Specificities of changes of interleukins in the blood, bone marrow and liquor of critical patients

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In critical patients studying of immune status began in 1975 year. In this regard researches are presented in more than 200 works of Institute. Using research results 2 doctoral and 18 probations are maintained. Work results demonstrated that critical patients have immunodeficiency. These include both immune competent and progenitor cells.

During dyeing T lymphocytes loose ability of immune memory, there suppressor activity falls, apoptotic ability also experiences changes which in turn promotes auto aggressive mechanisms. Mechanism of immune status is related to cytokine profile which is generally not well studied. In this regard interleukin changes are studied in blood, bone marrow and CSF of critical patients.

Key words: Blood, Bone marrow, Interleukins.

Actuality:

The research of immune status in critical patients has been begun since 70s of the XX century. From this viewpoint our institute has a priority, where the research of above mentioned problem has been carried out since 1975 and still remainsthe main line of our researches.The researches about immune status are discussed in about 200 works of the Institute. The two DPhils and 18 Candidates' dissertation are made based on the results of the researches. The results of the researches have made clear that there is a certain presence of immunosuppression in critical patients, both in immunocompetent and progenitive cells. During the death the T lymphocytes lose the ability of immune memory, the suppressive activity is diminished, changes the ability of apoptosis that favors the development of autoagressive mechanism. The mechanism of immune status is mostly connected to the profile of cytokines, the study of which is less known and is almost unidentified in critical patients. The cytokines are the mediators of intercellular interaction and play the important role in regulation of the organism's reaction. Any inflammatory reaction in the organism is formed with the participation of cytokines. Its quantitative definition has a great importance in assessment of pathological process activity, in disease recognition and in its flow gravity as well as in

monitoring of the treatment efficiency and in prognosis of the disease outcome. The content of cytokines in serum and in plasma does not completely describe the immune system of an individual, but as an integral identifier, still describe the tension of herd immunity. The goal of our research was to study the cytokine profile in critical patients, their participation in death process and their eventual use in progress of the disease and in outcome prognosis. The following cytokines were studied in the blood plasma of patients: IL-4 IL-6, IL-8 TNF a and IL -10 TNF a- is a cytokine inducing pleotrophic inflammation, performing the regulatory and effector functions during the immune response and inflammation. The main producers of TNFa are monocytes and macrophages. Besides it is secreted from granulocytes and natural killers. The main inductors of TNF are the components of microorganisms, including bacterial lypo-polysaccharides. The inductors might be interleukin1, interleukin2, as well as alpha and beta interferons. TNF a has a characteristic of selective cytotoxicity towards some carcinogen cells; makes active granulocytes, macrophages, endothelial cells and hepatocytes (protein production of the acute phase); stimulates the differentiation of neutrophil fibroblasts as well as angiogenesis, haemopoiesis, has the anticancer and antiviral activity.The increased concentration of TNF a is identified in cases of acute bacterial, viral and parasitic inflammations, as well as during cancer diseases. The increased concentration of liquor is identified in cases of multiple sclerosis and meningitis. It takes part in autoimmune reactions. IL-4 is a glycoprotein, the main producer of which is TH-2 lymphocyte IL-4 enhances the eosinophilia, the IgG4 secretion and B-cell response; it stimulates an IgE synthesis has anticancer effect, suppresses the secretion of inflammatory cytokines and prostaglandins. The concentration of IL-4 in blood increases during allergic reactions and other inflammatory processes. In normal situations, in healthy donors, its concentration is not more than 6 pg/ml. IL-8 belongs to chemokines and is produced by mononuclear phagocytes, polymorphonuclear leukocytes and by different cytokines of endothelial cells (IL-1, TNF a), as well as during stimulation with bacteria, viruses and products of their metabolism. Actually, in healthy donors the quantity of IL-8 is not more than 30 pg/ml. Its increased indices are identified in cases of grave bacterial infections (sepsis, pulmonary chronic obstructive diseases). The determination of IL-8 might be used for control and prognosis of above mentioned diseases. IL-6 is the pleiotropic cytokine with wide range of biological activity. It is produced by lymphoid and some non-lymphoid tissues. IL-6 regulates immune response, acute phase response, inflammation carcinogenesis and haemopoiesis. One of the functions of IL-6 is to take part in production of antibodies produced by B-lymphocytes. The

increase of IL-6 level is detected in cases of various inflammations, in cases of gastrointestinal exacerbation. In healthy donors its concentration is not more than 30pg/ml. The imbalance of cytokines mainly inflammatory and anti-inflammatory cytokines plays an important role in allergic, autoimmune, carcinogenic, infectious and cardiovascular diseases.

Materials and methods:

48 patients were surveyed, who passed the course of treatment in the Institute of Critical Medicine, age range - from 35 up to 82 years.

Including diseases:

a) Ischemic stroke-14
b) Hemorrhagic stroke -14
c) Cardiac failure -12
d) Sepsis -8

In all patients multiple organ failure was detected with additional diseases such as: diabetes, atherosclerosis, pneumonia, chronic cardiac insufficiency. All the patients were treated with traditional method. All of them were on the artificial respiration and parenteral nutrition. The drug treatment was administered according to the symptoms. The patients were examined on the first, sixth and seventh day after their hospitalization. It should be mentioned that the examination of cytokines' concentration in blood is often used in practice, but in liquor and in spinal marrow the concentration of cytokines is not determined almost at all. The cytokines were determined by the method of immune-enzyme analysis. The results were calculated by immune-enzyme reader RAITO 2100. During the researches the immune-enzyme analysis test-systems Vector-Best manufactured by Russia were used:

Interleukin-4 - immune-enzyme analysis –VECTOR BEST

Interleukin -6 - immune-enzyme analysis – VECTOR BEST

Interleukin -8 - immune-enzyme analysis – VECTOR BEST

Interleukin -10 - immune-enzyme analysis – VECTOR BEST

TNF - immune-enzyme analysis – VECTOR BEST

Immune-enzyme analysis were conducted according to the instructions attached to the generally accepted method of "Sandwich ELISA".

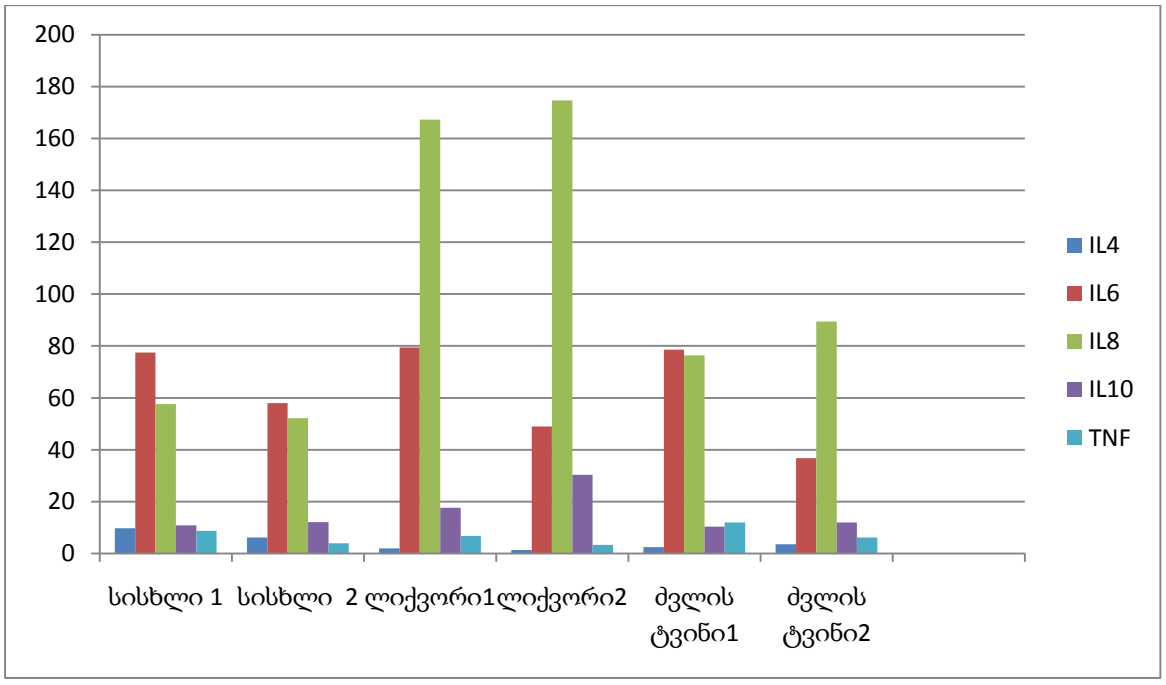
The results of the research are given in the table.

As it is seen from the table the profile of the cytokines are always changed in critical situations. At

material	groups	IL4	IL6 <30pg/ml	IL8 30pg/ml	IL10	TNF-α
blood	before treatment	9,8±2,2	77,5±4,0	57,6±3,5	10,8±0,6	8,7±1,6
	after treatment	6,2±2,9 >0,05	58,0±12,9 <0,05	52,1±9,9 >0,05	12,1±1,8 >0,05	4,0±1,0 <0,05
liquor	before treatment	2,0±0,2	79,4±5,2	167,3±9,9	17,6±6,4	6,8±1,2
	after treatment	1,4±0,4 >0,05	43,0±13,4 <0,05	174,6±34,1 >0,05	30,4±8,3 >0,05	3,3±0,9 <0,05
spinal marrow,	before treatment	2,5±0,2	75,6±4,4	76,4±15,3	10,4±1,0	12,0±1,6
	after treatment	3,6±1,0 >0,05	36,7±4,2 <0,001	89,4±12,2 >0,05	12,0±1,4 >0,05	6,2±1,6 <0,05

the same time the proinflammatory cytokines are increased especially before the treatment. In the process of treatment the concentration of cytokines (IL-6, IL8, TNF) in blood and in liquor in some cases is decreased, and the concentration of anti-inflammatory cytokines (IL4 and IL10) is increased.

The interleukin changes in blood, liquor and in spinal marrow of critical patients.



Blood 1 Blood 2 Liquor 1 Liquor 2 Spinal marrow 1 Spinal marrow 2

As it can be seen from the chart, before the treatment the level of interleukin-6 is almost the same in all the biological liquids and after the treatment its concentration is perceptibly decreased. The concentration of IL-8 is increased in all cases, but during the treatment in some cases it does not

change in some of them (in liquor) it is even increased. This might be explained by the different mechanism of IL-8 regulation, or by the time factor (in short time we had for researches, the changes were not detected). TNF changes were not detected in our case; the changes in anti-inflammatory cytokines, namely IL-10 changes were detected only in liquor after the treatment start. In cases of 48 patients 28 of them had a fatal outcome. The situation of 10 patients having perceptible increase of IL-10 in liquor and peripheral blood has been improved. The situation was improved in cases of those patients having the perceptible decrease of IL-6 after the treatment start. Thus, during our researches the perceptible increase of proinflammatory cytokines has been determined in critical patients. The increase of IL-10 after the treatment start is a hopeful forecast. The unchanged level of IL-4 and IL-8 is an indicator of situation aggravation and practically, of death. Our researches are the minimal part of explanation of death process mechanism. It is necessary to continue the researches in this direction that is being done at the basis of our institute. We hope that these researches will contribute to the issues of critical situation management.

Summary:

The goal of our researches was to study cytokine profile in critical patients, their participation in death process and their eventual use in use in progress of the disease and in outcome prognosis.

The following cytokines were studied in the blood plasma of patients: IL-4 IL-6, IL-8 TNF a and IL -10.

In all patients multiple organ failure was detected with additional diseases such as: diabetes, atherosclerosis, pneumonia, chronic cardiac insufficiency. All the patients were treated with traditional method. All of them were on the artificial respiration and parenteral nutrition.

Thus, during our researches the perceptible increase of proinflammatory cytokines has been determined in critical patients. The increase of IL-10 after the treatment start is a hopeful prognosis. The unchanged level of IL-4 and IL-8 is an indicator of situation aggravation and practically, of death.

ინტერლეიკინების ცვლილებათა თავისებურებანი კრიტიკულ ავამყოფთა სისხლში, ძვლის ტვინში და ლიქვორში.

ნ.ბარნაბიშვილი, ზ.ხელაძე, ზვ.ხელაძე, ნ.ქავჯაია, ე.ივანიძე, მ.ტყემალაძე, თ.მაზმიშვილი (თბილისი, საქართველო)

კრიტიკულ ავადმყოფებში იმუნური სტატუსის საკითხის შესწავლა 1975 წლიდან წარმოებს. ამ თვალსაზრისით კვლევები წარმოდგენილია ინსტიტუტის 200-მდე ნაშრომში. გამოკვლევების შედეგების გამოყენებით დაცულია 2 სადოქტორო და 18 საკანდიდატო დისერტაცია. შრომების შედეგებმა ცხადჰყო, რომ კრიტიკულ ავადმყოფებში ადგილი აქვს იმუნო დეფიციტს. ეს მოიცავს როგორც იმუნურკომპეტენტურ, ასევე პროგენიტურ უჯრედებს. კვდომის დროს T ლიმფოციტები კარგავენ იმუნური მეხსიერეების უნარს, უქვეითდებათ სუპრესორული აქტივობა, ცვლილებას განიცდის აპოპტოზის უნარიც, რაც თავის მხრივ აუტოაგრესიული მექანიზმების განვითარებას უწყობს ხელს. იმუნური სტატუსის მექანიზმი მნიშვნელოვნად არის დაკავშირებული ციტოკინების პროფილთან, რაც ზოგადად ნაკლებად არის შესწავლილი და თითქმის არ არის შესწავლილი კრიტიკულ ავადმყოფებში. ამასთან დაკავშირებით შესწავლილია ინტერლეიკინების ცვლილებები კრიტიკულ ავადმყოფთა სისხლში, ძვლის ტვინსა და ლიქვორში.

გასაღები სიტყვები: სისხლი, ძვლის ტვინი, ინტერლეიკინები.