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V.V. Shcherbak, T.O. Kryuchko, L.M. Bubyr, O.V. Izmailova, O.A. Poda
Poltava State Medical University, Poltava, Ukraine

Changes of immunological biomarkers in pediatric patients with seasonal allergic rhinitis

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Abstract. ***Background.*** *Seasonal allergic rhinitis shows a constant upward trend not only among the adults but also in the children's population. A significant percentage of patients have a moderate and severe course of the disease, which leads to a significant deterioration in their quality of life. Since the pathogenesis of allergic rhinitis is represented by a complex cascade of immunological reactions involving a number of cytokines that play a role in the development of allergic inflammation of the upper respiratory tract, the priority direction of our work was an in-depth study of the pathogenetic mechanisms of this disease. The purpose was to investigate the levels of interleukin-33 (IL-33) and its suppression of tumorigenicity 2 (ST2) receptor in the blood serum of children with hay fever, depending on the age and severity of the disease, followed by the determination of correlations of these indicators with various clinical and immunological parameters of seasonal allergic rhinitis.* ***Materials and methods.*** *Forty-two patients aged 6–17 years with seasonal allergic rhinitis and 26 healthy children (controls) took part in the study. Severity of nasal and extra-nasal symptoms of allergic rhinitis was determined using a visual analog scale. Skin prick testing was performed according to generally accepted standards during remission using a standard pollen panel. Quantitative measurement of serum indicators of interleukin-33 and ST2 was carried out by the method of solid-phase enzyme immunoassay. A mandatory condition for conducting the study was the presence of written informed consent from the parents.* ***Results.*** *The study of serum levels of IL-33 and ST2 showed statistically higher concentrations in the group of children with a severe course of seasonal allergic rhinitis, a direct correlation of medium strength between content of the above cytokines in children with a mild SAR ($\tau = 0.65$; $p < 0.05$) and a strong dependence in patients with moderate ($\tau = 0.76$; $p < 0.01$) and severe ($\tau = 0.80$; $p < 0.05$) course. When comparing the mean values of IL-33 and ST2, no significant changes were found depending on age and the presence of mono- or polysensitisation among patients with allergic rhinitis. In children with seasonal allergic rhinitis and concomitant bronchial asthma, there was a statistically significant tendency towards a more severe course of the disease with correspondingly higher values of IL-33 and ST2 compared to patients with isolated seasonal allergic rhinitis.* ***Conclusions.*** *The obtained results of the immunological study made it possible to emphasize the diagnostic significance of IL-33 and its receptor ST2 as potential biomarkers in the development of allergic inflammation in pediatric patients with seasonal manifestations of rhinitis.*

Keywords: *allergic rhinitis; children; biomarker; cytokines; bronchial asthma; allergy*

Introduction

Seasonal allergic rhinitis (SAR), also known as hay fever, or pollinosis, is a fairly common allergic disease [1, 2]. According to foreign data, the precise prevalence of seasonal allergic rhinitis among children changes in different geographical regions and climatic conditions [3]. However, in general, 10 to 40 % of children suffer from SAR [4]. It is important to keep in mind that these figures may vary depending on factors such as genetic predisposition, allergen exposure, and air quality [5]. In recent years, many scientific papers have demonstrated an upward trend in allergic rhinitis (AR) cases but the figures are not precise and have a wide range of variations, which, accordingly, requires further epidemiological studies [6].

Patients with SAR have a significantly reduced quality of life due to the impact of disease symptoms on different are-

Для кореспонденції: Щербак Вікторія Валеріївна, асистент кафедри педіатрії № 2, Полтавський державний медичний університет, вул. Шевченка, 36, м. Полтава, 36040, Україна; e-mail: v.shcherbak@pdmu.edu.ua; тел. +38 (099) 780 87 01
For correspondence: Shcherbak Viktoriia, Assistant of the Department of Pediatrics 2, Poltava State Medical University, Shevchenko st., 36, Poltava, 36040, Ukraine; e-mail: v.shcherbak@pdmu.edu.ua; tel. +38 (099) 780 87 01
Full list of authors' information is available at the end of the article.

as of daily activity, reducing activity and learning at school, sports, etc. [7]. Children of primary and secondary school age are more affected, complaining more often of impaired concentration, memory, and task performance. In addition, symptoms such as nasal congestion and coughing interfere with a child's normal sleep which further leads to daytime fatigue, malaise, and irritation.

Timely and qualified treatment of patients is a significantly important aspect, as it affects the well-being of a child and the progression of the pathological process with the development of sinusitis, otitis media, and asthma. There is a close relationship between allergic rhinitis and asthma, which has common pathogenetic mechanisms of inflammatory processes, as about 40 to 50 % of patients with AR have manifestations of asthma [8], which forms a new phenotype of the disease and can be characterised by a refractory course [9]. In turn, this creates prerequisites for the search for new therapeutic approaches to the management of such patients.

Protocol symptomatic pharmacotherapy of AR is the leading direction in the treatment of this disease, but according to foreign scientists, it remains ineffective for 20 % of patients [10]. However, despite a large number of studies on this problem, the issue of in-depth study of the pathogenesis of AR is becoming increasingly relevant to better understand the molecular mechanisms of allergic disease development and identify new potential targets in the pathological process.

In recent years, most foreign and domestic studies have focused on the role of interleukin-33 (IL-33) in patients with inflammatory, autoimmune, and allergic diseases [11, 12]. It is known that this cytokine interacts with suppression of tumorigenicity 2 (ST2) receptor and, accordingly, transmits its signals to cells [13]. ST2 is a transmembrane protein found on the surface of various cell types, including immune, epithelial, and endothelial cells. After interacting with IL-33, the ST2 receptor activates intracellular signaling pathways, which leads to the activation of the immune response [14]. Activation of the ST2 receptor promotes the production of other pro-inflammatory cytokines, such as IL-4, IL-5, and IL-13. These cytokines affect various cells of the immune system, promoting inflammatory reactions, and activation of eosinophils, mastocytes, and other cells [15]. Studies on the role of IL-33 and its ST2 receptor in the immunology and pathophysiology of AR are ongoing, as their detailed mechanism of action has not yet been fully elucidated.

The purpose was to study the levels of interleukin-33 and its receptor ST2 in the blood serum of children with pollinosis, depending on the age and severity of the disease, and then to determine the correlations of these parameters with various clinical and immunological parameters of seasonal allergic rhinitis.

Materials and methods

To participate in the study, we selected 42 patients aged 6–17 years with SAR who sought medical and consultative care at the Centre for Specialised Pediatric Care "M.V. Sklifosovsky Poltava Regional Clinical Hospital of the Poltava Regional Council" in 2022. The control group included 26 healthy children of the appropriate age without any allergic or chronic diseases in their medical history. This prospective cohort study was conducted at the Department of Pediatrics 2 of Poltava State Medical University (PSMU) and the Research Institute of Genetic and Immunological Bases of Pathology and Pharmacogenetics of PSMU. All patients met the following inclusion criteria: the presence of long-term characteristic clinical symptoms of the disease, which are based on the Allergic Rhinitis and its Impact on Asthma (ARIA) guidelines; the presence of specific diagnostic criteria, such as positive skin tests to weed, grass, and/or tree pollen (hypersensitivity reaction "++" or papule size over 3 mm). Some of the children also had concomitant diagnoses of asthma and atopic dermatitis at the time of the examination, which were verified in accordance with current diagnostic and treatment protocols and international guidelines. Thus, among the examined patients, 13 (31 %) children with SAR had concomitant asthma. Exclusion criteria were: congenital and acquired upper respiratory tract anomalies/disorders; systemic immunological disorders; treatment with systemic steroids; decompensated kidney, liver, or heart disease; age under 6 and over 18 years; participation in other studies; parental refusal to participate in the study. The study was conducted with the written informed consent of the parents, following the Declaration of Helsinki, and was approved by the Ethics and Bioethics Committee of PSMU.

The overall assessment of the severity of nasal and non-nasal symptoms of AR was determined using a visual analogue scale (VAS), which is also an important tool in patient management, rational selection of stepwise pharmacotherapy and achievement of adequate control throughout the disease. The scale reflected clinical manifestations of AR from their complete absence (0 mm) to mild — 0–30 mm, moderate — 31–70 mm and severe — 71–100 mm [16].

Skin prick testing was performed following generally accepted standards during remission. A standard pollen panel (Immunolog LLC, Vinnytsia) was used for the study, which included the most common allergens of trees, weeds, and cereals. Prick testing was carried out in the absence of skin diseases in the acute stage, acute and chronic infectious diseases, tuberculosis; withdrawal of antihistamines at least ten days before, and topical steroid ointments at least 1 month before the study.

Quantitative measurement of interleukin-33 was performed by enzyme-linked immunosorbent assay (ELISA) using Human IL-33 ELISA Kit, a reagent kit for the quantitative determination of human interleukin-33 (Invitrogen by Thermo Fisher Scientific Inc., Vienna, Austria), according to the manufacturer's instructions. Serum level of ST2 was determined by ELISA using the Presage® ST2 Assay and Presage® ST2 Control Kit (Critical Diagnostics, San Diego, USA) according to the manufacturer's recommendations. The obtained concentrations of interleukin-33 and ST2 were calculated using LabLine-026 (LabLine, Austria), a microplate photometer for ELISA.

The results of clinical and laboratory studies were processed using a standard computer program for statistical analysis KyensLab Inc., version 6.0. The interpretation of descriptive statistics was performed with the determination of the mean value (M), the error of the mean value (m), and the standard deviation (δ). The statistical significance of the

differences in the results between the groups was determined using parametric and non-parametric methods of statistical analysis, depending on the normality of the distribution of the relevant values. In particular, to compare a series of data with a normal distribution, the Student's t-test was used, while in the case of non-normal distribution, the Mann-Whitney U-test was applied. To assess the correlations between IL-33, ST2 levels, and other clinical and laboratory parameters, linear correlation analysis and Spearman's rank correlation were performed. Statistical significance was set at the level of $p < 0.05$.

Results and discussion

A detailed analysis of clinical and immunological parameters was carried out among 42 children with seasonal manifestations of upper respiratory tract allergy. For population control, the study included 26 practically healthy children without any manifestations of atopy in the medical history and at the time of examination. The assessment of age and gender characteristics did not reveal any significant differences between patients with seasonal allergic rhinitis and healthy participants ($p > 0.05$). The average age of the examined children with SAR was 11.90 ± 0.50 years, with a greater prevalence in the age structure of those of secondary school age (45.24 %), while younger and older children accounted for 28.57 and 26.19 %, respectively. The gender distribution was typical, with almost twice as many males (64.29 vs. 35.71 %, respectively), which once again confirms the numerous literature data on the predominant development of allergic diseases among boys. The proportion of boys and girls in the control group was equal and amounted to 50 % for each sex. The analysis of the data obtained demonstrated early (up to 6 years) manifestation of the disease in more than half of patients (57.14 %) with SAR. The average duration of rhinitis was 5.05 ± 0.41 years and had no significant effect on the severity of symptoms, which is consistent with the results of some foreign researchers [17].

A study on the spectrum of associated allergic diseases showed that one in three patients had concomitant allergic conjunctivitis (35.71 %) and asthma (30.95 %). These results are consistent with recent studies by both local and foreign authors, which demonstrate that children with AR have a triple risk of developing asthma [18, 19]. Atopic dermatitis accompanied only two (4.76 %) patients with SAR.

When assessing the severity of AR symptoms using a VAS, we found a moderate severity in half of the subjects (52.38 %), and a severe disease in every fourth (26.19 %) patient (Fig. 1). Mild severity was recorded only in one in five patients (21.43 %) and was characterised by the absence of a significant impact on the child's health, performance and leisure. At the same time, patients with moderate and severe forms, along with classic nasal symptoms, reported impaired daily activity, sleep, rest, and deterioration in the quality of education, and in some cases required restriction of their usual physical activity.

In our opinion, assessing the severity of AR should be an integral diagnostic element in the management of patients with seasonal allergic rhinitis, as regardless of the severity of the disease, patients require a specialised personalised approach to adequately control its course and prevent complications. Therefore, a visual analogue scale is an important and accessible tool that should be used in the daily practice of an allergist by the international ARIA guidelines. In addition, it is worth remembering the importance of the influence of circadian rhythms on the severity of SAR symptoms, as most patients report an increase in symptoms at night and in the morning, which may be due to the active release of proinflammatory mediators IL-4, IL-5, IL-13, including interleukin-33 [20]. In this regard, there is a need to further investigate the role of immunopathogenetic mechanisms of respiratory allergy depending on molecular circadian rhythms [21, 22].

The study of the sensitisation spectrum provides important information about allergens that cause respiratory allergy symptoms and, accordingly, reveals patterns of atopic reactivity development. When analysing the structure of sensitisation to pollen allergens, we found that the highest proportion of children (92.86 %) were hypersensitive to weeds, which once again confirms the regional peculiarities of the prevalence of sensitisation and the data from local monitoring of pollen concentrations in the air [23, 24]. In addition, more than half of the patients (66.67 %) had sensitisation to the cereal family. In 45.24 % of cases, seasonal allergy manifestations were provoked by tree pollen. In the presence of multivalent autosensitisation, a combination of sensitivity to weeds and cereals was more common.

The study on the frequency of hypersensitivity to one (monosensitivity) or several (polysensitivity) groups of allergens is important both diagnostically for the selection of the most frequent causative allergens and therapeutically, at first for correct and timely allergen-specific treatment. Therefore, we analysed the prevalence of sensitisation to aeroallergens among patients with SAR, which revealed the presence of hypersensitivity to one group of pollen allergens

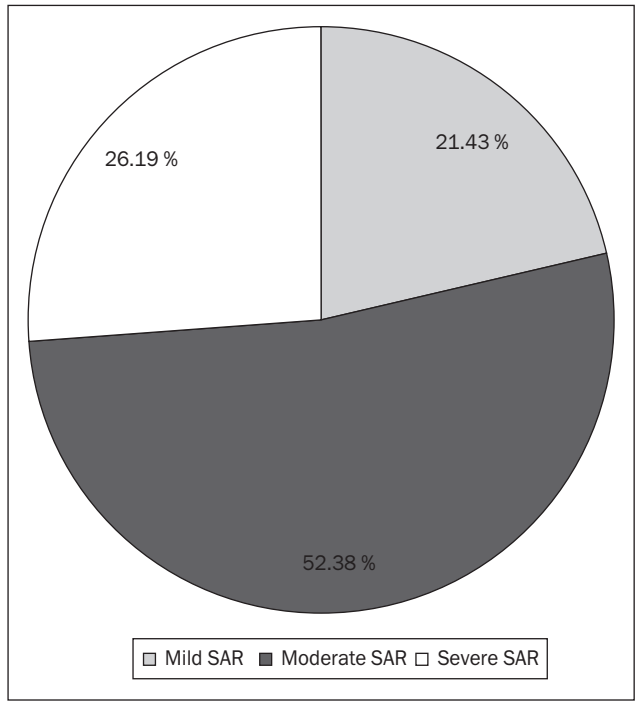

Figure 1. Severity of seasonal allergic rhinitis according to a visual analogue scale

in almost every fourth case (23.8 %), to two groups in almost half (47.62 %) of patients and to three groups in about one in three children (28.57 %). A high percentage of polysensitised patients confirms higher prevalence of hypersensitivity to several structurally different allergens and is of significant clinical and epidemiological importance in the management of people with respiratory allergy [25]. At the same time, the identification of real polyallergy remains an important task, which is confirmed by the presence of clinical symptoms of the disease that correlate with positive results of skin allergy testing and levels of specific immunoglobulins E (IgE).

The mechanisms of allergic inflammation in the human body are a complex cascade of immunological Th2-mediated reactions involving a large number of cytokine-chemokine mediators, whose role is still being studied. The hyperproduction of proinflammatory cytokines such as interleukin-4, -5, -13, immunoglobulin E, and eosinophils remains well known in the development of allergic inflammation. Recently, there has been a growing number of studies in the scientometric space on the role of IL-33 and its receptor ST2 in the pathogenetic mechanisms of various allergic pathologies. Therefore, one of the objectives of our study was an in-depth analysis of immunological status parameters to determine the levels of IgE, IL-33, and ST2 in patients with seasonal allergic rhinitis.

Since allergic rhinitis is realised by IgE-mediated allergic reactions, one of the primary steps at the laboratory stage was to determine serum IgE concentration in children with SAR. Thus, almost all patients with respiratory manifestations of allergy had elevated levels of IgE whose mean value was 549.88 ± 62.85 IU/ml (min 103.00, max 1,628.00 IU/ml), which confirms its role in the pathogenesis of allergic inflammation. Further study of the immunological profile (Fig. 2) revealed a statistically significant increase in the ratio of IL-33/ST2 levels compared to those in the group of healthy children ($p < 0.001$). Thus, the mean value of IL-33 (21.42 ± 0.89 pg/ml) and ST2 (38.560 ± 2.280 ng/ml) in the main group was twice as high as in the control group (10.16 ± 0.43 pg/ml and 19.69 ± 0.67 ng/ml, respectively). These results are in line with recently published foreign studies that showed high levels of serum IL-33 in children with AR compared to the control group, but according to their data, the average ST2 levels did not differ significantly among the respondents [26, 27].

In a comparative assessment of immunological parameters in sick children depending on sensitisation (mono- or polysensitisation) to different groups of pollen allergens (trees, cereals and weeds), we found no statistically significant differences between the levels of cytokine and its receptor. Thus, the concentrations of IL-33 and ST2 in monosensitised patients were 20.84 ± 1.27 pg/ml and 36.72 ± 2.77 ng/ml, respectively, and did not differ statistically from those in polysensitised patients. In children sensitised to groups 2 and 3 allergens, the average values of IL-33 and ST2 were 22.03 ± 1.42 pg/ml, 39.46 ± 3.18 ng/ml, and 20.87 ± 1.85 pg/ml, 38.61 ± 5.75 ng/ml, respectively ($p > 0.05$). However, to date, there have been no similar studies comparing the levels of interleukin-33 and ST2 depending on the number of causative pollen allergens. In our opinion, this may be an important aspect of laboratory diagnosis for the verification of allergic inflammation, even with minimal sensitisation to a single allergen.

In addition, we conducted a detailed analysis of IL-33 and ST2 serum levels, taking into account the age distribution of children. When comparing the mean values of these immunological markers shown in Table 1, it was noted that there were no age-dependent changes among patients with SAR in different age groups. The obtained results are fully comparable with the similar data of the control group ($p > 0.05$) and exclude the possibility of the age aspect influence on the development of immunologically mediated allergic inflammation.

Also, the study found out the peculiarities of the levels of immunological parameters IL-33, ST2 depending on the severity of seasonal allergic rhinitis, assessed by a VAS (Table 2). IL-33 content in the group of severe SAR (27.57 ± 1.90 pg/ml; $p < 0.01$) was statistically significantly (1.3 and 1.6 times, respectively) higher than that of patients with moderate (21.18 ± 1.02 pg/ml; $p < 0.01$) and mild (16.85 ± 0.78 pg/ml; $p < 0.001$) course of the disease. The analysis of serum ST2 concentration showed a statistically higher probability of the results obtained in patients with severe rhinitis (53.29 ± 5.15 ng/ml) compared to the group with mild severity (30.81 ± 3.22 ng/ml) of SAR ($p < 0.01$). The data obtained are consistent with other studies [28] and once again confirm the involvement of IL-33 in the induction of allergen-specific Th2 cell activation, which explains its regulatory role in the allergic immune response and an

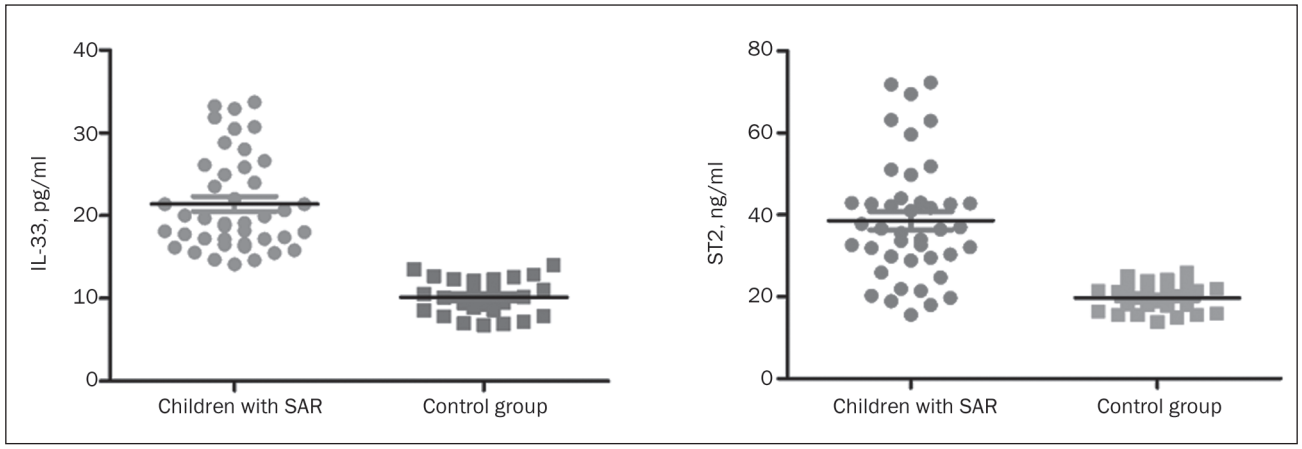

Figure 2. Comparative assessment of cytokine profile in the examined children

increase in serum concentration with increasing severity of SAR. Peng H.S. et al. also reported positive correlations between the severity of AR and IL-33 levels.

When comparing the correlation elements listed in Table 3, we found a direct strong correlation between the level of IL-33, its ST2 receptor ($\tau = 0.83$; $p < 0.001$), severity of nasal clinical symptoms ($\tau = 0.80$; $p < 0.01$) with a high degree of data reliability. The results obtained indicate the possibility of using these biomarkers to assess the severity of the disease and, subsequently, to predict the effectiveness of protocol treatment (including allergen-specific immunotherapy). In addition, the analysis of correlation structures revealed a statistically significant medium strength association between IL-33 and ST2 with a topical nasal ($\tau = 0.52$, $p < 0.001$ and $\tau = 0.38$, $p < 0.05$) and systemic eosinophilia ($\tau = 0.34$, $p < 0.05$ and $\tau = 0.31$, $p < 0.05$), which emphasises their diagnostic significance as criteria for allergic inflammation. Also, during the statistical processing of the results obtained, a direct medium strength relationship was found between the levels of IL-33 and IgE ($\tau = 0.41$; $p < 0.01$), which confirms the nature of IgE-mediated allergic reactions and determines the place of IL-33 in the algorithm for the diagnosis of patients with seasonal allergic rhinitis. The concentration of IL-33 and ST2 had an inverse moderate correlation with the level of IgA ($\tau = -0.36$ and $\tau = -0.37$, respectively, at $p < 0.05$), which logically explains a decrease in the latter in case of prolonged persistent allergic inflammation among patients with nasal manifestations of seasonal allergy. The results of the study did not demonstrate statistically significant correlations ($p > 0.05$) between the concentration of total IgE, the severity of SAR ($\tau = 0.21$), the systemic level of eosinophilic inflammation in the blood ($\tau = 0.18$) and serum values of ST2 ($\tau = 0.12$) and immunoglobulin A ($\tau = -0.25$).

The ARIA study is of particular interest in the study of multimorbid allergic diseases. It dates back to 1999 during a meeting of the regular session of the World Health Organization, where the interrelationships between these diseases were identified, based on which practical clinical guidelines for their treatment were developed. Due to numerous scientific papers reflecting a significant proportion of rhinitis comorbidity with other allergic diseases, primarily asthma, the concept of "one airway, one disease" is gaining more and more

Table 1. The levels of immunological parameters IL-33, ST2 and their correlations with the age of the examined patients (n = 42), M ± m

Cytokines	Primary school age (n = 12)	Secondary school age (n = 19)	High school age (n = 11)	p
IL-33, pg/ml	20.92 ± 1.34	21.47 ± 1.43	21.87 ± 2.01	> 0.05
ST2, ng/ml	37.03 ± 4.02	37.91 ± 3.29	41.37 ± 5.21	

Table 2. The levels of immunological parameters IL-33/ST2 and their correlations with the severity of SAR in the examined patients (n = 42)

Cytokines	Mild SAR (n = 11)	Moderate SAR (n = 22)	Severe SAR (n = 9)
IL-33, pg/ml (M ± m)	16.85 ± 0.78	21.18 ± 1.02*	27.57 ± 1.90$^{\#\#\Delta}$
ST2, ng/ml (M ± m)	30.81 ± 3.22	36.42 ± 2.63	53.29 ± 5.15$^{\#}$
Correlation coefficient τ	0.65	0.76	0.80
Probability value	p < 0.05	p < 0.01	p < 0.05

Notes: significance of indicators between patients: * — with moderate and mild SAR, p < 0.01; # — with severe and mild SAR, p < 0.01; ## — with severe and mild SAR, p < 0.001; Δ — with severe and moderate SAR, p < 0.01.

Table 3. Correlation between IL-33 levels and clinical and immunological determinants of seasonal allergic rhinitis in the examined patients (n = 42)

	IL-33, pg/ml	ST2, ng/ml	IgE, IU/ml	IgA, g/l	Blood eosinophils, %	Nasal eosinophils, %	Severity according to VAS
Severity according to VAS	0.80***	0.73***	0.21$^{\Delta}$	–0.46**	0.59***	0.65***	
Nasal eosinophils, %	0.52***	0.38*	0.37*	–0.37*	0.47**		
Blood eosinophils, %	0.34*	0.31*	0.18$^{\Delta}$	–0.43**			
IgA, g/l	–0.36*	–0.37*	–0.25				
IgE, IU/ml	0.41**	0.12$^{\Delta}$					
ST2, ng/ml	0.83***						
IL-33, pg/ml							

Notes: * — p < 0.05; ** — p < 0.01; *** — p < 0.001; Δ — p > 0.05.

Table 4. Comparative characteristics of cytokines in children with seasonal allergic rhinitis, taking into account comorbidity, M ± m

Cytokines	Isolated SAR (n = 29)	SAR and asthma (n = 13)	Control group (n = 26)
IL-33, pg/ml	18.16 ± 0.50*	28.68 ± 1.08#Δ	10.16 ± 0.43
ST2, ng/ml	32.26 ± 1.81*	52.64 ± 4.06#Δ	22.70 ± 1.39

Notes: significance of indicators between patients: * — with isolated SAR and control group, p < 0.001; # — with SAR/asthma and isolated SAR, p < 0.001; Δ — with SAR/asthma and control group, p < 0.001.

recognition as the upper and lower respiratory tracts are part of the same system. This fact emphasised the importance of timely verification of allergic rhinitis to prevent further evolution of the atopic march and development of bronchial hyperreactivity. However, according to the latest ARIA-MeDALL (MEchanisms of the Development of ALLergy, Bousquet J. et al., 2023), because of new views on the interpretation of polymorbidity and polysensitisation, there is a need to revise this concept for identifying and separating new phenotypes of allergic rhinitis such as isolated allergic rhinitis, rhinitis associated with asthma and severe allergic "asthmatic" phenotype, which combines these two diseases with allergic conjunctivitis. Thus, to analyse the most common clinical phenotypes of SAR, the subjects were divided into a subgroup with isolated rhinitis (n = 29) and SAR in combination with asthma (n = 13), which accounted for one third (30.95 %) of the total number of subjects in the main group.

In the presence of comorbidities among patients with SAR particularly concomitant asthma, there was a tendency to a statistically significant prevalence of severe disease (38.46 vs. 13.79 % in isolated AR; $\chi^2 = 1.94$; $p = 0.063$). Also, in patients with multimorbidity, the proportion of those with moderate disease was relatively higher (61.54 %), but it was not significant ($p > 0.05$). A mild course of the disease was recorded exclusively in patients with SAR ($\chi^2 = 4.86$; $p = 0.027$).

Comparison of laboratory parameters in children with SAR and concomitant asthma revealed higher levels of eosinophils, total IgE and IL-33/ST2. In particular, the average concentrations of interleukin-33 (28.68 ± 1.08 pg/ml) and ST2 (52.64 ± 4.06 ng/ml) in this subgroup were statistically significantly ($p < 0.001$) higher — by 1.6 and 2.8 times for IL-33, 1.6 and 2.3 times for ST2, compared to similar data in patients with isolated allergic rhinitis and healthy children (Table 4).

Comparative characteristics of the correlation between the levels of immunological parameters IL-33 and ST2 demonstrate the strongest statistically significant relationship in the subgroup of children with SAR and asthma ($\tau = 0.763$; $p < 0.01$), which, in our opinion, may be evidence of a more severe course of this disease phenotype. In the comparison groups, a direct medium strength dependence was found between the levels of these indicators, which explains the pathogenetic mechanisms of allergic inflammation through the corresponding correlation of IL-33 with its receptor.

Conclusions

1. The analysis of serum concentrations of IL-33 and ST2 showed statistically higher levels in the group with severe SAR and revealed a direct medium strength correlation between IL-33 and ST2 levels in patients with mild SAR ($\tau = 0.65$; $p < 0.05$) and a strong correlation in those with moderate ($\tau = 0.76$; $p < 0.01$) and severe ($\tau = 0.80$; $p < 0.05$) disease, which confirms the role of the IL-33/ST2 axis in inducing allergen-specific Th2 cell activation in children with SAR.

2. Comparison of the mean values of the studied immunological profile parameters, namely IL-33 and ST2, did not reveal any significant dependence on age, mono- or polysensitisation, or duration of allergic history.

3. Phenotypically, comorbid patients with seasonal allergic rhinitis and asthma are characterised by a tendency to statistically significant prevalence of a more severe course of the disease, with correspondingly higher values of IL-33 and its receptor ST2 compared to children with isolated SAR, which confirms their role as immunological markers indicating the severity of the immune response to allergic inflammation in pediatric patients.

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References

1. Bousquet J, Agache I, Berger U, et al. Differences in Reporting the Ragweed Pollen Season Using Google Trends across 15 Countries. Int Arch Allergy Immunol. 2018;176(3-4):181-188. doi:10.1159/000488391.

2. Kaidashev I, Morokhovets H, Rodinkova V, DuBuske L, Bousquet J. Assessment of Google Trends terms reporting allergies and the grass pollen season in Ukraine. World Allergy Organ J. 2020 Sep 30;13(10):100465. doi:10.1016/j.waojou.2020.100465.

3. Singh AB, Kumar P. Climate change and allergic diseases: An overview. Front Allergy. 2022 Oct 13;3:964987. doi:10.3389/falgy.2022.964987.

4. Marushko YuV, Moskovenko OD. Characteristics of sensitization and experience of using desloratadine in the treatment of seasonal allergic rhinitis in children. Zdorov'e rebenka. 2019;14(7):54-57. (in Ukrainian).

5. Lau S, Matricardi PM, Wahn U, Lee YA, Keil T. Allergy and atopy from infancy to adulthood: Messages from the German birth cohort MAS. Ann Allergy Asthma Immunol. 2019 Jan;122(1):25-32. doi:10.1016/j.anai.2018.05.012.

6. Zhang Y, Lan F, Zhang L. Advances and highlights in allergic rhinitis. Allergy. 2021 Nov;76(11):3383-3389. doi:10.1111/all.15044.

7. Yamasaki A, Burks CA, Bhattacharyya N. Cognitive and Quality of Life-Related Burdens of Illness in Pediatric Allergic Airway Disease. Otolaryngol Head Neck Surg. 2020 Apr;162(4):566-571. doi:10.1177/0194599820908202.

8. Bousquet J, Melén E, Haahtela T, et al. Rhinitis associated with asthma is distinct from rhinitis alone: The ARIA-MeDALL hypothesis. Allergy. 2023 May;78(5):1169-1203. doi:10.1111/all.15679.

9. Paiva Ferreira LKD, Paiva Ferreira LAM, Monteiro TM, Bezerra GC, Bernardo LR, Piuvezam MR. Combined allergic rhinitis and asthma syndrome (CARAS). Int Immunopharmacol. 2019 Sep;74:105718. doi:10.1016/j.intimp.2019.105718.

10. Wang M, Zhao N, Wang C, Jin ZB, Zhang L. Immunomodulatory properties of mesenchymal stem cells: A potential therapeutic strategy for allergic rhinitis. Allergy. 2023 Jun;78(6):1425-1440. doi:10.1111/all.15729.

11. Yemchenko YaO, Ischeikin KYe, Kaidashev IP, Izmailova OV. Il-33 as a biomarker of inflammatory activity in psoriasis patients with concomitant obesity. Svit medytsyny ta biolohii. 2021;(76):37-42. doi:10.26724/2079-8334-2021-2-76-37-42. (in Ukrainian).

12. Ramezani F, Babaie F, Aslani S, et al. The Role of the IL-33/ST2 Immune Pathway in Autoimmunity: New Insights and Perspectives. Immunol Invest. 2022 May;51(4):1060-1086. doi:10.1080/08820139.2021.1878212.

13. Sun J, Xia Y, Zhang D, Yu Z, Ning Y, Tan Z. Knowledge mapping of interleukin-33: a bibliometric study. Am J Transl Res. 2023 Feb 15;15(2):914-931.

14. Peng HS, Zhu XH. Research progress on the role of IL-33/ST2 axis in pathogenesis of allergic rhinitis. Lin Chuang Er Bi Yan Hou Tou Jing Wai Ke Za Zhi. 2019 Oct;33(10):910-914. doi:10.13201/j.issn.1001-1781.2019.10.003. (in Chinese).

15. Han X, Krempski JW, Nadeau K. Advances and novel developments in mechanisms of allergic inflammation. Allergy. 2020 Dec;75(12):3100-3111. doi:10.1111/all.14632.

16. Klimek L, Bergmann KC, Biedermann T, et al. Visual analogue scales (VAS): Measuring instruments for the documentation of symptoms and therapy monitoring in cases of allergic rhinitis in everyday health care: Position Paper of the German Society of Allergology (AeDA) and the German Society of Allergy and Clinical Immunology (DGAKI), ENT Section, in collaboration with the working group on Clinical Immunology, Allergology and Environmental Medicine of the German Society of Oto-rhinolaryngology, Head and Neck Surgery (DGHNOKHC). Allergo J Int. 2017;26(1):16-24. doi:10.1007/s40629-016-0006-7.

17. Bousquet PJ, Devillier P, Tadmouri A, Mesbah K, Demoly P, Bousquet J. Clinical relevance of cluster analysis in phenotyping allergic rhinitis in a real-life study. Int Arch Allergy Immunol. 2015;166(3):231-240. doi:10.1159/000381339.

18. Marushko YuV, Abaturov OE, Beketova GV, Berezenko VS, Pochynok TV, Churylina AV. New views on the diagnosis and treatment of asthma in children. Zdorov'e rebenka. 2022;17(2):54-60. doi:10.22141/2224-0551.17.2.2022.1503. (in Ukrainian).

19. Yan Z, Liu L, Jiao L, Wen X, Liu J, Wang N. Bioinformatics Analysis and Identification of Underlying Biomarkers Potentially Linking Allergic Rhinitis and Asthma. Med Sci Monit. 2020 May 27;26:e924934. doi:10.12659/MSM.924934.

20. Christ P, Sowa AS, Froy O, Lorentz A. The Circadian Clock Drives Mast Cell Functions in Allergic Reactions. Front Immunol. 2018 Jul 6;9:1526. doi:10.3389/fimmu.2018.01526.

21. Kryuchko TO, Tkachenko OYa, Shcherbak VV, Kolenko IO, Bubyr LM. The current state of the problem with the diagnosis and treatment of allergic rhinitis in pediatric practice. Zdorov'e rebenka. 2021;16(5):375-383. doi:10.22141/2224-0551.16.5.2021.239718. (in Ukrainian).

22. Nakao A. Clockwork allergy: How the circadian clock underpins allergic reactions. J Allergy Clin Immunol. 2018 Oct;142(4):1021-1031. doi:10.1016/j.jaci.2018.08.007.

23. Shcherbak VV, Bubyr LM, Kuzmenko NV, Oliinichenko MO, Klymko DM. Dynamic changes in ethiological structure of pollen sensitization in children with seasonal allergic rhinitis. Actual problems of modern medicine: Bulletin of Ukrainian Medical Stomatological Academy. 2022;22(3-4):100-104. doi:10.31718/2077-1096.22.3.4.100. (in Ukrainian).

24. Rodinkova V, Palamarchuk O, Yasniuk M, et al. P020 Global warming extends the ragweed season and duration of elevated ragweed pollen levels in Ukraine. Annals of Allergy, Asthma and Immunology. 2021;127(5):S22. doi:10.1016/j.anai.2021.08.065.

25. Raciborski F, Bousquet J, Bousqet J, et al. Dissociating polysensitization and multimorbidity in children and adults from a Polish general population cohort. Clin Transl Allergy. 2019 Feb 11;9:4. doi:10.1186/s13601-019-0246-y.

26. Qiao Y, Chen J. Serum levels of IL-31, IL-33 and ST2 in allergic rhinitis of children in China. Cell Mol Biol (Noisy-le-grand). 2018 Sep 30;64(12):52-55.

27. Glück J, Rymarczyk B, Rogala B. Serum IL-33 but not ST2 level is elevated in intermittent allergic rhinitis and is a marker of the disease severity. Inflamm Res. 2012 Jun;61(6):547-50. doi:10.1007/s00011-012-0443-9.

28. Zhu K, Xia C, Chen J, et al. Serum Soluble ST2 Correlated with Symptom Severity and Clinical Response of Sublingual Immunotherapy for House Dust Mite-Induced Allergic Rhinitis Patients. Mediators Inflamm. 2021 May 30;2021:5576596. doi:10.1155/2021/5576596.

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Information about authors

Viktoriia Shcherbak, Assistant of the Department of Pediatrics 2, Poltava State Medical University, Poltava, Ukraine; e-mail: v.shcherbak@pdmu.edu.ua; https://orcid.org/0000-0002-9268-6454

Tetyana Kryuchko, MD, PhD, Professor, Head of the Department of Pediatrics 2, Poltava State Medical University, Poltava, Ukraine; e-mail: drkryuchko@gmail.com; fax: +380(532)606491; phone: +380(50)3277023; https://orcid.org/0000-0002-5034-4181

L.M. Bubyr, PhD, Assistant of the Department of Pediatrics 2, Poltava State Medical University, Poltava, Ukraine; e-mail: lyudmila.bubyr@ukr.net; https://orcid.org/0000-0001-9736-3593

O.V. Izmailova, Senior Researcher of the Research Institute of Genetic and Immunological Bases of Pathology and Pharmacogenetics, Poltava State Medical University, Poltava, Ukraine; e-mail: o.izmailova@pdmu.edu.ua; https://orcid.org/0000-0003-4770-3494

O.A. Poda, PhD, Associate Professor of the Department of Pediatrics 2, Poltava State Medical University, Poltava, Ukraine; e-mail: olha.poda@gmail.com; https://orcid.org/0000-0002-2314-8628

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Щербак В.В., Крючко Т.О., Бубир Л.М., Ізмайлова О.В., Пода О.А.
Полтавський державний медичний університет, м. Полтава, Україна

Зміни імунологічних біомаркерів у педіатричних пацієнтів із сезонним алергічним ринітом

Резюме. Актуальність. Існує постійна тенденція до зростання частоти сезонного алергічного риніту не лише серед дорослого населення, але й у дитячій популяції. Значний відсоток пацієнтів мають середній та тяжкий перебіг захворювання, що призводить до суттєвого погіршення якості їхнього життя. Оскільки патогенез алергічного риніту представлений складним каскадом імунологічних реакцій із залученням низки цитокінів, що беруть участь у розвитку алергічного запалення верхніх дихальних шляхів, пріоритетним напрямком нашої роботи стало поглиблене вивчення патогенетичних механізмів цього захворювання. **Мета:** дослідити рівні інтерлейкіну-33 (ІЛ-33) та його рецептора ST2 у сироватці крові дітей із полінозом залежно від віку та тяжкості перебігу захворювання з наступним визначенням кореляцій цих показників із різними клініко-імунологічними параметрами сезонного алергічного риніту. **Матеріали та методи.** У дослідженні взяли участь 42 хворі віком 6–17 років із сезонним алергічним ринітом та 26 здорових дітей, які ввійшли в групу контролю. Ступень тяжкості назальних і екстраназальних симптомів алергічного риніту оцінювали за допомогою візуальної аналогової шкали. Шкірне прик-тестування проводилося відповідно до загальноприйнятих стандартів у період ремісії з використанням стандартної пилкової панелі. Кількісне вимірювання сироваткових показників інтерлейкіну-33 та ST2 здійснювали методом твердофазного імуноферментного аналізу. Обов'язковою умовою проведення дослідження була наявність письмової інформованої згоди батьків. **Результати.** При дослідженні сироваткових рівнів ІЛ-33 та ST2 показано їх статистично вищі значення в групі дітей із тяжким перебігом сезонного алергічного риніту й встановлено пряму кореляцію середньої сили між умістом ІЛ-33 та ST2 у пацієнтів із легким захворюванням ($\tau = 0,65$; $p < 0,05$) і сильну залежність в осіб із середнім ($\tau = 0,76$; $p < 0,01$) та тяжким ($\tau = 0,80$; $p < 0,05$) перебігом. При порівнянні середньостатистичних величин ІЛ-33 та ST2 не було виявлено вірогідних змін залежно від віку та наявності моно- чи полісенсибілізації серед пацієнтів з алергічним ринітом. В осіб із сезонним алергічним ринітом та супутньою бронхіальною астмою відмічалася тенденція до статистично значимого превалювання більш тяжкого перебігу захворювання з вищими рівнями ІЛ-33 та ST2 порівняно з хворими з ізольованим сезонним алергічним ринітом. **Висновки.** Отримані результати імунологічного дослідження дозволили підкреслити діагностичну значимість ІЛ-33 та його рецептора ST2 як потенційних біомаркерів розвитку алергічного запалення в пацієнтів педіатричного профілю з сезонними проявами риніту.

Ключові слова: алергічний риніт; біомаркер; цитокіни; бронхіальна астма; діти; алергія