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CYTOKINES AND IRON-CONTAINING PROTEINS LEVELS AT CHILDREN WITH PNEUMONIA AND ANEMIA


Sumy state university, Sumy, Ukraine

Objective: To examine levels of interleukins 4 and 8, transferrin and ferritin in children with community-acquired pneumonia and anemia.

Methods: By enzyme-linked immunosorbent assay was examined 74 children 1-3 years age at 1-4 day and 12-14 day of hospitalization.

Results: In children with pneumonia and anemia found a significant increase in the level of pro- and anti-inflammatory cytokines. Transferrin level was higher in children with pneumonia and anemia and ferritin levels, by contrast, was lower. There was a ferritin with interleukin 8 and a positive correlation with negative interleukin 4. Transferrin has a negative dependence on interleukin 8.

Conclusion: At children with pneumonia and anemia were observed more significant and lasting changes in the level of cytokines and iron-containing proteins.

Key words: community acquired pneumonia, anemia, cytokines, iron-containing proteins, children

Community-acquired pneumonia (CAP) is one of the most widespread infectious pathology with serious prognosis. The mortality rate of children with pneumonia is on average 13.1 per 100 000. Secondary immunodeficient states occupy significant place among the predictors of the pneumonia occurrence [1,2, 3]. As it is known, the age immaturity of specific and nonspecific immunological protection of the child during the first years of life is shown by more easy occurrence of an infectious disease, more severe course, significant risk of complications and generalization of the infection. A major contribution to the formation of an adequate immune response and regulation of intercellular interaction of all immunocompetent cells in realization of effect or protection belongs to cytokines, especially their balance of the oppositional pools. Thus, the occurrence of immune deficiency is the result of disbalance in cytokine system [3, 4, 5].

Another factor that affects almost all links of the immune reactions is the microelement iron. Deficient income of this element to the organism of the child is accompanied by decreased activity of natural killer cells, neutrophils, insufficient production of .myeloperoxidase, synthesis of IgAand IgG, as well as inefficient and inadequate for the level of stimulation the production of several cytokines [6, 7, 8].

The factors of nonspecific humoral protection also include iron-containing proteins ferritin and transferrin, the level of which is impaired by iron deficiency anemia (IDA). These proteins are involved into iron metabolism in the organism, functioning of the cellular and humoral-immunity systems, has cytotoxic and cytotropic characteristics [6,7,8,9,10].

That is why the search for new criteria for determining the degree and nature of the immunological response during the community-acquired pneumonia, associated with iron deficiency anemia in preschool children is actual and priority area of research in Pediatrics.

The aim of this work was to study the content of interleukin (IL) 4 and 8, transferrin and ferritin and their interconnections in the blood serum of preschool children, who are suffering from the community-acquired pneumonia, associated with iron deficiency anemia.

Materials and methods

Under the medical observation were 55 patients with the CAP, aged from 1 to 2 years old, who were on an inpatient treatment in the department of
infectious disease of "Sumy city children’s clinical hospital of St. Zinaida". Depending on the presence of IDA children were divided into the following groups: group I - patients with CAP without IDA (23 children); group II - patients with CAP and with the mild IDA (32 children). The control group included 19 almost healthy peers.

The diagnosis of pneumonia verified on the basis of complaints from parents of sick children, anamnesis, objective symptoms, data of laboratory and instrumental methods of examination according to national protocols of medical care to children with pulmonary diseases. The degree of iron deficiency anemia was evaluated in accordance with the level of hemoglobin according to the national protocols of medical care to children with pulmonary diseases. The content of serum ferritin (SF) in patients of group I at the end of the treatment; P5-6 - the difference between the indices of the children of 1 group at the beginning of treatment; P2-5 - the difference between the indices of the children of the control group and of 1 group at the end of the treatment; P3-4 - the difference between the indices of the children of 1 group at the beginning of treatment; P2-4 - the difference between the indices of the control group and of 1 group at the beginning of treatment; P2-3 - the difference between the indices of the children of the control group and of 1 group at the end of the treatment; P5-6 - the difference between the indices of the children of 1 group at the beginning of treatment; P2-6 - the difference between the indices of the children of the control group and almost healthy peers.

Table 1. Dynamics of the cytokines’ level in the preschool aged children, who are suffering from the community-acquired pneumonia, associated with iron deficiency anemia, M±m

<table>
<thead>
<tr>
<th>Index (PG/ml)</th>
<th>Control group n=19</th>
<th>Group 1 n=22</th>
<th>Group II n=32</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Beginning of the treatment (n=22)</td>
<td>End of the treatment (n=11)</td>
<td>Beginning of the treatment (n=32)</td>
</tr>
<tr>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>2</td>
<td>100,75±6,01</td>
<td>134,62±13,57</td>
<td>122,26±6,29</td>
</tr>
<tr>
<td>p&lt;0,001</td>
<td>p&lt;0,001</td>
<td>p&lt;0,05</td>
<td>p&lt;0,001</td>
</tr>
<tr>
<td>IL-4</td>
<td>22,44±1,61</td>
<td>48,95±10,18</td>
<td>73,92±6,79</td>
</tr>
<tr>
<td>p&lt;0,001</td>
<td>p&lt;0,001</td>
<td>p&lt;0,05</td>
<td></td>
</tr>
<tr>
<td>IL-8</td>
<td>0,21±0,03</td>
<td>22,88±11,48</td>
<td>73,92±6,79</td>
</tr>
<tr>
<td>p&lt;0,001</td>
<td>p&lt;0,05</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Notes: P2-3 - the difference between the indices of the children of the control group and of 1 group at the beginning of treatment; P2-4 - the difference between the indices of the children of the control group and of 1 group at the end of the treatment; P3-4 - the difference between the indices of the children of 1 group at the beginning and at the end of the treatment; P2-5 - the difference between the indices of the children of the control group and of 1 group at the beginning of treatment; P3-5 - the difference between the indices of the children of 1 and 2 groups at the beginning of treatment; P2-6 - the difference between the indices of the children of the control group and of 1 group at the end of the treatment; P5-6 - the difference between the indices of the children of 1 group at the beginning and at the end of the treatment.

The analysis of interleukin-8 in patients of the group I at the beginning of the disease determined significant increase of children indices of the control group (Table 1). The concentration of interleukin-4 in patients of this subgroup was also increasing significantly.

Patients of group II were marked by increased levels of interleukin-8 comparing to the indices of almost healthy children and patients of group I. The content of interleukin-4 in patients of group II were also increased as compared to children in the control group and group I.

In the analysis of the level of serum transferrin (STF) in children of the groups I at the beginning of the illness significant reduction is determined relative to the index of the control group (Table 2). The content of serum ferritin (SF) in patients of group I was significantly increased. The patients of group II were marked with higher content of STF against indexes of almost healthy children. Meanwhile, the level of SF in patients of group II was decreased comparing to the children in the control group. While the comparison the data received from groups I and II of children at the beginning of the illness, was found that patients with CAP with IDA concentration of STF was significantly higher than in patients with CAP without, and the content of SF was significantly lower.
Table 2. Dynamics of the concentration of transferrin and ferritin in the blood serum of preschool aged children, who are suffering from community-acquired pneumonia, (M±m)

<table>
<thead>
<tr>
<th>Index</th>
<th>1 control group (n=18)</th>
<th>Group I (n=23)</th>
<th>Group II (n=32)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Beginning of the treatment (n=23)</td>
<td>End of the treatment (n=12)</td>
<td>Beginning of the treatment (n=32)</td>
</tr>
<tr>
<td>Transferrin, mg/DL</td>
<td>278.24±11.37</td>
<td>139.17±16.93</td>
<td>311.33±24.26</td>
</tr>
<tr>
<td></td>
<td>P&lt;0.001</td>
<td>P&lt;0.001</td>
<td>P&lt;0.001</td>
</tr>
<tr>
<td>Ferritin, ng/ml</td>
<td>58.25±5.19</td>
<td>121.50±7.50</td>
<td>80.93±9.53</td>
</tr>
<tr>
<td></td>
<td>P&lt;0.001</td>
<td>P&lt;0.001</td>
<td>P&lt;0.001</td>
</tr>
</tbody>
</table>

Notes: p - credibility of differences; p1-2, p1-4 - between indices of children of control group and 1 and 2 groups at the beginning of the treatment; p2-4 - between indices at the beginning of the treatment of children from 1 and 2 groups; p2-3, p4-5 - between indices at the beginning and at the end of the treatment of children from 1 and 2 groups; p1-3, p1-5 - between indices of children of control group and children from 1 and 2 at the end of the treatment.

To determine a interconnections between the content of iron-containing proteins and cytokines, we analyzed the correlation between concentrations of serum ferritin, transferrin and IL-4, IL-8 (Table 3).

<table>
<thead>
<tr>
<th>* Index</th>
<th>Group 1</th>
<th>Group II</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ferritin/IL-4</td>
<td>-0.5652**</td>
<td>-0.5359**</td>
</tr>
<tr>
<td>Ferritin/IL-8</td>
<td>0.5202*</td>
<td>-0.5271**</td>
</tr>
<tr>
<td>Transferrin/IL-4</td>
<td>-0.3195</td>
<td>0.5643**</td>
</tr>
<tr>
<td>Transferrin/IL-8</td>
<td>0.4804*</td>
<td>0.4444*</td>
</tr>
</tbody>
</table>

Note. The credibility of differences between the indicators of the content of transferrin, ferritin and indicators of the levels of interleukins 4 and 8: * - p<0.05, ** - p<0.01.

Thus, among children in the group I was found negative correlative interconnection of the average force between the concentrations of ferritin and IL-4, the levels of transferring and IL-8 and positive correlation of medium strength ferritin/IL-8. Patients of the group II had a negative correlation of medium strength ferritin/IL-4, ferritin/IL-8 and a positive correlative interconnections between the content of transferring and IL-4, IL-8. The increase in the content of proinflammatory IL-8 in the acute period, perhaps, is the manifestation of the cells reaction of the monocyte - macrophage link to antigenic timulus. Increased levels of IL-4 shows the parallel activation of anti-inflammatory defense mechanisms of the organism. Along with this, there was significantly high production of interleukins of children, who has CAP with IDA, comparing to patients with CAP without IDA may indicate the intensity and severity of the infectious process in the organism. It also gives reason to consider changes in the content of IL-8 and IL-4 as a marker of activity of the inflammatory response in the child's organism [3, 4, 5].

In the period of acute inflammation were found changes in the levels of iron-containing proteins in children, who has CAP without IDA, possibly connected with the peculiarities of the influence of cytokines on the iron transportation, reducing the availability of bimetal for pathogens in the body, by reducing the number of transferrin receptors on the cell surface and increased synthesis of ferritin for iron depositing. Meanwhile, in the case of iron deficiency in patients with CAP with IDA reverse processes take place: suppressed synthesis of ferritin and increased level of transferring [6, 7, 8].

In the reconvalescence period the above mentioned figures have improved, but the distinctness of these changes also depended on the presence of IDA in children. Patients of the group I had significant increase of the level of interleukin-4 as to the indicator during acute period, whereas the content of interleukin-8 in the dynamics of treatment did not change. However, both indicators did not reach normal indices in healthy children.

The children in group II had increased level of interleukin-4 as to indexes of patients during acute period and almost healthy children and a trend to
decrease in the content of interleukin-8, but remained significantly higher than in the control group.

All patients in the reconvalescence period had certain dynamics in levels of iron-containing proteins, however, the degree of manifestation of the changes depended on the presence of IDA. So, in the dynamics of treatment in patients of the group I was significant increase in the level of STF, as well as reducing the amount of the SF. Along with this full normalization of both indicators didn’t occur. Patients of group II had further increase in the content of STF, which was significantly higher than the index of patients during the acute phase (p < 0.05) and the children of the control group (p < 0.001). In the blood serum of patients from group II was lower level of SF compared to patients during acute period and almost healthy children (table 2).

Therefore, in the early reconvalescence period, the level of IL-8 remained increase without clinical display of the illness, which indicates the incompleteness of the inflammatory process. Synthesis of anti-inflammatory IL-4 in the dynamics of illness is greatly increased, especially when concomitant iron deficiency anemia, which could indicate a structural alteration of the immune response towards an anti-inflammatory orientation with the stimulation of humoral, cellular and local levels of protection.

Patients with CAP without IDA reconvalescence was characterized only by improving the content of transferrin and ferritin, but they did not reach their level of almost healthy children. More significant imbalance of iron-containing proteins was observed in patients with CAP, associated with the IDA, which is probably affected by iron deficiency in the organism, that was increased during infectious process. This is confirmed by increased level of transferrin (p<0.05) and decreased ferritin (p<0.001) in patients with community-acquired pneumonia associated with iron deficiency anemia.

**Conclusion**

In children with pneumonia was an increase in the level of pro- and anti-inflammatory cytokines, more significant in the presence iron deficiency anemia.

The quantities of iron-containing proteins also changed, which was characterized by decrease transferrin and increase ferritin and was more pronounced in children with pneumonia and anemia.

Changes in the level of pro-and anti-inflammatory cytokines and iron-containing proteins had a lasting character and traced throughout the entire period of observation.

**List of literature:**


