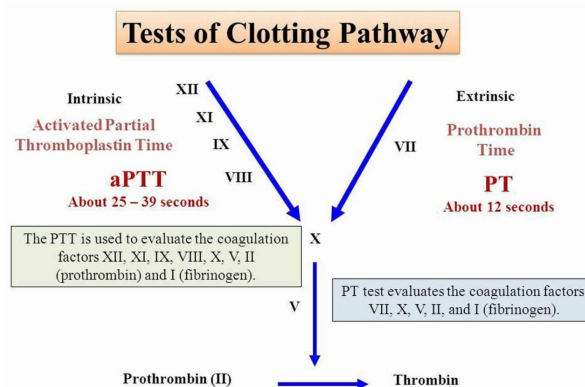


Figure 6. Partial thromboplastin time

Normal values: 38-65 sec.

Increase in kaolin-cepahalin time:

1. Physiological factors: in newborns, especially premature.
2. Drugs: use of large doses of heparin; - in newborns whose mothers took anticoagulants in the second and third trimesters.

Reduction of kaolin-cepahalin time:

1. Physiological factors: pregnancy, III trimester.
- 2.3. Prothrombin (thromboplastin) time

(Figure 7). Prothrombin time is an indicator of the external activation system of prothrombin. It is widely used to monitor treatment with indirect anticoagulants because the study does not require venipuncture and can be performed often enough. Determine the clotting time of plasma with the addition of thromboplastin and calcium chloride.

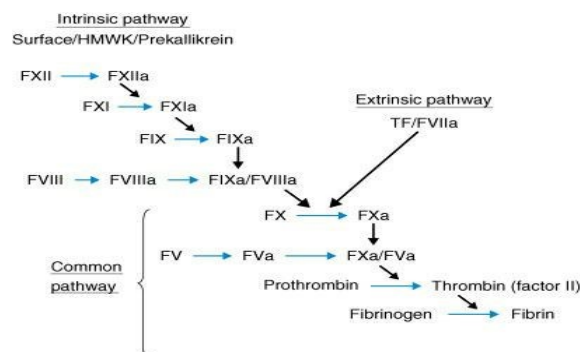


Figure 7. Prothrombin (thromboplastin) time

Normal values depend on the activity of the thromboplastin used, so the prothrombin index (PI) is more often determined.

$$INR = \left(\frac{PT_{Patient}}{PT_{mean-normal}} \right)^{ISI}$$

PT = 12–20 sec; PI = 90–105%

Increase in prothrombin time (PT) - decrease in prothrombin index (PI).

1. Physiological factors: in newborns, especially for 2-3 days; in premature infants up to 2 weeks of age.

2. Nutritional factors: deficiency of vitamin K in food [5].

3. Chemical factors: intoxication with chloroform and other halogenated hydrocarbon compounds, phosphorus, dioxanes.

4. Drugs: treatment with indirect anticoagulants, fibrinolytics, salicylates, novocephalin, citramon, ascofen, anabolic steroids, thiazide diuretics, iron supplements; in newborns whose mothers used indirect anticoagulants during pregnancy (II-III trimesters) (Figure 8).

Decrease in prothrombin time (PT) - increase in prothrombin index (PI):

1. Physiological factors: increased coagulation in pregnant women and during childbirth.

2. Drugs: overdose of vicasol.

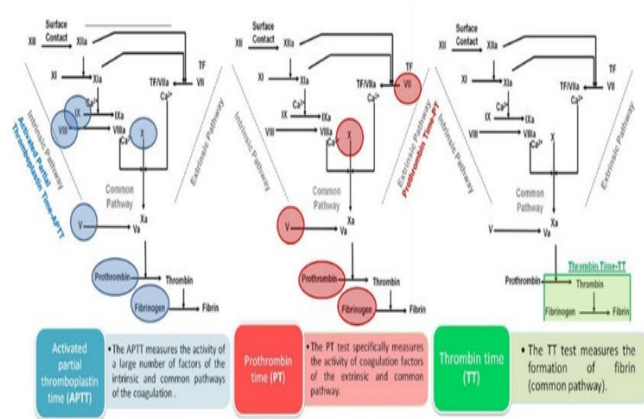


Figure 8. Activated partial thromboplastin, prothrombin, thrombin time

2.2.4. Thrombin time

Thrombin time is an indicator of the time of transition of fibrinogen to fibrin (Figure 9). The clotting time of blood plasma is determined by adding a solution of thrombin with standard activity [7].

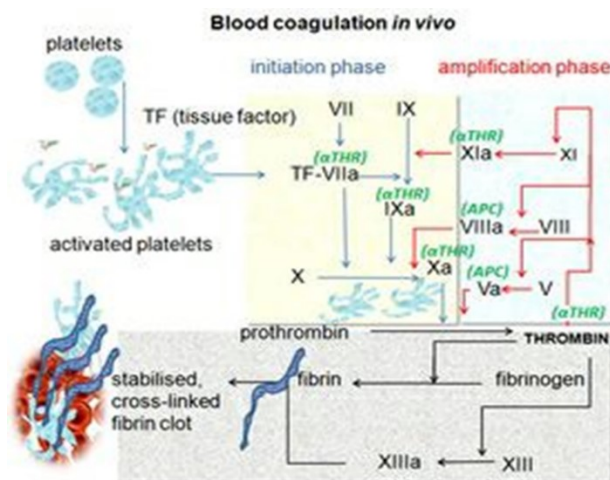


Figure 9. Thrombin time

Normal values: about 15 sec. Plasma does not coagulate:

1. Drugs: treatment with heparin.

2.2.5. Fibrinogen (Figure 10).

Fibrinogen is a precursor of insoluble fibrin, determined by drying the convolution formed

by adding a standard activity of thrombin solution to a certain volume of plasma (**Figure 11**) [6].

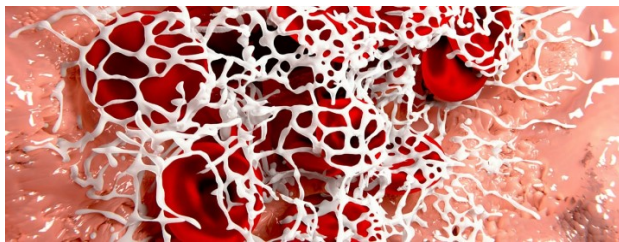


Figure 10. Fibrinogen formation

Normal values: 1.8–3.5 g / l.

Increasing the concentration of fibrinogen:

1. Physiological factors: menstruation; pregnancy, III trimester.
2. Nutritional factors: deficiency of vitamin C in food.
3. Physical factors: radiation exposure.
4. Chemical factors: intoxication with salvarsan, chloroform.



Figure 11. Structure of fibrinogen

Decreased fibrinogen concentration:

1. Chemical factors: poisoning by hepatotropic poisons; chlorinated naphthalenes, chlorinated hydrocarbons (chloroform, carbon tetrachloride), azo dyes, benzene, styrene, phosphorus, arsenic, heavy metals, organophosphorus pesticides.
2. Drugs: taking phenobarbital, streptokinase, urokinase.

2.2.6. Fibrinase activity

Fibrinase, or Factor XIII, or fibrin stabilizing factor, is found in the vascular wall, platelets, erythrocytes, kidneys, lungs, muscles, placenta, and binds fibrin monomers into a fibrin polymer [8]. Normal values: 35 ± 5 s

Decreased fibrinase activity:

1. Physiological factors: pregnancy.
2. Nutritional factors: deficiency of vitamins in food.
3. Physical factors: ionizing radiation.

2.3. Fibrinolytic activity of blood (**Figure 12**)

2.3.1. Time of lysis of euglobulins

The lysis time of the euglobulin fraction (devoid of antiplasmin) of plasma depends on the amount of fibrinogen, plasmin, plasminogen activators. The time of spontaneous lysis of the convolution obtained from the euglobulin fraction of platelet plasma when adding a solution of calcium chloride is determined [9]. Normal values: 100-300 sec.

Activation of fibrinolysis (lysis time of euglobulins is reduced):

1. Therapeutic measures: surgical interventions; taking adrenaline, dextrin, streptokinase and other fibrinolytics.

Inhibition of fibrinolysis (euglobulin lysis time is extended):

1. Physiological factors: pregnancy; in premature infants.
2. Drugs: taking trasilol and other antiproteases, oral contraceptives.

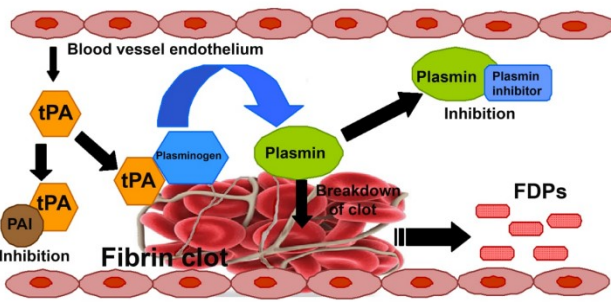


Figure 12. Fibrinolytic activity of blood

2.3.2. Fibrin degradation products (PDF)

PDFs are normally contained in very small quantities and are not defined by commonly used methods. At increase of concentration of PDF to 10 mkg / ml ethanol and protamine sulfate tests become positive (observe formation of a deposit in blood serum after addition to it of solution of protamine sulfate or ethanol). Determination of PDF can also be performed using antibodies against fibrin / fibrinogen fragments (Figure 13) [10].

Positive test:

1. Physiological factors: pregnancy, III trimester.
2. Drugs: treatment with fibrinolytic drugs.

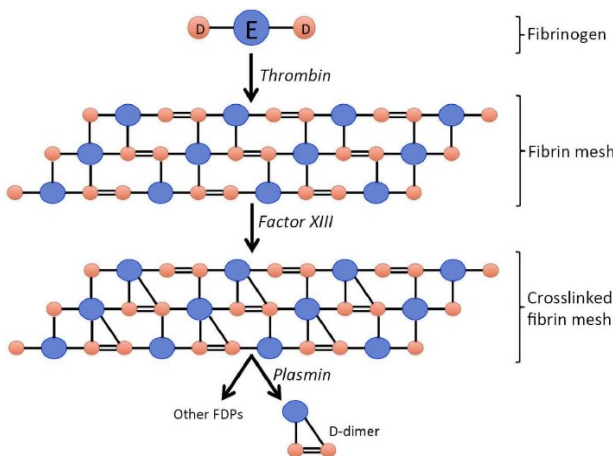


Figure 13. Fibrin degradation products

2.4. Anticoagulants

Protein C

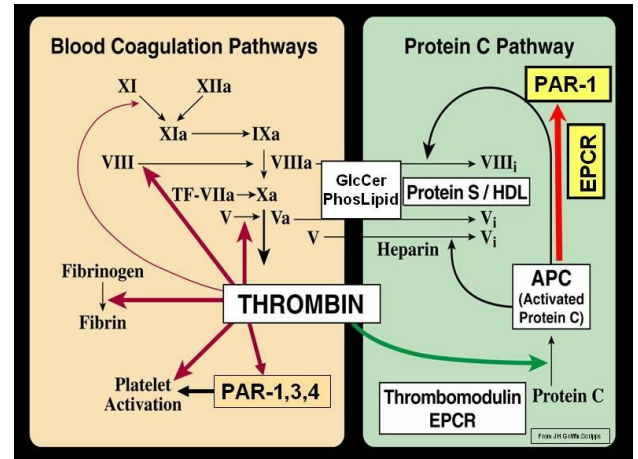
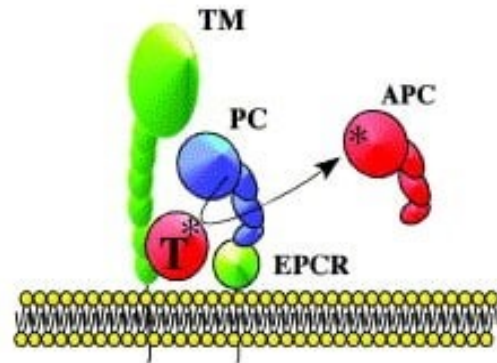


Figure 14. Protein C

A Activation of protein C



B FVa degradation

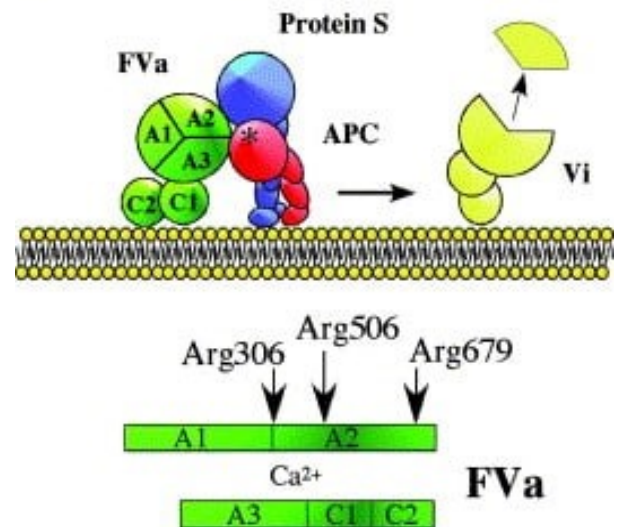


Figure 15. Activation of Protein C

Protein C is an active vitamin K-dependent anticoagulant (Figure 15) [11]. Normal values: about 1 g / l. Reducing the content of protein C:

1. Drugs: treatment with anticoagulants of indirect action (Figure 16).

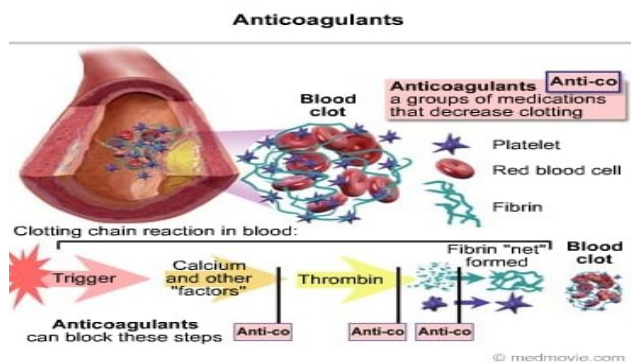


Figure 16. Anticoagulants

2.4.1. Antithrombin III (BP III)

Plasma antithrombin (Figure 17) activity primarily depends on BP III and, to a lesser extent, on α_2 -macroglobulin, α_1 -antitrypsin, C1-esterase inhibitor and others. This test is important for the control of heparin therapy.

BP III is determined after incubation of a mixture of serum and thrombin solution of standard activity. The final thrombin is determined in the thrombin-fibrinogen reaction.

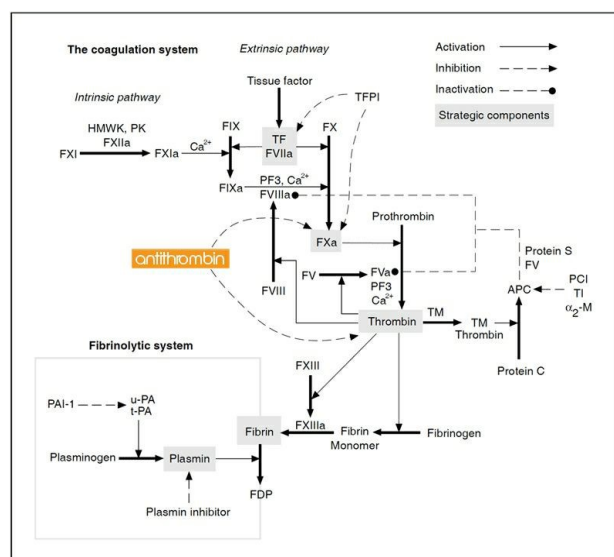


Figure 17. Antithrombin III (BP III)

Normal values: 75–125% (100% is the antithrombin activity of whole donor plasma).
Concentration: 20–50 mg / dl

Increasing the activity of ATIII:

1. Physiological factors: during menstruation.
2. Nutritional factors: vitamin K deficiency in food.
3. Drugs: use of anabolic drugs; taking anticoagulants of indirect action.

Decreased activity of ATIII:

1. Physiological factors: the middle of the menstrual cycle; the last months of pregnancy; postpartum period.
2. Drugs: long-term use of oral contraceptives, estrogens; long-term treatment with corticosteroids; use of clofibrate, norepinephrine, rifampicin; treatment with L-asparaginase; treatment with large doses of heparin (with a decrease in blood pressure of III more than 25%, the effectiveness of heparin therapy is significantly reduced).

Plasma tolerance to heparin. Plasma recalcification time and convolution formation after heparin injection into the plasma are measured. The indicators of this test allow us to judge the interaction of coagulation and anticoagulation mechanisms of the blood coagulation system. Normal values: 11–16 min.

Reducing the clotting time means an increase in plasma tolerance to heparin and indicates activation of the coagulation system. An increase in clotting time means a decrease in plasma tolerance to heparin and indicates an

increase in anticoagulant activity, or a deficiency of coagulation factors [12].

Increasing plasma tolerance to heparin:

1. Physiological factors: the last months of pregnancy.

Decreased plasma tolerance to heparin.

1. Drugs: taking anticoagulants of direct (heparin) and indirect (phenylin, dicoumarin) action.

3. Influence of inflammatory process on indicators of thrombocyte link of hemostasis and their disturbance at chronic inflammation

Currently, there is so much information about the complications of many inflammatory and non-inflammatory diseases, disorders of systemic and local microcirculation, thrombosis and thromboembolism. Including microcirculation disorders are observed in inflammatory diseases of CO. Endothelial activation and dysregulation of endothelial and dendritic cell relationships have been found to accelerate the development of atherosclerosis and cardiovascular disease, because the adhesion and migration of dendritic cells enhance atherogenesis (**Figure 18**). Inhibition of NO synthesis by endothelial cells, hypoxia, oxidized fine LP, tumor necrosis factor (TNF) in inflammation also increase the adhesion and migration of dendritic cells (**Figure 19**).

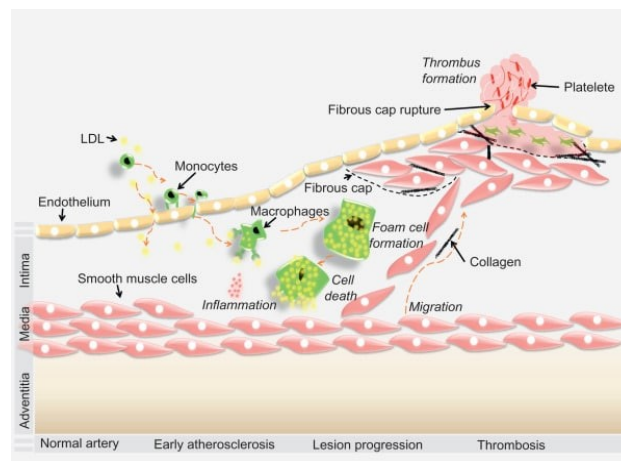


Figure 18. Atherogenesis

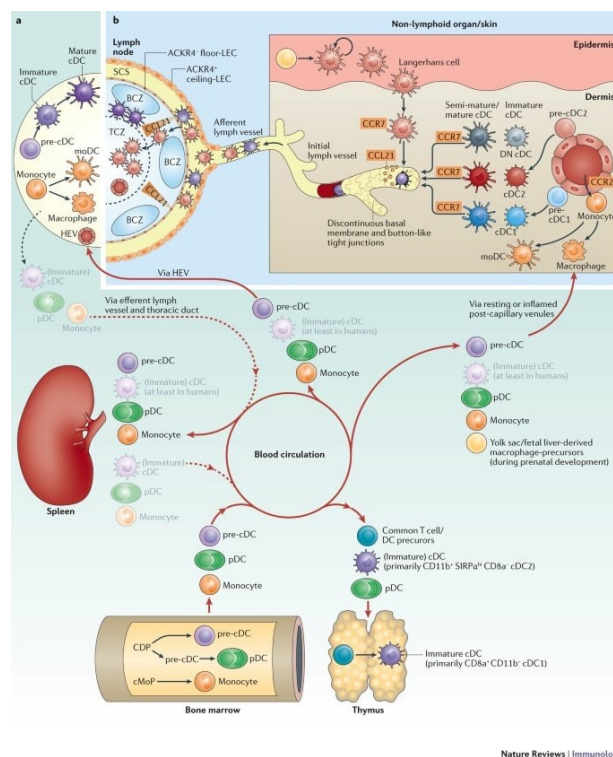


Figure 19. Adhesion and migration of dendritic cells

As a result, the interaction of activated endothelial cells and inflammatory cells is extremely complicated (**Figure 20**). Therefore, the study of the parietal effects of the endothelium is very important [13]. An increase the level of VFW in blood plasma is considered to be a marker of endothelial activation and / or damage.

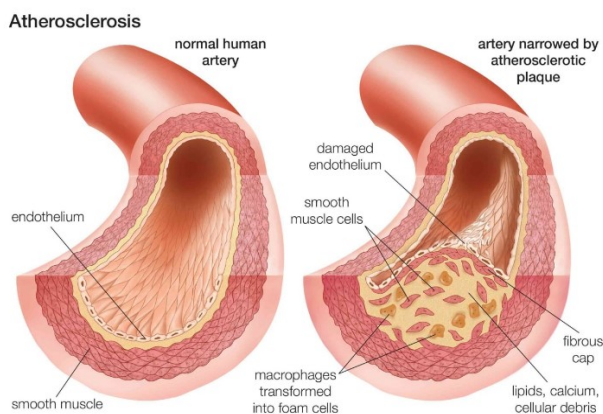


Figure 20. Atherosclerosis in the inflammatory process

By its nature, the von Willebrand factor is a glycoprotein (from 500,000 to 2 million daltons) (**Figure 21**). The FW subunits contain binding domains to the glycoprotein receptors Tr, collagen, heparin, coagulation factor V111 [14]. The main object of FW synthesis is endothelial cells. In this case, most of the synthesized FW is deposited in the granules of endothelial cells-cells of Weibel-Palade and, upon activation of cells is able to be rapidly released into the extracellular environment - into the blood.

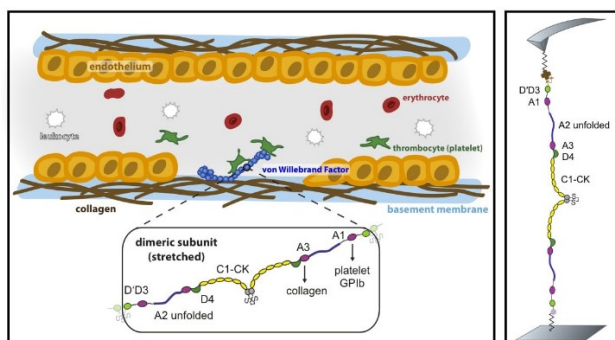


Figure 21. Von Willebrand factor

The role of FW in the circulation consists of the transport of the coagulation factor procoagulant V111, which ensures its stability and accumulation at the sites of vascular damage and thrombosis. However, the

most significant value of FW is its participation in vascular - TR interaction. At the stages of adhesion, flattening and aggregation of Tr, FW functions as a "bridge" between the subendothelial structures of the damaged vascular wall and Tr, as well as between the other ones. Currently, there are two approaches to the determination of FW: 1) determination of the level of FW antigen by using specific antibodies; 2) by ristocetin-induced aggregation of standard or formalized Tr.

For some diseases, the diagnostic value of FW as a marker of endothelial dysfunction has been proved. This was first shown by Boneu B. and co-authors (1975), who observed an increased concentration of FW in ischemic lesions of the extremities and sepsis. It was also shown an increase the level of FW in the blood plasma of patients with hypertension, ischemic stroke, pulmonary hypertension.

It was observed decreased plasma FW levels in patients after thrombolysis, who used tissue plasminogen activator.

In addition, there are indications on the shift in the functioning of other links of hemostasis in inflammatory diseases (prostaglandins, prostacyclin, thromboxane A2 (TxA2), etc. in the literature sources [15], [16]. It is known that cyclic endoperoxides, which are formed due to the activation of lipid peroxydation in inflammatory processes, act as a substrate for of TXA2 synthesis in Tr and prostacyclin in vascular endothelium. TxA2 is a

powerful activator of Tr aggregation and causes vascular spasm, whereas prostacyclin, in contrast, inhibits Tr aggregation and dilates blood vessels. Changes in the balance between TxA₂ and prostacyclin towards prostacyclin cause activation of the coagulation system and thrombosis. Particularly, it is found a tendency to decrease the level of plasminogen activator and increase the content of its inhibitor, as well as the antigen U111 PV and ristocetin aggregation Tr in patients with chronic glomerulonephritis. In patients with chronic nephritis, an association has been found between the increased Tr aggregation function and oxidative "stress" (increased levels of DC and MDA in the blood). Deterioration of local microcirculation was found in children with vasomotor rhinitis. The fibrinolytic system in patients with lupus nephritis is depleted first by the local renal vascular channel, and then by the systemic, depending on the severity of the disease. Studies of the functions of microcirculatory hemostasis in patients with bronchial asthma have shown an increase in the amount of Tr, their ability to respond to the action of aggregating agents, violation of the content of TxA₂.

In patients with steroid-dependent bronchial asthma and chronic obstructive pulmonary disease, the authors observed activation of total fibrinolytic activity of blood plasma until the development of disseminated intravascular coagulation syndrome. In patients

with inflammation of the skin, both allergic contact dermatitis and atopic damage, or due to skin irritation, the expression of CS-1 fibronectin was found in the endothelial cells of inflamed vessels. In patients with psoriasis, there is an increase in the activity of Tr hemostasis on the background of reduced fibrinolytic activity of the blood, and at the same time Tr factors are significantly involved in the implementation of psoriatic inflammation.

The authors identified a correlation relationship between endothelial activation and release of adhesion molecules, impaired hemostasis, immunity and indicators that characterize the development of atherosclerosis in cases of rheumatoid arthritis. In patients with appendicitis there are adverse disorders of the LP spectrum of Tr membranes, which increase their Tr potential. Obliterative endarteritis is accompanied by a significant increase in the adhesive-aggregation properties of Tr in the lesion with a high risk of developing a generalized form of hemostasis.

In hypertension Tr endothelial dysfunction occurs with the involvement of proinflammatory cytokines, which indicates the immune-inflammatory nature of endothelial damage in high blood pressure[18]. In case of insulin-dependent diabetes mellitus, an increase of spontaneous aggregation was found. It is believed that these processes depend on the deficiency of NO secretion by damaged endothelial cells.

In case of STI lesions, which can cause inflammation of the CO, disorders of the hemostasis system and signs of endothelial lesions are also observed.

The works of J.I. Arkhipova and co-authors showed that most hemoblastoses and aplastic anemias are complicated by immune compromise, the development of mycoses in combination with hemorrhagic syndrome. Bleeding is caused not only by Tr-penia due to aplasia of the bone marrow, but also by the tropism of the mycosis pathogen to blood vessels. Some fungi, including yeast and, especially, mold, are able to invade blood vessels with the development of necrotizing angiitis, which is manifested by necrosis, heart attacks, hemorrhages in various cavities, organs and tissues.

In case of secondary syphilis, indicators of spontaneous and inductive aggregation of Tr appear, as well as EF, which characterizes hemostasiological endothelial dysfunction [16]. Patients with endothelial inflammation have reduced fibrinolytic properties and develop resistance to thrombolysis, which plays an important role in the occurrence of heart attacks [17].

Thrombin, activated during the inflammatory response, acts as an important mediator of neutrophil-dependent damage, joining the multicomponent inflammatory cascade system as an important link between inflammation and thrombosis.

Results and discussion

State of platelet of hemostasis in patients with chronic inflammatory process of genitals.

For laboratory diagnosis of the activity of the inflammatory process and the prognosis of complications, the assessment of the functional state of platelets is of great importance.

Therefore, in patients with chronic inflammatory process of the genitals gene, we studied the indicators of adhesive-aggregation functions of platelet hemostasis (Figure 22).

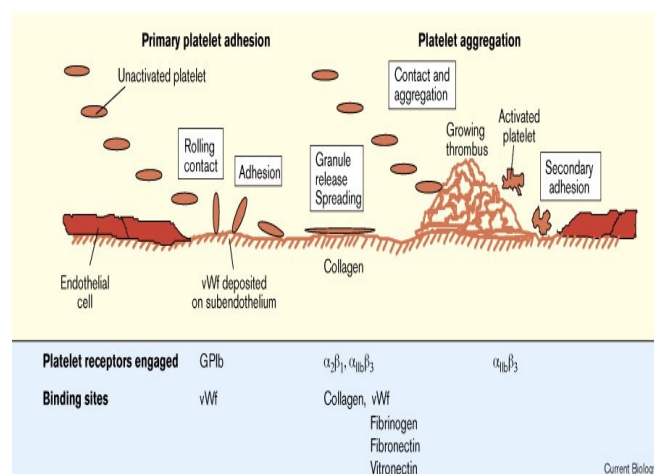


Figure 22. Platelet adhesion, activation and aggregation on exposed subendothelium, for example at a wound site.

The relative level of von Willebrand factor (VWF), the platelet component of which characterizes platelet aggregation, was also determined, and the vascular component is considered a marker of endothelial damage[19]. As a result of the study, an increase in the intensity of platelet adhesion (Table.1) was

found in men of the 2nd group by 1.2 times ($p < 0.05$) and in women of the 3rd group 1.4 times.

Table 1. Indicators of platelet hemostasis and von Willebrand factor in patients with chronic inflammatory process of the genitals.

| Group | | The degree of adhesion, % | ADP aggregation on start time, s | Degree of maximum aggregation, % | Relative level of VWF, % |
|-------|---|---------------------------|----------------------------------|----------------------------------|--------------------------|
| K 1 | M | 40,5±4,9 | 15,7±2,8 | 17,9±5,3 | 100 |
| | W | 38,9±4,1 | 14,9±3,6 | 20,2±4,9 | 100 |
| K 2 | M | 41,2±5,3 | 15,2±3,3 | 18,8±4,8 | 100 |
| | W | 40,0±3,8 | 14,3±5,3 | 21,1±7,2 | 100 |
| K 3 | M | 42,0±3,8 | 14,8±3,6 | 20,7±5,3 | 100 |
| | W | 40,7±4,7 | 14,0±2,8 | 23,4±5,5 | 100 |
| 1 | M | 42,8±2,3 | 8,38±3,3* | 36,6±6,4* | 44,3±39,1 |
| | W | 42,7±2,0 | 12,2±3,4 | 41,6±12,3* | 42,1±27,4 |
| 2 | M | 50,0±6,0* | 9,81±3,88* | 38,3±6,8* | 38,9±25,5 |
| | W | 40,4±2,3 | 10,2±2,3* | 36,4±6,6* | 34,8±22,0 |
| 3 | M | 39,6±2,3 | 12,5±5,3 | 39,4±7,9* | 61,8±46,1 |
| | W | 56,1±4,1* | 8,75±3,2* | 37,2±4,8* | 29,7±21,8 |

Note. * - $p < 0.05$; compared to control

The time of onset of platelet ADP aggregation was significantly reduced in men of the 1st and 2nd groups in 1.9 and 1.6 times

($p < 0.05$), respectively, and in women of the 2nd and 3rd groups in 1.5 ($p < 0.05$) and 1.6 ($p < 0.05$) times compared to K.

The intensity of maximal ADP platelet aggregation significantly exceeded that in all groups of patients.

Thus, in men of the 1st, 2nd and 3rd groups, this indicator was increased by 2.0 ($p < 0.05$) times.

In women of the 1st, 2nd and 3rd groups, the degree of maximal platelet ADP aggregation was increased by 2.0, 1.7 and 1.6 times ($p < 0.05$), respectively.

We observed that a certain level of VWF in all groups of people decreased and ranged from 30 to 62% of this control group (100%). Given the fact that the vascular component of VWF is a marker of the functional state of the vascular endothelium, we can assume that in patients with chronic inflammatory process of the genitals there was damage to endothelial cells, endothelial dysfunction with reduced VWF production.

When analyzing the function of platelet counts in groups of patients depending on the type of inflammatory pathogen (table 4.2) revealed a probable increase in platelet adhesion in all groups except the 5th with (viral infection) on average 1.4 times ($p < 0.05$); reduction of aggregation time by 1.3 times ($p < 0.05$); increase in maximum aggregation by 1.9-2.2 times ($p < 0.05$).

We watched as the relative level of VWF plasma levels probably decreased in all groups of patients with different sexual infections 2.4-3.2 times.

The decrease in this indicator in patients of group 4 with chlamydial infection was the most significant ($p < 0.05$), compared with K, 1-3 and 5-6 groups.

Table 2. Indicators of platelet (PLT) hemostasis and the content of von Willebrand factor in the plasma of patients with various types of genital infections

| Indicator unit of measurement | K | 1 | 2 | 3 | 4 | 5 | 6 |
|----------------------------------|----------|-----------|-----------|-----------|-----------|-----------|-----------|
| The degree of adhesion, % | 40,6±0,6 | 54,2±1,2* | 55±1,2* | 56,7±1,3* | 56,8±1,4* | 39±1,2 | 57,2±1,2 |
| ADP Aggregation start time, s | 14,9±0,4 | 9,7±0,3* | 9,1±0,2* | 9,2±0,3* | 9,4±0,3* | 11,3±0,4* | 10,4±0,4* |
| Degree of maximum aggregation, % | 19,9±1,1 | 40,1±1,6* | 38,3±1,3* | 38,6±1,3* | 43,4±1,2* | 37,6±1,3* | 40,5±1,3* |
| Relative level of VWF, % | 100 | 41,2±3,6* | 34,2±2,9* | 31,2±2,4* | 23,3±1,9* | 38,8±2,6* | 36,7±2,6* |

Notes:

- * - $p < 0.05$ compared with the control group
- ^ - $p < 0.05$, compared to all other groups

In general, disorders of adhesion-aggregation functions of platelets occurred in the direction of increasing thrombogenic potential, while the relative level of von Willebrand factor in patients with group 4 with (chlamydial infection) VWF decreased ($p < 0.05$).

This can lead to a disorder of regulation of the vascular-platelet hemostasis system, primarily in the direction of increase with a possible subsequent decrease in the activity of primary hemostasis due to depletion of von Willebrand factor and the consumption of circulating platelet pool.

Endothelial dysfunction and platelet dysfunction, on the one hand, leads to limited access of drugs to the lesion, and on the other hand, does not exclude the possibility of complications from the blood coagulation system.

Therefore, the question arises whether chronic inflammatory processes of the genitals can play a role in the development of platelet disorders of hemostasis.

To answer this question, we observed that the obtained data correlated with indicators that characterize inflammatory processes: local - in the source of inflammation, as well as in the circulating blood system.

It was also interesting to trace the associations of shifts in lipid metabolism and adhesion-aggregation function of platelets and the relative level of von Willebrand factor in the plasma of patients with chronic inflammatory diseases of the genitals.

We showed that the shift in the concentration of Low-density lipoprotein cholesterol (LDL-C) had strong feedback with the amount of PLT per unit volume of blood ($r = -0.73$) (Table 3), the time of onset of ADP

aggregation PLT ($r = -0,52$) and the relative level of VWF in blood plasma ($r = -0.52$), ($p < 0.05$).

The shift in the concentration of total cholesterol had strong feedback with the amount of PLT per unit volume of blood ($r = -0.61$), the time of onset of ADP aggregation PLT ($r = -0.74$), the relative level of VWF in blood plasma ($r = -0.86$), as well as a direct strong relationship with the intensity of maximum aggregation PLT ($r = +0.89$), ($p < 0.05$).

That is, an increase in the concentration of LDL-C and total cholesterol was associated with a decrease in the amount for VWF per unit volume of blood, acceleration and increase in the intensity of ADP aggregation of PLT and a decrease in the relative level of VWF.

At the same time, the value of the concentration of High-density lipoprotein cholesterol (HDL-C) had direct strong correlations with the number of PLT per unit volume of blood ($r = +0.73$), the time of onset of ADP aggregation PLT ($r = +0.51$), relative the level of VWF in plasma ($r = +0.64$), and strong feedback ($r = -0.63$) ($p < 0.05$) with the intensity of maximum ADP aggregation PLT.

This means that the increase in HDL-C was associated with an increase in PLT per unit volume of blood, probably due to a decrease in their consumption, a decrease in the rate and intensity of PLT aggregation, normalization of VWF levels in blood plasma.

Table 3. Correlation coefficients (r) of lipid metabolism and platelet count of hemostasis and the content of von Willebrand factor in blood plasma in patients with chronic inflammatory process of the genitals

| Indicator, unit of measurement | Number of PLT l^{-1} | ADP aggregation start time, s | Degree of maximum aggregation PLT, % | Relative level of VWF, % |
|------------------------------------|------------------------|-------------------------------|--------------------------------------|--------------------------|
| The level of LDL-C, $mmol l^{-1}$ | -0,73* | -0,52* | 0,48 | -0,52* |
| Conc. of cholesterol $mmol l^{-1}$ | -0,61* | -0,74* | 0,89* | -0,86* |
| Conc. of HDL-C $mmol l^{-1}$ | 0,73* | 0,51* | -0,63* | 0,64* |

Note. * - $p < 0.05$, compared with the control group.

In the surveyed individuals with the persistence of various agents of sexually transmitted infections, the probable strong feedback of the concentration of total cholesterol (CH) in the blood plasma and the time of onset of ADP aggregation ($r = -0.79$) and the relative content of VWF ($r = -0,96$) ($p < 0.05$), as well as strong direct connections with the maximum aggregation PLT ($r = +0.94$) ($p < 0.05$) (Table 4).

That is, the level of total cholesterol was associated with an acceleration and increase in ADP-aggregation of PLT and with a decrease in the relative level of VWF in blood plasma. Similar associations were observed with regard to the concentration of malonic dialdehyde (MDA) in blood plasma.

This indicator had strong feedback with the time of onset of ADP aggregation PLT ($r = -$

0.84) and the relative level of VWF ($r = -0.94$), as well as strong direct relationships with the intensity of ADP aggregation ($r = -0.97$).

Table 4. Correlation coefficients (r) of lipid metabolism and platelet count of hemostasis and the content of von Willebrand factor in blood plasma in subjects with various infectious agents of the genitals

| Indicator, unit of measurement | ADP aggregation start time, s | Degree of maximum aggregation PLT, % | Relative level of VWF, % |
|---|-------------------------------|--------------------------------------|--------------------------|
| Concentration of CH, mmol l ⁻¹ | -0,79* | 0,94* | -0,96* |
| Concentration of MDA mmol l ⁻¹ | -0,84* | 0,97* | 0,97* |

Note. * - $p < 0.05$, compared with the control group

Conclusions

Thus, the study found that increasing the level of proatherogenic substances in blood plasma (total cholesterol, low-density lipoprotein cholesterol, malonic dialdehyde) increases the aggregative activity of platelets and reduces the level of von Willebrand factor. At the same time, the increase in antiatherogenic substances high-density lipoprotein cholesterol (HDL-C) reduces platelet aggregation and increases the level of von Willebrand factor in blood plasma.

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