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Role and Correlation of Laboratory Biomarkers in the Assessment of Severe COVID-19 in Children

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ABSTRACT

At the end of 2019, the coronavirus infection quickly spread around the world and WHO declared the beginning of a pandemic on March 11, 2020. To this end, the authors decided to study the role of some laboratory biomarkers in assessing the severity of the course of the disease Covid-19 in children and the correlation between them. 551 patients applied to the Department of Pediatric Infectious Diseases. 55 patients without comorbidity, with confirmed "Upper Respiratory Diseases" were treated on an outpatient basis and entered the control group. 496 patients were diagnosed with Covid-19 and were included in the study group. Of the patients, 364 children were laboratory positive for C-19 and belonged to group I (covid pediatric). Laboratory C-19 in 132 children was assigned to the negative-II group (MIS-Ch). Patients were diagnosed with a number of biomarkers (inflammatory, respiratory and metabolic panel, coagulogram, interleukin-6, brain natriuretic peptide concentration) in the blood and the correlation between them. When comparing patients of groups I and II, it was found that the concentration of biomarkers was higher in patients of group II. This once again proves that the Covid-19 disease is more severe in patients of group II. In both groups, the correlation coefficient between biomarkers was also studied. Thus, there is a direct strong correlation between creatine kinase-MB and ferritin in group I and BNP and interleukin-6 in group II, and it is statistically significant.

Introduction

Coronavirus 2019 (Covid-19) is characterized by high infection and mortality, which sometimes occurs asymptomatically in children [1]. For this reason, the World Health Organization announced a coronavirus pandemic [2]. With severe and critical coronavirus disease-19, the concentration of different biomarkers is increased, as a result of which it is possible to assess and predict the severity of pneumonia [3]. The condition of sick Covid-19 can suddenly worsen and develop life-threatening complications [4]. Diagnosis of biomarkers in patients with Covid-19 is of great importance [5]. The course of COVID-19 is associated with increased blood clotting, left ventricular failure, myocardial infarction and the development of thrombohemorrhagic syndrome [6]. The results of studies in the blood increase the concentration of a number of enzymes that predict death: procalcitonin, C-reactive protein, D-dimer, ferritin, creatine kinase-MV, cerebral natriuretic [7]. Expert of the International Society of Thrombosis and

Hemostasis (ISTH) D. Wolff believes that if levels of D-dimer and ferritin COVID-19 are 3-4 times higher than normal in adults with concomitant and concomitant diseases (MIS-A), these patients have a key indicator of recurrence, which is why to the risk group [8]. A series of studies have shown that patients with N-end progesterone sodium peptide (NT-proBNP) Covid-19 have a higher risk of death. BNP represents its own peptide, secreted in response to elevated tension in the wall of the ventricle, and is considered one of the indicators of depressed systolic function of the left ventricle and unpredictable sudden [9]. The purpose of the study was to study the role of some laboratory biomarkers in assessing the severity of the disease Covid-19 in children and the correlation between them. The co-authors were allowed to write articles based on case histories and laboratory tests while maintaining the anonymity of the patients. We are aware of the decision of the Ministry of Health of the Republic of Azerbaijan on the Rules for the Ethical Conduct of Doctors by order No. 137 of 12/29/2011.

Materials and Methods of Research

In 2020-2021. 551 children applied to the Department of Children's Infectious Diseases. In 55 (9.98%) patients, there were no concomitant diseases and "upper respiratory tract disease" was confirmed, patients were treated on an outpatient basis and entered the control group. 496 patients (90.01%) were diagnosed with Covid-19 disease and included in the main group. Of the patients, 364 children (73.39%) were positive for the C-19 laboratory (PSR) and belonged to group I (covid children). Laboratory S-19 (RPS) in 132 children (26.62%) was assigned to the negative-II group (MIS-S). Of the patients of the main group, 297 (59.88%) were boys and 199 (40.12%) were girls. The patients were aged 2 to 14 (7.7 ± 0.4) years, 90–130 cm (1.15 ± 0.01) tall and weighed 10.5–65.0 kg (29.0 ± 9, 86). Children aged 2-6 years - 188 (37.9%), 7-11 years old - 108 (21.78%), 12-14 years old - 200 (40.33%). In the anamnesis of the main group, 213 (42.94%) of 496 patients had concomitant diseases (Table 1). Of the 496 patients treated in the hospital, 56 (11.29%) were in critical condition, 38 of them had sepsis (67.86%); 18 (32.14%) developed multiple organ (pulmonary-cardio-renal) insufficiency. Eleven of these patients were intubated on a Maguet ventilator with SIMV + PS + PEEP 4-5%; FiO2-45% is added. Of the 11 intubated patients, 7 died. Of the patients with a fatal outcome, 2 (28.57%) belong to group I and 5 (71.43%) belong to group II. The diagnosis was established on the basis of generally accepted clinical and anamnestic, laboratory, functional examinations, general and biochemical blood tests, inflammation, respiration, metabolic panel, interleukin-6, detailed coagulogram, brain natriuretic peptide (BNP).

Table 1: Co-morbidities in the study group.

Name of Diseases	n	%
Vitamin D deficiency	112	22,58
Superficial gastritis	8	1,61
Urinary tract infection	14	2,82
Diabetes	13	2,62
Anemia	29	5,85
Hypospody	9	1,81
Aneurysm in the coronary artery	3	0,61
Blepharoconjunctivitis	25	5,04
Total	213	42,94

Statistical processing of the obtained numerical data: for quantitative indicators, it was carried out in an EXCEL-2010 spreadsheet using the U-Wilcoxon (Mann-Whitney) rank test, the average values and average statistical errors of the obtained parameters were calculated, the results were summarized in tables and diagrams. Results and its discussion. In the study group of patients, the study was conducted in 2 stages: Stage I - the period of admission to the RITS; Phase II covers the recovery period (7-10 days of illness). When studying the results of our study, significant differences in the concentration of biomarkers were found, which is reflected in Figure 1. Patients with Covid-19 may develop cardiovascular complications such as heart failure, myocarditis, pericarditis, vasculitis, and cardiac arrhythmias [6]. If you look at the concentration of D-dimer, a marker of thrombosis and fibrinolysis, then in the control group it was 0.5 ng/ml. In group I, it was high at the first stage and was 3.6 times ($p0<0.05$) more than in the control group, but at the second stage it was 3.6 times ($p1<0.05$) lower than at the first stage and was at the level of the control group. In group II, the concentration of D-dimer at the first stage was 6.8 times ($p0<0.001$) compared with the control group and 1.9 times ($p2<0.01$) more than in the first group. At the second stage, this indicator sharply decreased by 4.05 times ($p1<0.001$) compared with the first stage but remained higher than in the control and group I, and 1.68 times ($p0<0.01$) ($p2<0.01$). Creatine kinase-MB (CK-MB), a biomarker of myocardial injury, is a meta-analysis that may be useful in assessing the risk of death in patients with COVID-19 [10].

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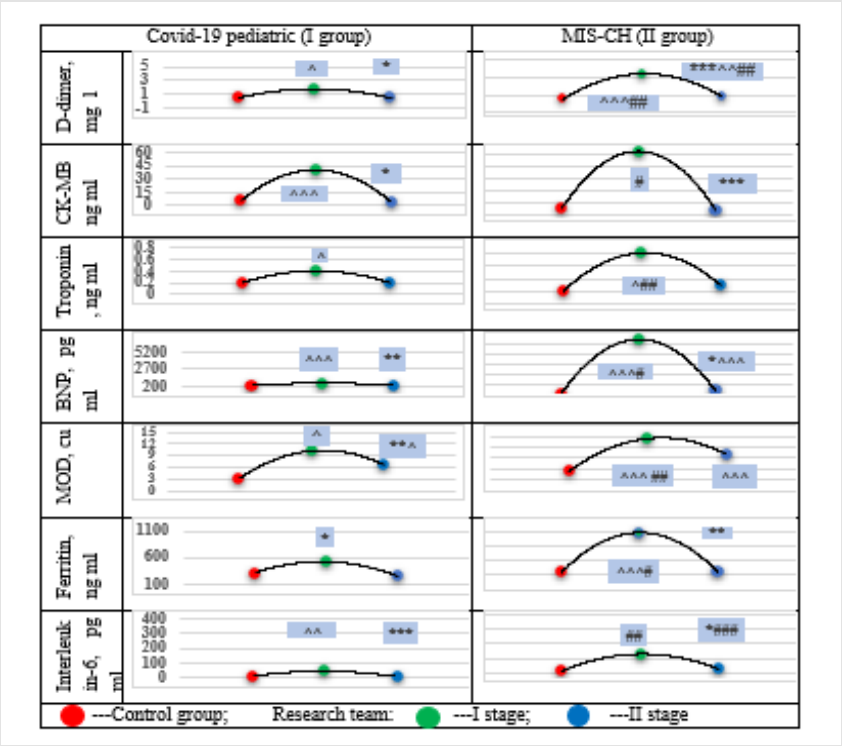

Figure 1: Dynamic changes in a number of biomarkers in comparison and research group patients.

Note: Statistical accuracy of the difference between the indicators at different stages in patients: ^ -p0<0.05 compared to the control group; ^^ - p0<0.01; ^^^ - p0<0.001; Compared to the first stage: * -p1<0.05; ** - p1<0.01; *** - p1<0.001; Compared to group I: # -p2<0.05; ## - p2<0.01; ### - p2<0,001.

CK-MB in the control group was 4.9±0.011 ng/ml. In group I, at the first stage, this indicator was 8.2 times (p0<0.001) higher than in the control group. At the second stage, this indicator decreased by 9.57 times (p1<0.05) compared to the first stage and reached the level of the control group. In group II, the concentration of CK-MB at the first stage was 10.89 times higher than in the control group, and 1.32 times higher than in the first group (p2<0.05). At the second stage, the control was at the group level, decreasing by 14.79 times (p1<0.001) compared with the first stage, and is statistically significant. Troponin secretion occurs early in the disease, reflecting the onset of myocardial infarction in 8-28% of patients with Covid-19 [11]. Therefore, it is important to study troponin and BNP concentrations from cardiac markers and myocardial oxygen demand (MOT) in children diagnosed with Covid-19. In children in the control group, troponin was 0.2±0.001 ng/ml. In group I, at the first stage, it was 2 times (p0<0.05) more than in the control group. At the second stage, the dynamics decreased and the control was at the group level. In group II, at the first stage, it was 4 times more than in the control group (p0<0.05), and 2 times more than in the first group (p2<0.01). Against the background of treatment, the dynamics of the control group decreased at the second stage.

The BNP concentration in the control group was 300.1±26.4 pg/ml. In group I at the first stage it was 2.27 times (p0<0.001) more than in the control group. Against the background of the measures taken at the second stage, this indicator was 1.06 times higher than in the control group but decreased by 2.12 times (p1<0.01) compared with the first stage and is statistically significant. In group II, at the first stage, it was 19.3 times (p0<0.001) compared with the control group and 8.5 times (p2<0.05) more than in group I. At the second stage, BNP decreased by 10.8 times (p1<0.05) compared with the first stage but was 19.3 times (p0<0.001) compared with the control group and 1.67 times more than in the first stage the first group. Hypotension, defined by septic status and blood hypoxemia as a result of Covid-19, can reduce the oxygen supply to the heart and thus lead to acute myocardial injury, especially in patients with primary chronic coronary syndrome. In this case, myocardial oxygen demand (MOD) may increase [9]. For this purpose, MOD was studied in children. MOD in the control group was 3.2 ± 0.21 shsh. In group I at the first stage it was 3.2 times (p0<0.05) more than in the control group. At the second stage, this indicator decreased by 1.52 times (p1<0.01) compared with the first stage but remained 2.1 times (p0<0.05) more than in the control group.

3.85 times in group II at the first stage compared with the control group (p0<0.001); Compared to group I, it remained 1.21 times (p2<0.01) more. Although at the second stage it decreased by 1.53 times compared with the first stage, it was 2.51 times (p0<0.001) more than in the control group.

The severity of ARDS in Covid disease is predicted based on blood ferritin levels. Thus, pro-inflammatory cytokines involved in ARDS increase ferritin synthesis and can accelerate the formation of toxic hydroxyl radicals [10]. The concentration of ferritin in the blood of the control group was within the normal range of 308.3±38.54 ng/ml. In group I at the first stage it was 1.68 times (p0<0.05) more than in the control group. At the second stage, the dynamics was 2.17 times lower than at the first stage (p1<0.01), 1.28 times lower than in the control group, but below the norm. In group II, at the first stage, it was 3.58 times (p0<0.001) compared with the control group and 2.12 (p2<0.05) more than in group I. Although at the second stage it decreased by 2.74 times (p1<0.01) compared to the first stage, it was 1.31 times less than in the control group; although 1.67 times more than in the first group, it was above the norm. It is important to evaluate cytokine production in COVID-19, especially the study of interleukin 6 as a pre-inflammatory cytokine that enhances the immune response [12]. Year-6 in the control group was 10 pg/ml. In group I, IL-6 was higher at the first stage, 4.28 times (p0<0.01) more than in the control group. At the second stage, this indicator decreased in dynamics by 7.51 times (p1<0.001) to the level of the control group. In group II, it was very high at the first stage, 12.95 times more than in the control group, 3.03 times more than in the first group (p2<0.01). Although the control in phase II was 2.8 times higher than in group I and decreased by 4.63 times (p1<0.05) compared with stage I, it was 4.91 times (p2<0.001) more than in group I and was statistically accurate.

The correlation between biomarkers in both groups was studied, the correlation between biomarkers in group I is shown in (Table 2). In group I, weakly flat between BNP and D-dimer (r = 0.19; p<0.01), weakly opposite with troponin (r = -0.32; p<0.01), weakly flat with MOC (r = 0, 38; p<0.01), weak reaction with ferritin (r = -0.11; p<0.05), weak reaction with IL-6 (r = -0.34; p<0.01); d-dimer weakly flat with troponin (r = 0.3; p<0.001), slightly opposite with ferritin (r = -0.2; p<0.001), slightly opposite with IL-6 (r = -0.1; p <0.001); weak contrast with troponin (r=-0.34; p<0.001), weak contrast with IL-6 (r=0.31; p<0.001); Weak reversion of CK-MB by IL-6 (r = -0.23; p<0.001); MOD weakly flat with ferritin (r = 0.17; p<0.001), slightly opposite with IL-6 (r = -0.25; p<0.001); ferritin has a moderately opposite (r=-0.52; p<0.01) correlation with IL-6 (Table 3). Of the biomarkers, only CK-MB had a strong direct correlation between ferritin, the correlation coefficient was r = 0.85 (p<0.001) and was statistically accurate, as shown in Figure 2. In group II, weakly direct (r = 0.29; p<0.001) between BNP and D-dimer, slightly opposite with troponin (r = -0.22; p<0.001), slightly opposite with CK-MB (r = -0 , 2), slightly flat with MOD (r = 0.14; p<0.001), slightly opposite with ferritin (r = -0.21; p<0.01); D-dimer moderately direct with troponin (r = 0.69), slightly opposite with CK-MB (r = -0.41; p<0.001), moderately direct with IL-6 (r = 0.79; p<0.01); Troponin weakly positive in CK-MB (r = 0.25; p<0.001), slightly opposite in MOD (r = -0.5), strongly direct in ferritin (r = 0.72), weakly opposite in IL-6 (r = 0.13; p<0.01); CK-MB is slightly opposite to MOD (r = -0.16), direct strong with ferritin (r = 0.7), slightly opposite to IL-6 (r = -0.48; p<0.05); MOD has a weakly opposite (r = -0.34) correlation with ferritin and a weakly direct (r = 0.34; p<0.05) correlation with IL-6.With D-dimer of ferritin and IL-6; ferritin also did not correlate with IL-6. Only a strong direct (r = 1.0; p<0.001) correlation between BNP and IL-6 is available, which is statistically significant Figure 3.

Table 2: Correlation between laboratory biomarkers in group I patients.

I group	D-dimer	Troponin	CK-MB	MOC	Ferritin	IL-6
BNP	↑**	↓**	↓	↑**	↓*	↓**
D-dimer		↑***	↓	↓	↓***	↓***
Troponin			↓	↓	↓***	↓***
CK-MB				↑	↑↑↑***	↓***
MOC					↑***	↓***
Ferritin						↓↓**

Note: Statistical accuracy of the difference between laboratory biomarkers in patients, compared: * -p<0.05; ** - p<0.01; *** - p<0.001. ↑- straight weak (r = 0-0.5); ↑↑ -Straight average (r = 0.5-0.7); ↑↑↑ -Straight (r = 0.7-1.0); ↓-reverse weak (r = 0 - (- 0.5)); ↓↓-reverse mean (r = -0.5 - (- 0.7)); ↓↓↓ -reverse is strong (r = -0.7 - (- 1.0)).

Table 3: Correlation between laboratory biomarkers in group II patients.

II group	D-dimer	Troponin	CK-MB	MOC	Ferritin	IL-6
BNP	↑***	↓***	↓	↑***	↓**	↑↑↑***
D-dimer		↑***	↓***	-	-	↑↑**
Troponin			↑***	↓↓	↑↑↑	↓**
CK-MB				↓	↑↑↑	↓*
MOC					↓	↑*
Ferritin						-

Note: Statistical accuracy of the difference between laboratory biomarkers in patients, compared: * -p<0.05; ** - p<0.01; *** - p<0.001. ↑- straight weak (r = 0-0.5); ↑↑ -Straight average (r = 0.5-0.7); ↑↑↑ -Straight (r = 0.7-1.0); ↓-reverse weak (r = 0 - (- 0.5)); ↓↓-reverse mean (r = -0.5 - (- 0.7)); ↓↓↓ -reverse is strong (r = -0.7 - (- 1.0)).

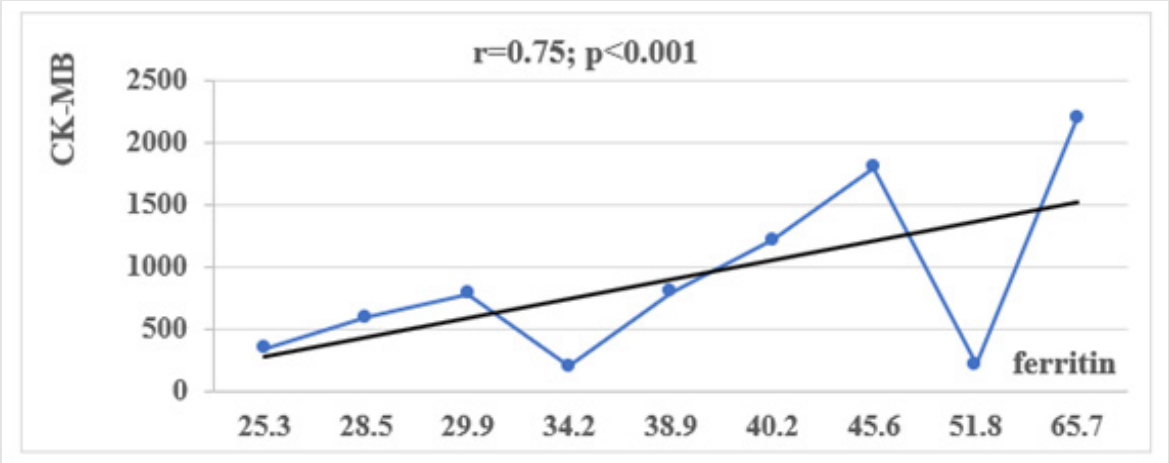

Figure 2: Correlation of CK-MB and ferritin in group I patients.

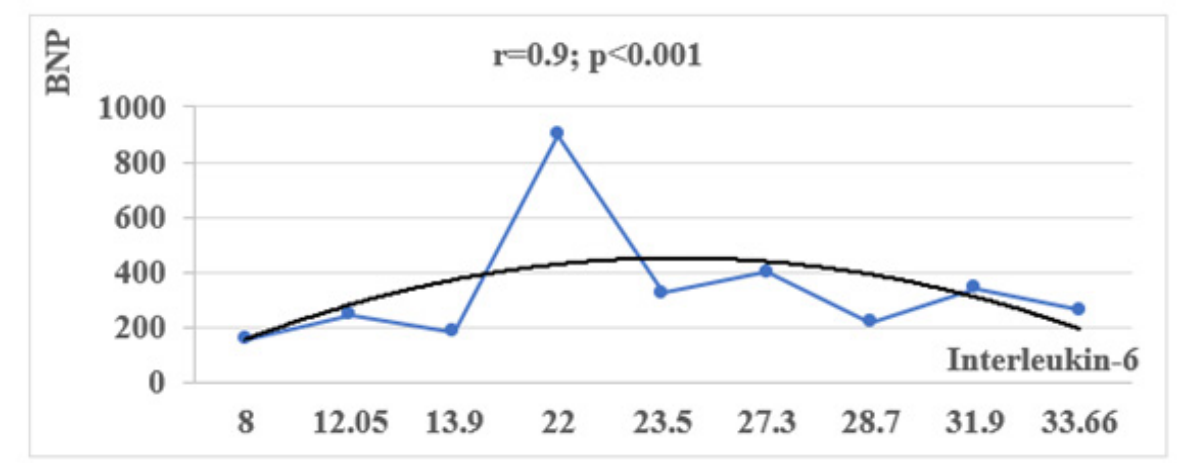

Figure 3: Correlation of brainatriuretic peptide and interleukin-6 in group II patients.

Result

a) D-dimer was higher in group I at the first stage, 3.6 times (p0<0.05) more than in the control group, and at the second stage it decreased 3.6 times (p1<0.05) and reached the level of the control group. In group II, 6.8 times (p0<0.001) from the control group at the first stage. Although at the second stage it decreased by 4.05 times (p1<0.001), in the control (p0<0.01) and 1.68 times (p2<0.01) it was higher than in the first group, and statistically significant.

b) Creatine kinase-MB was 8.2 times (p0<0.001) higher in group I than in the control group at the first stage. At the second stage, it decreased by 9.57 times (p1<0.05). In group II at the first stage it was 10.89 times more than in the control group, and 1.32 times more than in group I (p2<0.05). At the second stage, it decreased by 14.79 times (p1<0.001) and was at the level of the control group and was statistically significant.

c) Troponin in group I at stage I was twice as high as in the control group (p0<0.05). At the second stage, the dynamics decreased and the control was at the group level. In group II, at the first stage, the control was 4 times more than in the group (p0<0.05), 2 times more than in the first group (p2<0.01). At the second stage, the dynamics decreased and reached the level of the control group.

d) Brain natriuretic peptide in group I was 2.27 times (p0<0.001) more than in the control group in stage I. At the second stage, it decreased by 2.12 times (p1<0.01). In group II at the first stage it was 19.3 times more than in the control group (p0<0.001), 8.5 times more than in group I (p2<0.05). At the second stage, it decreased by 10.8 times (p1<0.05), remained 1.67 times more than in the first group.

e) MOD in group I was 3.2 times (p0<0.05) more than in the control group at the first stage. At the second stage, it decreased by 1.52 times (p1<0.01). In group II, 3.85 times (p0<0.001) from the control group at the first stage; 1.21 times more than in group I (p2<0.01). Although at the second stage it decreased by 1.53 times, it was 2.51 times (p0<0.001) more than in the control group and is statistically significant.

f) Ferritin in group I was 1.68 times (p0<0.05) more than in the control group at the first stage. At the second stage, it decreased by 2.17 times (p1<0.01) and was below the norm. In group II, at the first stage, the control was 3.58 times (p0<0.001) and 2.12 times (p2<0.05) more than in group I. At the second stage, it decreased by 2.74 times (p1<0.01), was above the norm and was statistically significant.

g) In patients of group I, there is a strong strong (p<0.001) correlation between creatine kinase-MB and ferritin, and in group II between BNP and interleukin-6 and is statistically significant.

Discussion

Kernan K.F. and co-authors conducted a systematic study and meta-analysis to evaluate biomarkers of anemia and iron metabolism (hemoglobin, ferritin, transferrin, soluble transferrin receptor, hepcidin, haptoglobin, unsaturated iron binding capacity, erythropoietin, erythrocyte free protoporphyrin) in patients with Covid-19. Although hemoglobin levels were low in elderly and comorbid patients in intensive care, ferritin levels were found to be high [10]. According to F. Heidari-Benya and co-authors, a high level of procalcitonin in the blood is not the main indicator of the severity of Covid-19 disease. A high level of procalcitonin indicates the presence of bacterial pneumonia, and not viral, and the development of a septic state [13]. P. Mehta and M. Brown achieved good results in the treatment of patients with the use of high doses of immunoglobulins to limit the action of antibodies by determining the severity of the disease based on the concentration of interleukin-6 in the blood of patients with Covid-19. Anakinra, which has immunosuppressive activity by blocking interleukin-1 receptors, has also been shown to be effective in patients with MIS-CH [14]. A number of researchers consider the use of metipred in the early stages to block secondary inflammatory processes in the treatment of children with multisystem inflammatory syndrome by strictly controlling the concentration of CRP and leukocytes in the blood [15]. Also W.J. Guan in his studies showed that the concentration of IL-6 in the blood can also increase under the influence of inflammatory parameters [16]. Thus, comparing our results with studies conducted by world scientists, it can be seen that the concentration of all biomarkers in the blood increases dramatically with covid-19 and MIS-CH. Based on the concentration of these markers, certain judgments can be made about the severity of the disease, the principles of treatment, and the level of mortality, and we have proved the existence of a correlation between biomarkers.

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