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ABSTRACT

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PROGNOSTIC VALUE OF IL-4, IL-6, AND IL-8 LEVELS IN CHILDREN WITH SEVERE COMMUNITY-ACQUIRED PNEUMONIA IN RELATION TO SARS-COV-2 INFECTION

Introduction. Pneumonia is one of the most dangerous infectious diseases and a common reason for hospitalization among children. Interleukins (ILs) play a key role in activating the immune response during inflammation and in subsequent tissue repair. Assessing cytokine levels can be important for determining disease severity and outcomes.

Objective: To evaluate changes in anti-inflammatory IL-4 and pro-inflammatory IL-6 and IL- 8 levels in children with severe community-acquired pneumonia (CAP), taking into account SARS- CoV- 2 infection.

Materials and Methods. We examined 81 children with severe CAP, including 52 patients with confirmed SARS-CoV-2 infection and 29 patients without coronavirus infection. The control group consisted of 32 somatically healthy children. Cytokine levels were determined by ELISA during both the acute phase and the convalescent period. Statistical analysis was performed using Microsoft Excel and JASP. A two-tailed independent Student's *t*-test was applied, with $p < 0.05$ considered statistically significant.

Results. The acute phase of pneumonia in children was characterized by elevated levels of both the anti-inflammatory cytokine IL-4 and the pro-inflammatory cytokines IL-6 and IL-8, particularly in patients infected with SARS-CoV-2. During convalescence, the levels of pro-inflammatory cytokines decreased, while anti-inflammatory IL-4 showed a further increase.

Conclusions. The changes in cytokine balance observed in children with CAP reflect the activity of the immune response to inflammation. These findings may serve as markers of disease severity, predictors of the clinical course of the infectious process, and indicators for evaluating treatment effectiveness.

Keywords: community-acquired pneumonia, cytokines, children, SARS-CoV-2, immune response.

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РЕЗЮМЕ

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ПРОГНОСТИЧНЕ ЗНАЧЕННЯ РІВНІВ IL-4, IL-6 ТА IL-8 У ДІТЕЙ З ТЯЖКОЮ ПОЗАЛІКАРНЯНОЮ ПНЕВМОНІЄЮ, ВРАХОВУЮЧИ ІНФЕКЦІЮ SARS-COV-2

Вступ. Пневмонія є однією з найнебезпечніших інфекційних хвороб і поширеною причиною госпіталізації дітей. Інтерлейкіни (ІЛ) відіграють ключову роль в активації імунної відповіді під час запалення та подальшого відновлення тканин. Оцінка рівня цитокінів може бути важливою для визначення тяжкості захворювання та його наслідків.

Мета. Оцінити зміни рівнів протизапального IL-4 та прозапальних IL-6 та IL-8 у дітей з тяжкою позалікарняною пневмонією (ПП), враховуючи інфекцію SARS-CoV-2.

Матеріали та методи. Ми обстежили 81 дитину з тяжкою ПП, з них 52 пацієнти з підтвердженою інфекцією SARS-CoV-2 та 29 без коронавірусної інфекції. Контрольна група складалася з 32 соматично здорових дітей. Рівень цитокінів визначали методом ІФА як у гострій фазі, так і в період одужання. Статистичний аналіз проводили за допомогою Microsoft Excel та JASP. Застосовували двосторонній незалежний t-критерій Стьюдента, при цьому $p < 0,05$ вважали статистично значущим.

Результати. Гостра фаза пневмонії у дітей характеризувалася підвищеним рівнем як протизапального цитокіну IL-4, так і прозапальних цитокінів IL-6 та IL-8, особливо у пацієнтів, інфікованих SARS-CoV-2. Під час одужання рівень прозапальних цитокінів знизився, тоді як протизапальний IL-4 показав подальше підвищення.

Висновки. Зміни в балансі цитокінів, що спостерігаються у дітей з ПП, відображають активність імунної відповіді на запалення. Ці дані можуть слугувати маркерами тяжкості захворювання, предикторами клінічного перебігу інфекційного процесу та показниками для оцінки ефективності лікування.

Ключові слова: позалікарняна пневмонія, цитокіни, діти, SARS-CoV-2, імунна відповідь.

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ABBREVIATIONS

CAP — community-acquired pneumonia
IL — interleukin

INTRODUCTION

Acute respiratory diseases, particularly community-acquired pneumonia (CAP), remain a major problem in pediatrics. Pneumonia is among the leading causes of infectious morbidity and mortality in children [1, 2]. The development and implementation of new treatment protocols and vaccination programs have reduced the prevalence and mortality of CAP; however, these rates remain high, especially in young children [1, 3, 4]. Importantly, this pathology also affects older pediatric age groups. According to national data, the incidence of pneumonia has shown an increasing trend over recent decades [5, 6]. The etiological structure of pneumonia is diverse: bacteria, viruses, fungi, combinations thereof,

some of which (in particular Klebsiella pneumoniae) can cause a complicated course of the inflammatory process [7, 8]. It should be noted that in recent years, the spectrum of pathogens has changed, particularly in connection with the global coronavirus pandemic [6, 9]. The severity of pulmonary inflammation depends on both the etiological factor and the intensity of the immune response [9, 10]. The latter is regulated by the coordinated action of multiple components, including anti-inflammatory (IL-4) and pro-inflammatory (IL-6, IL-8) cytokines, whose balance is crucial for an adequate immune response [11, 12]. International studies have shown that excessively elevated levels of pro- and anti-inflammatory cytokines (a cytokine storm) may be associated with severe pneumonia, treatment failure and increased mortality [9, 12–17]. Researchers also suggest that measuring cytokine levels may reflect pneumonia severity and serve as a tool for monitoring treatment effectiveness [9, 18]. Nevertheless, despite progress in studying cytokine markers, the literature contains a limited number of studies assessing IL-4, IL-6, and IL-8 levels in patients aged 6 to 17 years with CAP, leaving an incomplete understanding of the pediatric immune response in pulmonary inflammation. These considerations justify the need to investigate cytokine levels in children with CAP, particularly in the context of SARS-CoV-2 infection.

Study objective: to evaluate changes in anti-inflammatory IL-4 and pro-inflammatory IL-6 and IL-8 in children aged 6 to 17 years with CAP during the acute phase and convalescence, taking into account SARS-CoV-2 infection.

MATERIALS AND METHODS

A total of 133 children of both sexes, aged 6 years to 17 years 11 months 29 days, were examined to achieve the study objective. Among them, 81 patients with community-acquired pneumonia were hospitalized in the Infectious Diseases Department of the Municipal Non-Commercial Enterprise "St. Zinaida Children's Clinical Hospital" of the Sumy City Council, where they received standard therapy. The mean age of these patients was (11.1 ± 0.34) years. Based on the presence of SARS-CoV-2 infection, they were divided into two groups:

- Group I — 52 children with CAP and confirmed SARS-CoV-2 infection.
- Group II — 29 children with CAP without coronavirus infection.

The control group included 32 somatically healthy children matched by sex; their mean age was (10.84 ± 0.57) years (p>0.05).

The diagnosis of pneumonia was established according to the Clinical Guideline *"Community-acquired pneumonia in children"* (2022). SARS-CoV-2 infection was confirmed by polymerase chain reaction (PCR) of nasopharyngeal swabs. Cytokine levels were measured in serum using enzyme-linked immunosorbent assay (ELISA). Biological samples were collected during the acute phase (hospital days 1–2). All measurements were performed using standard protocols and instructions.

Statistical analysis was conducted with Microsoft Excel (2013) and JASP software. The arithmetic mean (M) and standard error (m) were calculated. Independent samples were compared using a two-sided Student's t-test. A p-value <0.05 was considered statistically significant.

The study was carried out in compliance with the principles of the Helsinki Declaration. All procedures were non-invasive and did not cause psychological harm to patients.

RESULTS

We found significant changes in cytokine balance in children with CAP. The mean level of the anti-inflammatory cytokine IL-4 increased during the acute phase reaching (34.87±1.03) pg/mL, which was significantly higher than in controls (17.18±0.48) pg/mL (p<0.001). During convalescence, IL-4 increased further compared with the acute phase to (35.95±1.02) pg/mL (p>0.05) (Figure 1).

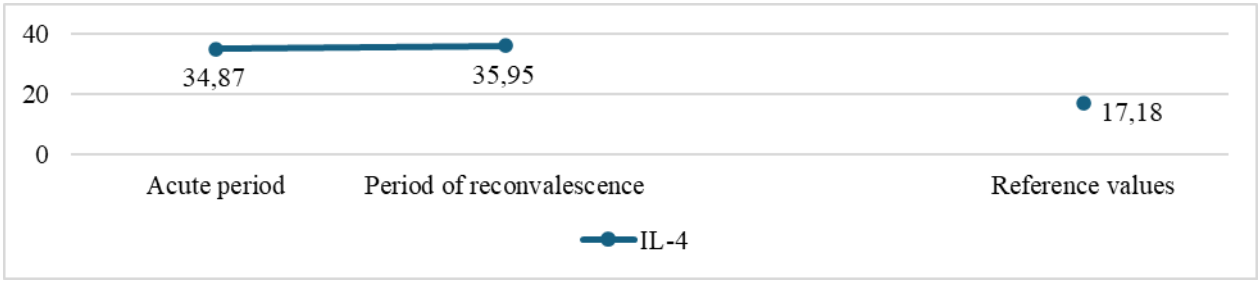

Figure 1. Dynamics of anti-inflammatory cytokine IL-4 by disease phase

Pro-inflammatory IL-6 and IL-8 also increased in the acute phase. Mean IL-6 was (21.38±0.92) pg/mL, significantly higher than in controls (4.92±0.28 pg/mL (p<0.001). IL-8 also increased significantly compared with healthy children (111.93±1.85) vs (23.53±0.63) pg/mL (p<0.001). After treatment mean IL-6 and IL-8

in children with CAP were (10.90±0.42) pg/mL (p<0.001) and (53.43±1.04) pg/mL (p<0.001) respectively. Notably, both pro- and anti-inflammatory cytokines in convalescence remained significantly higher than control values (p<0.001) (Figure 2).

In Group I (CAP with SARS-CoV-2 infection), both pro- and anti-inflammatory cytokines increased during the acute phase. IL-4 was (33.89±1.29) pg/mL, significantly higher than controls (p<0.001). IL-6 and IL-8 was also significantly elevated compared with healthy children: (22.07±1.15) pg/mL and (117.14±2.33) pg/mL respectively (both p<0.001). In Group II, cytokine concentrations were also higher than in controls: IL-4 (36.61±1.68) pg/mL (p<0.001); IL-6 (20.13±1.55) pg/mL (p<0.001); IL-8 (102.59±2.17) pg/mL (p<0.001).

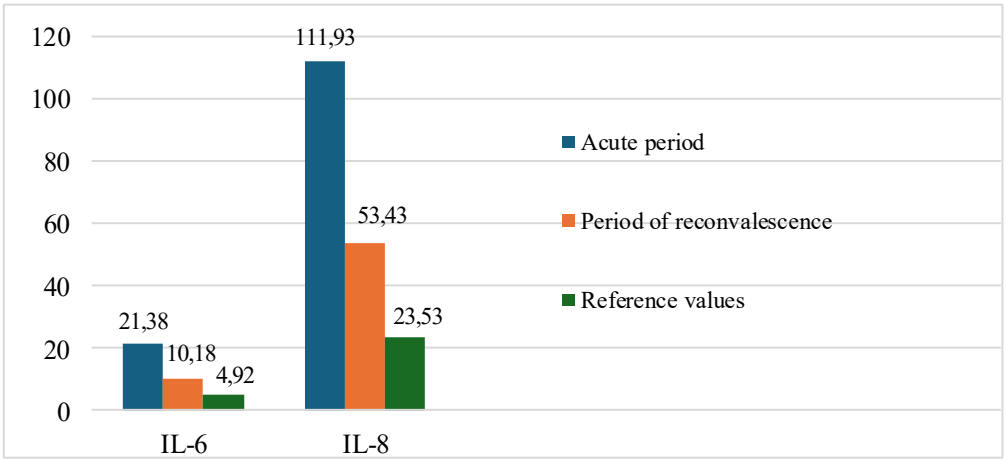

Figure 2. Dynamics of pro-inflammatory cytokines by disease phase

A comparative analysis of the two CAP groups in the acute phase showed that anti-inflammatory IL-4 and pro-inflammatory IL-8 were significantly higher in Group I (p<0.05 and p<0.001, respectively). IL-6 did not differ significantly between groups (p>0.05).

During convalescence in Group I, IL-6 and IL-8 decreased compared with the acute phase to (11.21±0.53) pg/mL and (56.0±1.34) pg/mL, respectively, but remained 2.27- and 2.37-fold higher than control values. IL-4 increased relative to the acute phase to (34.99±1.29) pg/mL, 2.03-fold higher than in controls. In Group II, IL-4 also increased during convalescence by 2.04-fold to (35.07±1.61) pg/mL. Pro-inflammatory cytokines decreased compared with the acute phase: IL-6 (10.18±0.71) pg/mL (2.06-fold higher than controls) and IL-8 (48.76±1.28) pg/mL (2.07-fold higher).

In convalescence, pro-inflammatory cytokines were significantly higher in Group I compared with Group II, whereas IL-4 did not differ significantly between groups. A detailed summary is presented in Table 1.

The described decreases in pro-inflammatory cytokines during convalescence suggest a reduction in inflammatory activity on standard therapy, possibly due to pathogen elimination, although levels remain elevated because the disease has not fully resolved. The increase in anti-inflammatory IL-4 indicates completion of the active inflammatory phase and transition to tissue repair.

Overall, these findings indicate a similar pattern of acute and convalescent phases of CAP in children. However, the immune response was more pronounced in patients with SARS-CoV-2 infection, which may indicate a more severe systemic inflammatory process.

DISCUSSION

Our study characterizes the cytokine balance (IL-4, IL-6, IL-8) in children with severe community-acquired pneumonia (CAP), with consideration of concurrent SARS-CoV-2 infection. The findings contribute to a better understanding of immune system dynamics in pediatric infectious diseases with severe clinical courses.

In line with previous reports, we observed significant elevations of both pro-inflammatory (IL-6, IL-8) and anti-inflammatory (IL-4) cytokines during the acute phase of CAP [19–25]. Some international studies have reported only minor deviations in IL-6 levels during acute SARS-CoV-2 infection in children compared with age-matched norms [26–28]; however, those cohorts predominantly included patients with mild COVID-19. Other evidence indicates that IL-8 levels are elevated in severe pneumonia and are particularly high in children with severe SARS-CoV-2 infection compared with mild cases [29].

Conversely, several publications report no significant differences in cytokine profiles (IL-4, IL-6) between mild and severe coronavirus-associated pneumonia [22, 28]. In adults with COVID-19, decreased IL-4 has been described, which may reflect age-related differences in immune response [29].

Table 1 – Levels of pro-inflammatory and anti-inflammatory cytokines in patients with community-acquired pneumonia

Indicator	Acute period		Period of convalescence		Reference values (n=32)
	Group I (n=52)	Group II (n=29)	Group I (n=52)	Group II (n=29)	
IL-4, pg/mL	33,89±1,29 p_{1-2}<0,05 p_{1-3}>0,05 p_{1-5}<0,001	36,61±1,68 p_{2-4}>0,05 p_{2-5}<0,001	34,99±1,29 p_{3-4}>0,05 p_{3-5}<0,001	37,28±1,66 p_{4-5}<0,001	17,18±0,48
IL-6, pg/mL	22,07±1,15 p_{1-2}<0,001 p_{1-3}<0,001 p_{1-5}<0,001	20,13±1,55 p_{2-4}<0,001 p_{2-5}<0,001	11,21±0,53 p_{3-4}<0,001 p_{3-5}<0,001	10,35±0,69 p_{4-5}<0,001	4,92±0,28
IL-8, pg/mL	117,14±2,33 p_{1-2}<0,001 p_{1-3}<0,001 p_{1-5}<0,001	102,59±2,17 p_{2-4}<0,001 p_{2-5}<0,001	56,0±1,34 p_{3-4}<0,001 p_{3-5}<0,001	48,76±1,28 p_{4-5}<0,001	23,53±0,63

Notes: P – significance of data discrepancies, p_{1-2} – significance of differences in pro- and anti-inflammatory cytokine levels in the blood serum of patients in groups I and II during the acute phase of the disease, p_{1-3} – reliability of the difference in serum cytokine levels between patients in group I during the acute and convalescence periods, p_{1-5} – reliability of the difference in serum cytokine levels in patients in groups I and the control group , p_{2-4} – reliability of indicators for patients in group II during the acute and convalescent periods, p_{2-5} – reliability of values for patients in groups II and the control group, p_{3-4} – reliability of indicators for patients in groups I and II during the convalescent period, p_{3-5} – reliability of indicators of group I during the convalescence period and control values, p_{4-5} – reliability of values of patients in group II and control values

According to published data, CAP convalescence is generally characterized by a reduction in pro-inflammatory cytokines and a modest rise in anti-inflammatory IL-4 [29, 30].

Taken together, our results confirm that severe pediatric pneumonia is accompanied by substantial immunological changes, which are particularly pronounced in the presence of SARS-CoV-2 infection. Cytokine profiling may therefore serve as a valuable marker for prognostication and treatment monitoring in this patient population.

CONCLUSIONS

- The acute phase of severe CAP in children is characterized by increased pro-inflammatory (IL- 6, IL-8) and anti-inflammatory (IL-4) cytokines.
- SARS-CoV-2 infection is associated with a more pronounced immune response compared with CAP without coronavirus infection.
- During convalescence, pro-inflammatory cytokines decrease but remain above control values; anti-inflammatory IL-4 increases. The cytokine profile may be used to assess treatment effectiveness and to predict disease course.

PROSPECTS FOR FUTURE RESEARCH

Future studies should comprehensively assess cytokine profiles in children with CAP, considering disease severity, comorbidities and complications. It is also advisable to investigate cytokine dynamics under modified therapies and their association with disease duration and outcomes.

AUTHOR CONTRIBUTIONS

The author independently conducted all stages of the research, including conception, data collection and study execution, analysis and statistical processing of the results and formulation of conclusions. The author wrote, edited and approved the final manuscript.

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CONFLICT OF INTEREST

The author declares no conflict of interest.

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ARTIFICIAL INTELLIGENCE DISCLOSURE

The author did not use artificial intelligence technologies in writing or editing the manuscript.

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