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IMMUNE STATUS OF PATIENTS WITH CHRONIC MICROBIAL ECZEMA AND ELEVATED IGG4 LEVELS TO MOLD ALLERGENS

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According to the World Allergy Organization, 40 % of the global population has one or more allergic diseases and daily struggles with the fear of potential asthma attacks, complications, or even death from various allergic reactions. A clear dependence of the development of allergic diseases on the degree of air contamination by fungi and their spores has been established. Therefore, the study of specific immune reactions to antigens of mold and yeast-like fungi is extremely relevant. Overall, elevated levels of specific IgG4 were detected in 95.7 % of patients to at least one of the studied allergens. The frequency of elevated specific IgG4 levels to each of the studied mold allergens was as follows: *Mucor racemosus* – 65.2 %, *Candida albicans* – 54.3 %, *Aspergillus fumigatus* – 41.3 %, *Alternaria tenuis* – 34.8 %, *Penicillium notatum* – 6.5 %, *Cladosporium herbarum* – 4.3 %. For the etiological diagnosis of exacerbations in chronic microbial eczema and uncontrolled disease progression, the determination of specific fungal IgE and IgG is informative. This will clarify the nature of sensitization and enable adequate treatment for patients with chronic microbial eczema.

Key words: sensitization, mold fungi, microbial eczema, IgG4.

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ІМУННИЙ СТАТУС ПАЦІЄНТІВ ІЗ ХРОНІЧНОЮ МІКРОБНОЮ ЕКЗЕМОЮ ТА ПІДВИЩЕНИМ РІВНЕМ IGG4 ДО АЛЕРГЕНІВ ПЛІСНЯВИХ ГРИБІВ

Відповідно до даних Всесвітньої організації алергії (World Allergy Organization), 40 % населення планети мають одну чи декілька алергічних хвороб та щоденно борються зі страхом можливого нападу астми або розвитком ускладнень, чи, навіть, смерті від тих чи інших алергічних реакцій. Встановлено чітку залежність розвитку алергічних захворювань від ступеня обсіменіння повітря грибами та їх спорами. Тому надзвичайно актуальне дослідження специфічних імунних реакцій на антигени пліснявих та дріжджоподібних грибів. В цілому підвищений рівень специфічного IgG4 виявлено у 95,7 % хворих до хоча б одного з досліджуваних алергенів. Частота зустрічаємості підвищеного рівню специфічного IgG4 до кожного з досліджуваних нами алергенів пліснявих грибів, отримані результати: до *Mucor racemosus* – 65,2 %, до *Candida albicans* – 54,3 %, до *Aspergillus fumigatus* – 41,3 %, до *Alternaria tenuis* – 34,8 %, до *Penicillium notatum* – 6,5 %, до *Cladosporium herbarum* – 4,3 %. Для етіологічної діагностики загострення хронічної мікробної екземи та неконтрольованого перебігу захворювання інформативним є визначення специфічних грибкових IgE та IgG, що дозволить уточнити характер сенсibilізації та провести адекватне лікування хворих на хронічну мікробну екзему.

Ключові слова: сенсibilізація, плісняві гриби, мікробна екзема, IgG4.

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According to the data from the World Allergy Organization (WAO), 40 % of the global population has one or more allergic diseases and daily struggles with the fear of potential asthma attacks, complications, or even death from various allergic reactions. Of particular concern to the scientific community and practical healthcare institutions is the rising prevalence of these indicators among the pediatric population. According to the results of the international ISAAC study (The International Study of Asthma and Allergies in Childhood), the prevalence of bronchial asthma among children aged 6-7 years was found to range from 4.1–5.6 % in Indonesia, Iran, and India to 23.3–32.1 % in Brazil, Australia, and certain European countries. Among children aged 13-14 years, the prevalence ranged from 2.1–4.4 % in Indonesia, China, and Georgia to 32.2 % in the United Kingdom. Similarly high rates were observed for allergic rhinitis (its prevalence ranged from 9.8 % in Albania to 44.5–66.6 % in Hong Kong, France, and Argentina) and atopic dermatitis (from 4.1 % in Albania and 11.9 % in Poland to 21.8 % in Finland) [1].

A clear dependence of the development of allergic diseases on the degree of air contamination by fungi and their spores has been established. Therefore, the study of specific immune reactions to antigens of mold and yeast-like fungi is extremely relevant. Fungi, along with other allergens, can cause mycogenic sensitization and allergy in humans [6]. Allergic skin diseases, in terms of severity and prevalence, are second only to bronchial asthma [9]. The problems of pathogenesis and treatment of eczema are the most relevant in modern medicine. According to various researchers, eczema accounts for 15 to 40 % of all skin diseases. Microbial eczema is an inflammatory skin disease (epidermis and dermis) of neuroallergic nature, the pathognomonic sign of which is serous (eczematous) wells, characterized by a recurrent and persistent

course without long remissions and evolutionary polymorphism. Accounting for 40-50 % of all allergic dermatoses, it is one of the most frequently encountered skin diseases in elderly and senile individuals [4].

The purpose of the study was to investigate the level of specific IgG4 to allergens of non-pathogenic and opportunistic mold fungi (*Mucor racemosus*, *Alternaria tenuis*, *Candida albicans*, *Penicillium notatum*, *Cladosporium herbarum*, *Aspergillus fumigatus*) in patients with chronic microbial eczema to improve the treatment of this pathology.

Materials and methods. The study examined the level of specific IgG4 in the serum of 86 patients with a confirmed diagnosis of microbial eczema, who had been ill for at least 1 year and were undergoing treatment in the day hospital of the Dermatovenereological Center of the M.V. Sklifosovsky Poltava Regional Clinical Hospital of the Poltava Regional Council from 2019 to 2024. The clinical picture was characterized by the typical spread of the skin-pathological process: asymmetric lesions on the skin of the shins, dorsal surfaces of the hands, lateral surfaces of the trunk, scalp, polymorphous rash (lesions with clear boundaries, in the form of microvesicles, pustules on a background of erythema, infiltration, purulent and yellowish crusts, scaling), severe itching, acute inflammatory skin reaction, severe pain on palpation; persistence of clinical course, and frequent disease relapses [7].

The level of specific IgG4 in serum was determined using the enzyme-linked immunosorbent assay (ELISA) method with monoclonal antibodies against human IgG4 labeled with peroxidase, produced by PJSC "DIAPROF-MED" (Ukraine); cellulose discs coated with allergens produced by Dr. Fooke (Germany). The optical density of the samples was measured using the "Stat fax 303 plus" ELISA analyzer (USA).

The selection of fungal species for the study was based on an analysis of literature and statistical data, according to which we used the six most common fungal allergens: *Mucor racemosus*, *Alternaria tenuis*, *Candida albicans*, *Penicillium notatum*, *Cladosporium herbarum*, *Aspergillus fumigatus*. Statistical analysis was performed using the STATISTICA 6.0 statistical package (Stat Soft, USA).

Results of the study and their discussion. The study included 86 patients aged 35 to 65 years with a diagnosis of chronic microbial eczema, established based on the clinical picture. Of these, 62 % had microbial eczema for 1–3 years, 27.5 % for 4–7 years, and 10.5 % for more than 10 years. Additionally, 37 % of the patients lived in the private sector of rural areas, 8.7 % were urban residents but lived in the private sector, and 54.3 % of the patients resided in urban multi-story buildings. The diagnostic value of specific IgG4 is determined by its level among healthy individuals, which in our study averaged 0.200 optical density (OD) units. A specific IgG4 level of 0.250–0.300 OD units was considered doubtful ("gray zone"), and a level above 0.300 OD units was defined as elevated. Overall, elevated levels of specific IgG4 were detected in 95.7 % of patients to at least one of the studied allergens (Table 1).

Table 1

Mean level of specific IgG4 in the examined patient group

Concentration of Specific IgG4 to Allergens (%):						
Level of sIgE	Mukor racemosus	Alternaria tenuis	Candida albicans	Penicillium notatum	Cladosporium herbarum	Aspergillus fumigatus
Normal <0.200	19.6	45.8	22.9	91.3	95.7	39.1
Doubtful 0.250–0.300	15.2	19.4	22.8	4.2	--	19.6
High >0.300	65.2	34.8	54.3	6.5	4.3	41.3

Regarding the frequency of elevated specific IgG4 levels to each of the studied mold allergens, the following results were obtained: *Mucor racemosus* – 65.2 %, *Candida albicans* – 54.3 %, *Aspergillus fumigatus* – 41.3 %, *Alternaria tenuis* – 34.8 %, *Penicillium notatum* – 6.5 %, *Cladosporium herbarum* – 4.3 %.

Among the group of patients with elevated specific IgG4 levels Simultaneous allergy to two or more allergens was detected in 71.8 % of the examined patients.

Of these: 43.3 % of the total patients had a combined allergy to two allergens, 17.4 % – to three allergens, 10.5 % – to four allergens simultaneously.

The most frequent combined allergies to mold allergens were: *M. racemosus* and *C. albicans* – 27.3 % of total combined sensitization cases, *A. fumigatus* and *M. racemosus* – 25 %, *M. racemosus* and *A. tenuis* – 20.5 %, *A. fumigatus* and *A. tenuis* – 15.9 % of cases.

Of the total number of patients residing in the private sector 60.9 % had elevated levels of specific IgG4 to *Candida albicans*, 47.5 % to *Mucor racemosus*, 43.5 % to *Aspergillus fumigatus*, 34.7 % to *Alternaria tenuis*.

From the group of patients with established elevated levels of specific IgG4 to the allergen of the fungus *Penicillium notatum*: No elevated levels of specific IgG4 only to the allergen of the fungus

Penicillium notatum were detected. No elevated levels of specific IgG4 to the allergen of the fungus *Penicillium notatum* in combination with one of the studied fungi were identified.

In the study of this patient group: 33.3 % of cases showed a combination of elevated specific IgG4 levels to the allergen of the fungus *Penicillium notatum* simultaneously with elevated levels to the allergens of fungi *Aspergillus fumigatus* and *Mucor racemosus*. A combination of elevated specific IgG4 levels to the allergen of the fungus *Penicillium notatum* and three other studied mold allergens was detected in 66.7 % of patients. In all cases of sensitivity to the allergen of the fungus *Penicillium notatum*. Simultaneous sensitization to the allergen of the fungus *Mucor racemosus* was observed.

In the study of the patient group with established elevated levels of specific IgG4 to the allergen of the fungus *Aspergillus fumigatus*, it was found that 21 % of patients had elevated levels of specific IgG4 only to the allergen of the fungus *Aspergillus fumigatus*. In 31.6 % of patients, elevated levels of specific IgG4 to the allergen of the fungus *Aspergillus fumigatus* were observed simultaneously with elevated levels of specific IgG4 to one of the studied fungal allergens. The same proportion (31.6 %) was observed in the group of patients with elevated levels of specific IgG4 to the allergen of the fungus *Aspergillus fumigatus* and simultaneous elevation of specific IgG4 levels to two of the studied fungal allergens. In 15.8 % of patients with established elevated levels of specific IgG4 to the allergen of the fungus *Aspergillus fumigatus*, elevated levels of specific IgG4 to three of the studied fungal allergens were simultaneously detected. Most frequently, simultaneous sensitivity to the allergen of the fungus *Aspergillus fumigatus* and sensitivity to the allergen of the fungus *Mucor racemosus* was observed in 57.9 % of patients.

In the study of the patient group with established elevated levels of specific IgG4 to the allergen of the fungus *Mucor racemosus*.

Sensitization specifically to this fungal species and none of the other studied allergens was detected quite rarely – in 6.7 % of patients. Elevated levels of specific IgG4 to the allergen of the fungus *Mucor racemosus* and simultaneously elevated levels of specific IgG4 to one of the studied fungal allergens were detected in 53.3 % of patients. Elevated levels of specific IgG4 to the allergen of the fungus *Mucor racemosus* and simultaneous elevation of specific IgG4 levels to two of the studied fungal allergens were detected in 26.7 % of patients, while elevation of specific IgG4 levels to three of the studied fungal allergens was established in 13.3 % of patients. Most frequently, simultaneous sensitivity to the allergen of the fungus *Mucor racemosus* and the allergen of the fungus *Candida albicans* was observed – in 56.7 % of examined patients.

In the analysis of the patient group with established elevated levels of specific IgG4 to the allergen of the fungus *Candida albicans*:

Elevation of specific IgG4 levels only to the allergen of the fungus *Candida albicans* was detected in 16 % of examined patients. Elevation of specific IgG4 levels to the allergen of the fungus *Candida albicans* and simultaneous elevation of specific IgG4 levels to one of the studied fungal allergens was detected in 48 % of examined patients. Elevated levels of specific IgG4 to the allergen of the fungus *Candida albicans* and simultaneous elevation of specific IgG4 levels to two of the studied fungal allergens were detected in 20 % of patients, while simultaneous elevation of specific IgG4 levels to the allergen of the fungus *Candida albicans* and three of the studied fungal allergens was established in 16 % of examined patients. In most cases, sensitivity to the allergen of the fungus *Candida albicans* and simultaneous sensitization to the allergens of the fungus *Mucor racemosus* were observed – in 68 % of examined patients.

From the studied group of patients with established elevated levels of specific IgG4 to the allergen of the fungus *Alternaria tenuis*:

Elevated levels of specific IgG4 only to the allergen of the fungus *Alternaria tenuis* were detected in 6.2 % of patients. Elevated levels of specific IgG4 to the allergen of the fungus *Alternaria tenuis* and simultaneously elevated levels of specific IgG4 to one of the studied fungal allergens were detected in 43.8 % of patients. Elevated levels of specific IgG4 to the allergen of the fungus *Alternaria tenuis* and simultaneously elevated levels to two of the studied fungal allergens were found in 25 % of patients, while simultaneously elevated levels of specific IgG4 to three of the studied fungal allergens were established in 25 % of examined patients. Most frequently, the combination of sensitivity to the allergen of the fungus *Alternaria tenuis* and sensitization to the allergen of the fungus *Mucor racemosus* was observed – in 62.5 % of examined patients. Equally common was the combination of sensitivity to the allergen of the fungus *Alternaria tenuis* and sensitization to the allergens of fungi *Aspergillus fumigatus* and *Candida albicans*.

In the study of the group of women with microbial eczema, who participated in the research and constituted 37 % of the total patient group, the following results were obtained:

Most frequently, elevated levels of specific IgG4 to the allergen of the fungus *Aspergillus fumigatus* were detected – in 52.9 % of female patients. Second in frequency was elevated levels of specific

IgG4 to the allergen of the fungus *Mucor racemosus* – in 47.1 % of patients. In 35.3 % of women with microbial eczema during the study, elevated levels of specific IgG4 to the allergen of the fungus *Alternaria tenuis* were established. The same percentage of elevated specific IgG4 levels was observed for the allergen of the fungus *Candida albicans* – 35.3 % of female patients. Elevated levels of specific IgG4 to the allergen of the fungus *Cladosporium herbarum* were detected in 29.4 % of women, while elevated levels to the allergen of the fungus *Penicillium notatum* were found least frequently – in 11.8 % of women with microbial eczema.

Additionally: No cases were identified where there was no elevation of specific IgG4 levels to at least one of the studied fungal allergens, meaning all women in the group exhibited sensitivity to at least one fungal allergen. Elevated levels to only one of the studied fungal allergens were detected in 29.4 % of female patients, most frequently to the allergen of *Aspergillus fumigatus*. Second, with equal frequency, was sensitivity to the allergen of *Candida albicans*. No mono-sensitivity to any other studied fungal allergens was detected. Simultaneous elevation of specific IgG4 levels to two of the studied fungal allergens was observed in 41.2 % of women with microbial eczema. Most frequently, this involved simultaneous elevation to the allergen of *Aspergillus fumigatus* either with *Mucor racemosus* or *Alternaria tenuis*. Simultaneous elevation of specific IgG4 levels to three of the studied fungal allergens was detected in 17.6 % of female patients. Notably, in these cases, as with dual sensitization, sensitivity to the allergens of *Aspergillus fumigatus*, *Mucor racemosus*, and *Alternaria tenuis* was consistently observed. Simultaneous elevation of specific IgG4 levels to four of the studied allergens was observed in 11.8 % of female patients, but no specific pattern was identified for this combined fungal sensitivity.

Unlike the results obtained from studying elevated specific IgE levels to fungal allergens in women, the analysis of elevated specific IgG4 levels in the same patient group with microbial eczema revealed: no cases of simultaneous elevation of specific IgG4 to five of the studied fungal allergens [5].

Analogous to the group of women who participated in the study, an analysis of the results of elevated levels of specific IgG4 to fungal allergens was conducted in men with microbial eczema, who constituted 63 % of the total patient group.

Among the total number of examined men, the majority – 72.4 % of patients – had elevated levels of specific IgG4 to the allergen of the fungus *Mucor racemosus*, unlike the group of women with microbial eczema, in whom elevated levels of specific IgG4 to the allergen of the fungus *Aspergillus fumigatus* were most frequently detected. Second in frequency was the elevation of specific IgG4 levels to the allergen of the fungus *Candida albicans* – in 65.5 % of male patients. Elevated levels of specific IgG4 to the allergen of the fungus *Aspergillus fumigatus* were detected in 34.5 % of patients. Elevated levels of specific IgG4 to the allergen of the fungus *Alternaria tenuis* were detected in 31 % of examined men with microbial eczema during the study. Least frequently, elevated levels of specific IgG4 to the allergen of the fungus *Penicillium notatum* were detected – in 3.4 % of male patients. No cases of elevated specific IgG4 levels to the allergen of the fungus *Cladosporium herbarum* were detected among the male group. No elevation of specific IgG4 levels to any of the studied fungal allergens was observed in 7 % of male patients with microbial eczema (Table 2).

Table 2

Frequency of occurrence of elevated levels of specific IgG4E to fungal allergens among examined patients, depending on the patient's sex

Patients	Frequency of Elevated Specific IgG4 to Fungal Allergens, %					
	Not Detected	To <i>Penicill. notatum</i>	To <i>Alternar. tenuis</i>	To <i>Mucor racemos.</i>	To <i>Aspergill. fumigatus</i>	To <i>Cladosp. herbar.</i>
Women	--	11.8	35.3	47.1	52.9	29.4
Men	7	3.4	31	72.4	34.5	--

Elevation of specific IgG4 levels to only one of the studied fungi was detected in 17.2 % of patients, with equal distribution of mono-sensitization to the allergen of the fungus *Candida albicans* and the allergen of the fungus *Mucor racemosus* observed in these cases. Simultaneous elevation of specific IgG4 levels to two of the studied fungal allergens was detected in 48.3 % of male patients, with 90 % of cases showing sensitivity to the same fungal allergens – *Candida albicans* and *Mucor racemosus* – which can likely be explained by the presence of shared allergenic components in the structure of these fungal species [2]. Simultaneous elevation of specific IgG4 levels to three of the studied fungal allergens was detected in 17.2 % of male patients, while simultaneous elevation of specific IgG4 levels to four of the studied fungal allergens was found in 10.3 % of patients. This differs from the results obtained in the study of elevated specific IgE levels to fungal allergens in men, where no cases of simultaneous elevation of specific IgE levels to four studied fungal allergens were detected.

Thus, in the examination of patients with a confirmed diagnosis of “Microbial Eczema” (duration of illness ≥ 1 year), the determination of specific IgG4 levels revealed a high prevalence of elevated specific IgG4 to fungal allergens: *Penicillium notatum*, *Aspergillus fumigatus*, *Mucor racemosus*, *Alternaria tenuis*, *Cladosporium herbarum*, and *Candida albicans*. Fungal sensitization is a characteristic feature in patients with chronic microbial eczema [3]. For etiological diagnosis of exacerbations in chronic microbial eczema and uncontrolled disease progression, the determination of specific fungal IgE and IgG is informative [8, 10], which clarifies the nature of sensitization and enables adequate treatment of this patient group. Further investigation into the role of fungal infection in the development and progression of microbial eczema is critically needed. The data obtained will open new prospects for improving treatment efficacy in patients with microbial eczema.

Conclusion

The results of the conducted studies indicate that in patients with chronic microbial eczema and elevated levels of specific IgG4, both cellular and humoral immunity disorders are observed, including impaired phagocytosis and dysregulation of the production of the main classes of immunoglobulins.

Thus, in patients with chronic microbial eczema and elevated levels of specific IgG4, a vicious circle of pathological reactions is formed: dysregulation of humoral immunity leads to the production of ineffective antibodies (IgG4), which, on the one hand, are unable to provide protection, and on the other hand, suppress phagocytosis, further deepening the defect in cellular immunity and maintaining chronic skin inflammation.

Such immunological abnormalities suggest that chronic microbial eczema is not merely a local dermatological problem but a systemic condition associated with overall immune insufficiency. This predisposes patients to a protracted and recurrent course of the disease, the development of secondary infectious complications, and reduced effectiveness of standard therapeutic approaches.

The findings highlight the importance of a comprehensive treatment strategy for these patients. In addition to anti-inflammatory and antimicrobial agents, the use of immunomodulatory interventions aimed at restoring phagocytic activity, normalizing immunoglobulin production, and correcting the imbalance between cellular and humoral immunity appears warranted. Such an approach may contribute to longer remission periods, reduced recurrence rates, and improved quality of life in affected individuals.

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