Immunological aspects of chronic kidney disease diagnosed in adult patients

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Abstract: The purpose of the study was to study the immune status of patients diagnosed with SBK, to develop immunological diagnostic and prognostic criteria. It was found that SD3+- and SD4+-lymphocytes decreased by 1.43 and 1.07 times, SD8+-lymphocytes increased by 1.25 times, and IRI decreased by 1.32 times (T-immunodeficiency). SD25+-lymphocytes increased by 1.80 and 1.59 times, and SD95+-lymphocytes increased by 1.28 and 1.13 times in the blood of these patients. It was found that the amount of SD16+-lymphocytes was reliably increased compared to the parameters of healthy individuals. An imbalance was observed in the concentration of immunoglobulins in blood serum, while IL-4 increased by 11.84 times in patients, IL-6 was reliably reduced by 1.46 times compared to healthy people. Dynamic determination of serum concentrations of IL-4 and IL-6 was recommended as diagnostic and prognostic criteria for patients diagnosed with SBK.

1 Introduction.
The development of clinical immunology in various pathological conditions, as well as in patients diagnosed with chronic kidney disease (CKD), provides information about the state of the immune system, changes in the quantitative and qualitative indicators of immunocompetent cells, various endogenous and exogenous factors that lead to this [5, 6, 8, 11].

The recommendation of diagnostic and prognostic criteria based on the laws related to changes in immunological indicators recommended by many researchers allows to determine the prospects of the end of various pathologies [4, 7].

SBK takes one of the leading places among many other diseases with its complications and death rates. Various clinical, instrumental, and laboratory methods are used to determine the prognosis of the end of the disease in these patients [1, 2, 12].

There are few works related to the comparative study of specific and non-specific resistance factors of the immune system in SBK, and those that exist are scattered, more emphasis is placed on the clinical aspects of the issue, there are no prognostic immunological criteria that determine the transition of the disease to the stage of hemodialysis in clinical practice [9, 10]. For this reason, studies focused on the solution of this issue have not lost their relevance.

The purpose of the study The study of the immune status of patients diagnosed with SBK consisted of developing immunological diagnostic and prognostic criteria.

2 Materials and Methods

135 SBK patients between the ages of 19 and 60 were included in the research. 87 of them (64.44±4.12%) were men, and 48 (35.56±4.12%) were women. It was found that men were diagnosed 1.81 times more than women (R<0.001). The majority of patients (71.85±3.87%, n=97) were aged between 21 and 50 years.

The inclusion of people in medical research was carried out in accordance with the Declaration of Helsinki adopted by the General Assembly of the World Medical Association (I). The principles of evidence-based medicine were followed in the organization and conduct of research.

Cellular (CD3+, CD4+, CD8+, CD16+, CD20+, CD25+, CD95+-lymphocytes) and humoral immunity (IgA, IgM, IgG), cytokine status (IL-4, IL-6) were determined in them. All immunological studies were performed using conventional immunological methods. All immunological studies were performed at the Institute of Immunology and Human Genomics of the Federal Republic of Uzbekistan and the Bukhara State Medical Institute.

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The statistical analysis of the obtained results was carried out on personal computers based on programs specially used for medical-biological research using traditional variational statistical methods.

3 Results and Discussion

Determining the immune status of adult patients with SBK began with the interpretation and analysis of cellular immune indicators. The obtained results showed that the amount of leukocytes in the blood of the patients was 5649±180x10⁹ /l, while the total amount of lymphocytes in the blood was on average 30.80±0.93%, and their absolute amount reached 1740±101 μl.

Although there was a reliable difference in the number of leukocytes in the blood, no reliable difference was found in the relative number of lymphocytes. The results showed that in healthy individuals (control group, n=20) SD3+-lymphocytes were 62.35±0.89%, while in the studied patients they were on average 43.72±0.56%, which is 1.43 times less reliable (R<0.05). A convincing decrease in the relative amount of SD3+-lymphocytes in patients was explained by the pathological condition developed in them.

Determination of SD4+-lymphocytes in the blood of patients diagnosed with SBK showed that, although this indicator was a small percentage in patients, it was convincingly lower than the parameters of healthy individuals - on average 30.43±0.57% vs. 32.45±0.66%, respectively. (1.07-fold decrease, R<0.05). If we take into account that SD4+-lymphocytes are assistants in the three-cell cooperative system that transmit antigen information from macrophages to antibody-producing V-lymphocytes, we can be sure that a decrease in their percentage will have a negative effect on the immune system.

When the synthesis of anti-antigen antibodies is sufficient during the performance of the immune system, when the antigens are eliminated from the body, SD8+-lymphocytes, the cells suppressing the activity of the immune system, their increase in number is a sign of the development of the pathological process. In the study, it was observed that the relative amount of SD8+-lymphocytes increased in patients compared to these parameters in healthy individuals. If in patients this indicator was on average 28.30±0.53%, then in healthy people it was recorded at an average level of 22.71±0.72% (1.25-fold reliable difference, R<0.05). It can be seen that the absolute numbers had a trend of changes in the same way as the relative parameters of the immune system. It is known that V-lymphocytes are immunity, has also been studied. In it, the relative amount of V-lymphocytes was determined and compared with the indicators of healthy people. It is known that V-lymphocytes are suitable for this antigen and provide an immune response. In our case, SD20+ marker-bearing V-lymphocytes were 62.35±0.89%, while in the studied patients they were on average 43.72±0.56%, which is 1.43 times lower (R<0.001).

Along with the T-joint of the immune system, the V-joint, which is responsible for humoral immunity, has also been studied. In it, the relative amount of V-lymphocytes was determined and compared with the indicators of healthy people. It is known that V-lymphocytes are suitable for this antigen and provide an immune response. In our case, SD20+ marker-bearing V-lymphocytes were 62.35±0.89%, while in the studied patients they were on average 43.72±0.56%, which is 1.43 times lower (R<0.001).

The increase of this cell in the peripheral blood of patients indicates that the activity of the immune system has decreased SD8+-lymphocytes increased reliably in patients, which led to decreased immune system activity in patients and decreased SD3+- and SD4+-lymphocytes. This condition is a target of secondary immunodeficiency developed in patients.

The parameters of the immunoregulatory index (IRI) were given as proof of the mentioned points. It is known that IRI is the ratio of SD4+-lymphocytes to SD8+-lymphocytes, and this index indicates the level of immune system activity. In our case, IRI was 1.08±0.02 units in patients diagnosed with SBK, which was 1.32 times lower than the parameters of healthy individuals (1.43±0.02 units) (R<0.05). This parameter was explained by a decrease in SD8+-lymphocytes due to an increase in the relative amount of SD8+-lymphocytes.

The fact that such changes occurred against the background of no significant changes in the total number of lymphocytes showed that only changes related to immunocompetent cells occurred, which was expressed in the form of T-immunodeficiency. Therefore, secondary immunodeficiency was observed in the immune system of patients diagnosed with SBK, which was expressed by the formation of T-immunodeficiency. All data are fully represented in Figure 1.

![Fig 1](https://example.com/fig1.png)

**Fig 1.** Comparative parameters of relative indicators of the immune system T-joint in adult patients with chronic kidney disease, % (LUS-lymphocyte total number).

Figure 1 shows the degree of development of T-immunodeficiency observed in patients.
Along with the T-joint of the immune system, the V-joint, which is responsible for humoral immunity, has also been studied. In it, the relative amount of V-lymphocytes was determined and compared with the indicators of healthy people. It is known that V-lymphocytes are differentiated from lymphopoietic stem cells in the bone marrow, and after T-helpers convey information about the antigen, they are synthesized into plasma cells, synthesize antibodies suitable for this antigen and provide an immune response. In our case, SD20+ marker-bearing V-lymphocytes (SD20+-lymphocytes) were 28.55±0.43% in patients and 23.98±0.93% in the control group, the difference between the numbers reached 1.19 times, and patients was statistically significantly higher in favor (R <0.05). The relative parameters of the V-joint of the immune system are compared with those of the T-joint in Fig. 2 for ease of evaluation.

Fig 2. Comparative parameters of relative indicators of the V-joint of the immune system in adult patients with chronic kidney disease, %.

As shown in Figure 2, an imbalance was observed in the T- and V-joints of the immune system, that is, when there was a deficiency in the T-joint (T-immunodeficiency), an increase in relative parameters was observed in the V-joint (V-hyperproduction). Such a situation is explained on the basis of academician R.V. Petrov's "mobile principle", that is, the immune system is explained by the decrease of one joint and the increase of another joint, so that the immune system keeps its activity in balance. In this case, the deficiency of SD3+-lymphocytes caused the hyperproduction of SD20+-lymphocytes and provided the immune response, but still this condition represents stress on the immune system. It can be seen that the absolute numbers had a trend of changes in the same way as the indicators of the relative numbers (Figure 3). Although the intensity of the changes differed, the trend of the changes was the same with the relative parameters.

At the next stage of scientific work, the relative and absolute quantities of proliferating lymphocytes (SD25+-lymphocytes) indicating the activation of immune system cells and apoptotic cells (SD95+-lymphocytes) indicating the apoptosis of immunocompetent cells were determined, the results were interpreted and analyzed.

Fig 3. Comparative indicators of absolute amounts of immune system cells in adult patients with chronic kidney disease, in absolute numbers

The relative and absolute parameters of the obtained indicators were reliably high in patients. If we look at the numbers, it turns out that the relative parameter of D25+-lymphocytes was significantly higher in patients by 1.80 times compared to healthy people - 25.26±0.49% versus 14.06±0.32%, respectively (R<0.001). A similar trend was observed for the absolute amount of this lymphocyte - 277±6 μl versus 440±9 μl, respectively (1.59-fold difference, R<0.001).

The increase of this cell in the peripheral blood of patients indicates that the activity of the immune system has increased, the amount of proliferating cells has increased, and the proliferation process has exceeded the norm. So, in order to ensure the functioning of the immune system, this system is strained, which means an increase in the number of proliferating (activated) lymphocytes.
A similar trend of changes in SIt was also observed on D95+-lymphocytes. It is known that apoptosis is the programmed death of cells, which normally occurs at the same time, but as a result of various effects, it becomes stronger and accelerates the death of immunocompetent cells. This causes a deficiency in the immune system (secondary immunodeficiency). In this case, SD95+-lymphocytes, like SD25+-lymphocytes, showed a statistically significant increase in relative and absolute amounts in patients compared to healthy individuals - on average 21.60±0.59% versus 16.89±0.47% (1.28 times difference, R<0.05) and 376±10 μl vs. 332±9 μl (1.13-fold difference, R<0.05). This situation indicated that the immune system is working under strain, and it is also an attempt to restore the immune balance by compensating the activity of immunocompetent cells against the background of secondary immunodeficiency.

At this point, S. is among the non-specific protective factors. We found it permissible to dwell on the results of the determination of natural killers that capture the D16+-marker on their surface. These NK-cells (natural killers) perform the task of identifying and destroying mutant or tumor cells that are generated in the body, regardless of antigen exposure. As a result of the formation of SBK, the tendency to the formation of mutant cells increases under the influence of chronic inflammation. This can be proved by the increase in relative and absolute values of SD16+-lymphocytes detected in patients. These indicators were 27.21±0.48% and 473±8 μl in patients with SBK, respectively, and were reliably higher than the parameters of healthy subjects by an average of 2.36 and 2.08 times -11.53±0.36%, respectively and 227±7 μl (P<0.001). This situation indicated a sharp activation of SD16+-lymphocytes in the body (Fig. 4). This situation is a sign of activation of SD16+-lymphocytes in the body, which means an increase in mutant cells being formed in the body. It has been proven that this condition is the result of negative changes in the body's immune system.

In addition to cellular (T- and V-cells of the immune system), non-specific protective factors (SD16+-lymphocytes), proliferative (SD25+-lymphocytes) and apoptosis cells (SD95+-lymphocytes), humoral immunity was studied in the body's immune system. In it, the concentrations of immunoglobulins (Ig) of the main class in the blood serum of patients and healthy individuals were comparatively analyzed. Clinically important immunoglobulins A, M, G (IgA, IgM, IgG) were detected in it. Their concentrations in blood serum were determined by traditional methods using IFA.

According to the results, the amount of immunoglobulins in healthy individuals was as follows: IgA - 1.26±0.15 g/l, IgM - 1.33±0.13 g/l, IgG - 13.53±0.89 g/l. In patients diagnosed with SBK, their blood serum concentrations changed differently, and an imbalance in their amount was observed, which was considered a sign of the development of a pathological condition.

It is known that IgA, found in blood serum, passes through tissues to the surfaces of mucous membranes in the form of slgA, providing local immunity. In this case, it was observed that the amount of immunoglobulins in patients increased reliably up to 4.15 times compared to healthy people - respectively 5.23±0.38 g/l versus 1.26±0.15 g/l (R<0.001). This condition is specific for SBK, this immunoglobulin was not increased in other pathologies, which was explained by the specific aspects of the pathological condition in the kidney.

Correspondingly, but smaller changes were also observed for IgM (R<0.05). It is known that IgM is distinguished among the immunoglobulins in the blood serum by its large molecular mass (900 daltons), the fact that it is the first to be synthesized when the antigen is exposed, it cannot pass through the placenta, and its structure is in the form of a pentamer. Also, this immunoglobulin is not synthesized in the secondary immune response, as it provides the primary immune response. In the studied pathology, its concentration was reliably unchanged in patients compared to healthy people - 1.60±0.20 g/l versus 1.33±0.15 g/l, respectively (R>0.05).
The most important among the studied immunoglobulins is IgG, which makes up 75% of all immunoglobulins in the blood serum. In the case of the primary immune response, it begins to be synthesized 5-6 days after the antigen enters the body and reaches its maximum on the 28th day, begins to synthesize. At the same time, unlike others, its molecular mass is small, so it can pass through the placenta. In other words, it is an immunoglobulin that performs the main function in inflammatory processes. The amount of this immunoglobulin in the studied SBK was significantly lower in patients by 1.93 times compared to the values of the control group - 13.53±0.89 g/l against 7.00±0.25 g/l, respectively (R<0.001).

This determined imbalance in the concentration of immunoglobulins in blood serum in SBK was explained by the specificity of the pathogenesis of the disease, the chronic nature of the pathological process, and the fact that the tendency of the inflammatory process to decrease is not observed.

In the process of studying the immune status of patients diagnosed with SBK, interpreting and analyzing the actual results, the following specific immunological features were identified:

first of all, all 20 indicators of immune status (relative number of lymphocytes, except for IgM concentration) were found to have changed reliably (R<0.05 - R<0.001), depending on the state of the immune system, their amount was below or above the normal limits, imbalance of immunocompetent cells was assessed as secondary immunodeficiency caused by a pathological condition in the body;

secondly, the relative and absolute amounts of the T-joint of the immune system were deficient compared to the normal parameters, while the relative and absolute amounts of the V-joint were reliably increased, this imbalance was explained by the "mobile principle";

thirdly, the ratio of proliferating active lymphocytes (SD25+-lymphocytes) and apoptotic cells (SD95+-lymphocytes) to the norm was statistically significantly increased in patients, which was explained by the state of immune system stress as a result of a chronic pathological process;

fourthly, the increase in relative and absolute amounts of SD16+-lymphocytes in the blood of patients is a sign of sharp activation of these cells, an increase in mutant cells generated in the body, which was interpreted as a consequence of negative changes in the immune system;

from the fifth, there was an imbalance in the concentrations of immunoglobulins in the blood serum of patients, while IgM remained within the norm, the amount of IgA increased dramatically, and the concentration of IgG decreased accordingly.

In order to clearly visualize the changes in the immune system of the studied SBK patients, the ratio of all indicators to the indicators of healthy individuals was presented and formed as a table.

Table 1. In patients with chronic kidney disease, the level of changes in the immune system compared to the norm, times

<table>
<thead>
<tr>
<th>Indicators</th>
<th>The degree of changes compared to the norm, times</th>
</tr>
</thead>
<tbody>
<tr>
<td>Leukocytes</td>
<td>1.10 * ↓</td>
</tr>
<tr>
<td>Lymphocytes</td>
<td>1.03 ↔</td>
</tr>
<tr>
<td>SD3+ lymphocytes</td>
<td>1.43 * ↓</td>
</tr>
<tr>
<td>SD4+ lymphocytes</td>
<td>1.07 * ↓</td>
</tr>
<tr>
<td>SD8+ lymphocytes</td>
<td>–1.25 * ↑</td>
</tr>
<tr>
<td>IRI</td>
<td>1.32 * ↓</td>
</tr>
<tr>
<td>SD20+ lymphocytes</td>
<td>–1.19 * ↑</td>
</tr>
<tr>
<td>SD25+ lymphocytes</td>
<td>–1.80 * ↑</td>
</tr>
<tr>
<td>SD95+ lymphocytes</td>
<td>–1.28 * ↑</td>
</tr>
<tr>
<td>SD16+ lymphocytes</td>
<td>–2.36 * ↑</td>
</tr>
<tr>
<td>IgA</td>
<td>–4.15 * ↑</td>
</tr>
<tr>
<td>IgM</td>
<td>1.20 ↔</td>
</tr>
<tr>
<td>IgG</td>
<td>1.93 * ↓</td>
</tr>
</tbody>
</table>

Note: * - sign of a reliable difference compared to the indicators of healthy individuals; ↑, ↓ - directions of changes; ↔ - there is no convincing difference; numbers with a negative sign are higher than those of healthy individuals.

Today, along with the cellular and humoral links of the immune system, the description of its cytokine status is one of the main conditions for evaluating the activity of this system. Assessment of cytokine status not only allows patients to assess the state of the immune system, but also to determine the trend and intensity of its changes. According to today's scientific sources, cytokines include: interferons, colony-stimulating factors, tumor necrosis
factor, interleukins, growth factor group, chemokines. The tasks performed by these groups are different, and their study is determined depending on the pathological condition.

These endogenous mediators were identified because pro-inflammatory and anti-inflammatory interleukins are important in inflammatory processes. Pathogenically important anti-inflammatory interleukin-4 (IL-4) and pro-inflammatory IL-6 were identified and the results evaluated. The concentrations of these cytokines were determined in the blood serum of patients using conventional enzyme immunoassay (IFA).

The obtained results showed that the serum concentration of IL-4 was 5.85±0.99 pg/ml, and the corresponding amount of IL-6 was 155.95±0.66 pg/ml in healthy individuals without SBK diagnosis.

If we logically derive from the activity of cytokines, it is natural that the amount of anti-inflammatory cytokines (for example, IL-4) should be low in the absence of inflammation, and the amount of pro-inflammatory cytokines should be higher than anti-inflammatory ones. If we take into account that anti-inflammatory cytokines suppress the transcription of their genes in pro-inflammatory cytokine-producing cells, induce the synthesis of interleukin receptor antagonists, increase the formation of soluble receptors, and reduce the density of pro-inflammatory receptors on the surface of cells through down-regulation, the compatible antagonistic activity of these cytokines is shown. will be in pathologies where various inflammatory processes are observed, the violation of this ratio ensures the strength and duration of this process [7].

The concentration of IL-4 in the blood serum of patients with SBK was markedly increased by 11.84 times - 69.27±5.50 pg/ml versus 5.85±0.99 pg/ml, respectively (R<0.001). It should be noted that such a sharp increase of IL-4 as an anti-inflammatory cytokine indicates the development of the inflammatory process in patients with SBK and its persistence. If we take into account that IL-4 induces the differentiation of normal T-helpers (Th0-cells) into Th2-cells in the immune system, the mechanism of their action in this studied pathology becomes clear.

Given that the concentration of pro-inflammatory cytokine IL-6 is increased during inflammatory processes, its clinical and immunological significance is known. However, it was found that the amount of this cytokine in the blood serum of patients with SBK was 106.67±11.58 pg/ml and was 1.46 times less than the parameters of healthy people (R<0.001). This situation was explained by the fact that the inflammatory process was observed for a long time in these patients, and the concentration of anti-inflammatory cytokines was high (Fig. 5).

![Fig 5. Comparative parameters of IL-4 and IL-6 concentrations in blood serum of patients with SBK, pg/ml](image_url)

The differences of studied cytokines between patients and healthy people are clearly visible from the given figure 5. This situation indicates that there was no reduction in the inflammatory process in the patient's body.

In this case, it was shown that the inflammatory process developed in these patients, as a result of which the strength and intensity of the immune response was low. Due to the fact that cytokine changes can provide information about the pathogenesis of the disease and its outcome, the dynamic determination of IL-4 and IL-6 concentrations in blood serum was recommended as diagnostic and prognostic immunological criteria for patients diagnosed with SBK.

4 Conclusions

It was found that there were significant changes in the immune system of adult patients with SBK, they reliably decreased SD3+ and SD4+ lymphocytes up to 1.43 and 1.07 times, respectively, compared to control parameters, and the relative amount of SD8+ lymphocytes was statistically significant up to 1.25 times increased was expressed by a convincing decrease of IRI by 1.32 times, which was expressed in the form of T-immunodeficiency. SD20+ lymphocytes were statistically significant 1.19 times higher in patients with SBK compared to controls.

S. in the blood of patients diagnosed with SBKD25+-, SD95+-lymphocyte counts were significantly higher in patients compared to healthy individuals, with an average increase of 1.80- and 1.59-fold in SD25+-lymphocytes, respectively, and an average increase of 1.28- and 1.13-fold in SD95+-lymphocytes, respectively. was expressed, this situation indicated that the immune system was working hard, which was evaluated as an attempt to restore the immune balance by compensating their activity against the background of secondary immunodeficiency.

It was found that the amount of SD16+-lymphocytes in the peripheral blood of patients with SBK increased reliably compared to the parameters of healthy individuals - this difference was 2.36 times in relative terms, and
2.08 times in terms of absolute numbers. This situation is a sign of sudden activation of SD16+ lymphocytes in the body, which is a sign of an increase in mutant cells formed in the body, and it was proved that the increase of natural killers is a consequence of negative changes in the body's immune system.

In patients diagnosed with SBK, there was an imbalance in the concentration of immunoglobulins in the blood serum, if the IgA parameter increased by 4.15 times compared to healthy individuals, the opposite was observed in the IgG concentration - it was 1.93 times convincingly lower than the values of healthy people, the concentration of IgM did not differ convincingly, such an imbalance explained by the specificity of the pathological condition;

It was found that IL-4 in blood serum of patients with SBK increased by 11.84 times compared to the parameters of healthy individuals, while IL-6 significantly decreased by 1.46 times compared to the parameters of healthy individuals. Such a situation indicated that the inflammatory process in these patients had developed and was ongoing, as a result of which the strength and intensity of the immune response was shown to be low. Due to the fact that such changes of IL-4 and IL-6 can provide full information about the pathogenesis of the disease and its outcome, dynamic determination of their concentrations in blood serum was recommended as diagnostic and prognostic criteria for patients diagnosed with SBK.

References