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ALTERNARIA ALTERNATA AS THE MAJOR FUNGAL ALLERGEN: MOLECULAR ANALYSIS OF FUNGAL SENSITIZATION PATTERNS

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Background. Among the agents causing respiratory and skin pathologies, fungal allergens play a significant role. The epidemiological situation regarding fungal sensitization in Ukraine, specifically in the Vinnytsia region, remains insufficiently studied. Conversely, its characteristics, defined at both the individual and population levels, may contribute to understanding patterns relevant to prevention of fungal allergy. The aim of our study was to evaluate the prevalence and analyze the sensitivity profiles to fungal allergens among residents of the Vinnytsia region.

Materials and Methods. We analyzed sensitization profiles to 13 fungal allergens in 2,623 residents of the Vinnytsia region using the ALEX multicomponent test. We considered sIgE levels for major molecules and extracts of *Alternaria, Aspergillus, Cladosporium, Malassezia, Penicillium*, and *Saccharomyces* (sensitivity threshold ≥ 0.31 kU/L). We examined the overall prevalence and isolated sensitization cases. Statistical analysis utilized non-parametric methods (Mann–Whitney *U*-test, χ^2 with Yates' correction, $p < 0.05$) in Statistica 8.0 and MS Excel.

Results. The overall prevalence of sensitization to fungal allergens was 9.57 %, and it was 1.85 times more common in children. The leading allergen was *Alternaria alternata* (Alt a 1), with sensitivity observed in 70.12 % of patients sensitized to fungal components. Among children, 82.93 % were sensitized to Alt a 1 alone. The mediansIgE value for Alt a 1 in children (25.64 kU/L) was significantly higher than in adults (7.87 kU/L) and was the highest among sIgE levels for all fungal components. *Aspergillus fumigatus* allergens (Asp f 1, Asp f 6) did not show isolated sensitization, which is consistent with their classification as secondary sensitizing agent, although their sIgE levels were

substantial. Adults showed more frequent sensitization to Asp f 1 and Asp f 6, while sensitivity to Alt a 1 and Mala s 6 was more prominent in children.

Conclusions. The obtained data confirm a high prevalence of fungal sensitization in the Vinnytsia region, particularly hypersensitivity to Alt a 1 among children. The identified age-related and immunological features of allergen sensitivity could contribute to the diagnosis and development of effective, personalized allergen-specific immunotherapy programs.

Keywords: allergic diseases, fungal sensitization, multicomponent molecular allergy diagnostics, molecular sensitization profiles, *Alternaria alternata*, *Malassezia sympodialis*, polysensitization

INTRODUCTION

Allergic diseases are among the key challenges of modern medicine, affecting the quality of life of millions of people worldwide. According to WHO data, allergies affect 20 % to 40 % of the population in various regions, and their prevalence continues to grow, including in Ukraine (Bohomolov & Zaikov, 2020; Rhee *et al*., 2014; Zheng *et al*., 2025). Fungal allergens play a significant role in the development of allergic reactions such as bronchial asthma (BA), allergic rhinitis (AR), atopic dermatitis (AD), and other IgE-mediated conditions. It is well known that allergy to fungi is the third most frequent cause of respiratory pathologies and is closely linked to a worsening asthma prognosis: fungal sensitization is associated with uncontrolled asthma (up to 75 % of cases) and an increased risk of hospitalization, particularly among children and adolescents (Fang *et al*., 2023; Forkel *et al*., 2021). Ubiquitous allergens that cause rhinitis, asthma, and severe respiratory conditions include, notably, the spores of *Alternaria alternata*, *Aspergillus* spp., and *Cladosporium* spp. (Sánchez *et al*., 2022). In Southeast Asia, sensitization to *Curvularia lunata* is observed (Sio *et al*., 2021).

According to previous studies, fungi provoke immune system responses via Th2 and Th17 pathways, leading to airway inflammation and asthma exacerbations, especially in childhood (Sánchez *et al*., 2022). Consequently, personalized antifungal therapy may reduce symptoms and the need for corticosteroids in patients with fungal asthma (Rapeport *et al*., 2020).

The patterns of vegetation and the species composition of the atmospheric fungal spectrum largely depend on climate, humidity, ecological zones, and the level of urbanization (Nageen *et al*., 2023). This leads to regional characteristics of fungal allergy. In the USA, sensitization is higher in urbanized areas and prairies/steppes compared to forests, with a higher risk in grassland ecosystems (Kwong *et al*., 2023). In Europe, for example in Germany, the frequency of fungal sensitization increased from 19.2 % to 22.5 % over two decades and is higher in steppe zones, with a predominance of patients aged 21–40 years (Forkel *et al*., 2021); in Ukraine, fungal hypersensitivity is lower in the Carpathians and higher in the steppe regions (Rodinkova *et al*., 2024).

Climate change also extends the fungal growing season. Notably, in the USA, the season has lengthened by three weeks, intensifying the impact of global warming on allergic manifestations (Wu *et al*., 2025). In Europe, higher concentrations of fungal spores are registered in warm, humid regions, such as Greece (up to 23.8 %), and lower concentrations in northern regions, such as Finland (2 %) (Sánchez *et al*., 2022).

In the Vinnytsia region, characterized by a temperate continental climate with high humidity, the population regularly contacts fungal spores both in the environment (parks, forests, agricultural lands) and in enclosed spaces (residential buildings, offices, public places). Specifically, the humid conditions characteristic of the Vinnytsia region favor the active growth of *Alternaria alternata*, *Aspergillus fumigatus*, *Cladosporium herbarum*, and *Malassezia sympodialis*, which are the primary sources of allergens (Abel-Fernández *et al*., 2023; Nowicka & Nawrot, 2019; Latgé & Chamilos, 2019). These microorganisms can cause both local and systemic allergic reactions, complicating their diagnosis and treatment.

Therefore, the aim of our work was to evaluate the prevalence of sensitivity to fungal allergens among the population of the Vinnytsia region and to analyze the age and gender characteristics of this sensitivity for a better understanding of the epidemiological situation in the region.

MATERIALS AND METHODS

To achieve the stated objective, we analyzed hypersensitivity profiles to fungal allergens in 2,623 residents of the Vinnytsia region. Data were obtained using the multicomponent molecular ALEX test, which allows for the determination of specific immunoglobulin E (sIgE) to particular allergenic components. The study included patients with asthma, allergic rhinitis, atopic dermatitis, and combined allergic conditions. Exclusion criteria included the absence of any allergy-related diseases and the absence of sensitization to fungal allergens.The ALEX panel includes 13 individual fungal molecules and extracts: Alt a 1 (the major allergen of *Alternaria alternata*, belonging to the AA1s protein family) (Zhang *et al*., 2019); Alt a 6 (Alternaria enolase); four *Aspergillus fumigatus* allergens, including the mitogillin family representative Asp f 1, the peroxisomal protein Asp f 3, Asp f 4 (undefined class), and Mn-superoxide dismutase Asp f 6. The analyzed allergenic components of *Cladosporium herbarum* included its extract Cla h, as well as mannitol dehydrogenase Cla h 8. Sensitivity to *Malassezia sympodialis* in ALEX was determined for Mala s 5 (undefined class), Mala s 6 (cyclophilin), and Mala s 11 (Mn-superoxide dismutase). The ALEX fungal allergen panel also included extracts of *Penicillium chrysogenum* and *Saccharomyces cerevisiae* (Pen ch and Sac c, respectively). According to the ALEX test reference values, the sensitization threshold for each component was set at 0.31 kU/L. When analyzing the number and percentage of patients sensitized to the described molecular components, both the total number and percentage of patients sensitive to a specific allergen in the sample, as well as the number and percentage of patients sensitive *only* to that fungal allergen, were considered. The first indicator included all patients who exhibited a reaction to the allergen. The second indicator reflected the percentage of patients sensitized to only one specific fungal allergen among all those studied, without concomitant hypersensitivity to other fungal components.

Statistical analysis was conducted using Statistica 8.0 and the Microsoft Office Excel software package. To assess the strength of the patients' immune response, median values of specific immunoglobulin E (sIgE) for each molecular component of the fungi were analyzed. These indicators were calculated separately for the total sample, as well as for children and adults. Due to the small number of patients hypersensitized to each component, the sample did not follow a Gaussian distribution; therefore, non-parametric

statistics were used. The Yates-corrected Chi-square test was used to compare the absolute frequencies of subjects sensitive to allergens, and the Mann–Whitney *U*-test was used to compare sIgE values between subjects. Statistical significance for all tests was set at ($p < 0.05$).

RESEARCH RESULTS

General results showed that 251 individuals, or 9.57 % of the total sample, were hypersensitive to at least one fungal allergen. Among the sensitized individuals, 163 (64.94 %) were children under 18 years of age, and 88 (35.06 %) were adults. Thus, the frequency of sensitivity among children was 1.85 times higher than among adults ($p < 0.0001$, $\chi^2 = 22.41$).

Hypersensitivity analysis to specific allergens. Alt a 1 (*Alternaria alternata*) was the leading fungal allergen: 176 patients (70.12 % of all fungal-sensitive patients) were sensitized to it. Notably, approximately 83.00 % exhibited monosensitization specifically to Alt a 1, a trend observed among both children and adults.

Other *Alternaria* components had significantly lower prevalence: Alt a 6 – 3.59 %; Asp f 1 (*Aspergillus fumigatus*) – 1.20 %, Asp f 3 – 5.58 %, Asp f 4 – 3.98 %, and Asp f 6 – 4.78 %. Monosensitization to these components was rare or absent.

Sensitivity to the *Cladosporium* extract (Cla h) was found in 4.38 % of patients, and to Cla h 8 in 3.98 %. Monosensitization was recorded only in isolated cases among both children and adults.

Malassezia sympodialis components played a notable role in the sensitization structure: Mala s 11 – 9.96 % (nearly half presented with monosensitization), Mala s 6 – 9.16 % (about one-third sensitive only to this allergen), and Mala s 5 – 4.38 % (over half of those sensitized were monosensitized). In most cases involving Mala s 5 and Mala s 11, children predominated, while for Mala s 6, children were also more frequently affected, though without statistically significant differences.

Sensitization to Pen ch (*Penicillium chrysogenum*) was low (2.79 %), with isolated cases of monosensitization primarily among children.

The yeast extract Sac c (*Saccharomyces cerevisiae*) also held significant importance: 9.16 % of patients were sensitive to it, and approximately one-third of these were sensitive to this allergen alone, with a similar proportion of children and adults in the sensitization structure (**Table 1**).

Analysis of allergen combinations in patient profiles. The analysis of fungal allergen combinations in the profiles of sensitized patients revealed that the most frequent profile was the presence of Alt a 1 as the sole agent of hypersensitivity. This profile, as mentioned above, was identified in 146 individuals, or 58.17 % of fungal-sensitized patients.

Ranking second in frequency, with a significant margin from the leader, was isolated sensitivity to Mala s 11 ($p < 0.0001$), observed in 12 patients (4.78 % of all fungal-sensitized individuals). The combination of Alt a 1 + Mala s 6 followed in prevalence, appearing in 8 patients (3.19 %). Among other common profiles, the combination of Mala s 6 and Sac c was observed in 7 individuals (2.79 %).

Two sensitization variants shared the same frequency, appearing in 6 patients each (2.39 %): isolated sensitivity to the Mala s 5 allergen and the combination of Asp f 6 + Mala s 11. Similarly, the combination of Alt a 1 + Asp f 3 and the isolated presence

Table 1. **Number and percentage of patients sensitized to molecular fungal components**

Allergen component name	Patients sensitive to the allergen, n (%)		Of which, patients sensitive only to this fungal allergen, n (%)		Children sensitive to the allergen, n (%)		Of which, children sensitive only to this fungal allergen, n (%)		Adults sensitive to the allergen, n (%)		Of which, adults sensitive only to this fungal allergen, n (%)	
	patients	%	patients	%	patients	%	patients	%	patients	%	patients	%
Alt a 1	176	70.12	146	82.95	123	69.89	102	82.93	53***	30.11	44	83.02
Alt a 6	9[a,b,c,d]	3.59	3[a,c]	33.33	5[a,b]	55.56	2[a]	40.00	4[a,c,d]	44.44	1[a]	25.00
Asp f 1	3[a]	1.20	0[a,c]	0.00	0[a, b]	0.00	0[a]	0.00	3[a,c]	100.00	0[a]	0.00
Asp f 3	14[a,e]	5.58	4[a]	28.57	9[a]	64.29	1[a]	11.11	5[a]	35.71	3[a]	60.00
Asp f 4	10[a,b,c,d]	3.98	4[a]	40.00	6[a]	60.00	1[a]	16.67	4[a,c]	40.00	3[a]	75.00
Asp f 6	12[a, e]	4.78	0[a,c]	0.00	3[a,b]	25.00	0[a]	0.00	9[a]	75.00	0[a]	0.00
Cla h	11[a,c,e]	4.38	2[a,c]	18.18	5[a,b]	45.45	1[a]	20.00	6[a]	54.55	1[a]	16.67
Cla h 8	10[a,b,c,d]	3.98	2[a,c]	20.00	5[a,b]	50.00	1[a]	20.00	5[a]	50.00	1[a]	20.00
Mala s 11	25[a,e]	9.96	12[a]	48.00	12[a]	48.00	7[a]	58.33	13[a]	52.00	5[a]	38.46
Mala s 5	11[a,c,e]	4.38	6[a]	54.55	6[a]	54.55	4[a]	66.67	5[a]	45.45	2[a]	40.00
Mala s 6	23[a,e]	9.16	7[a]	30.43	18[a]	78.26	5[a]	27.78	5*[a]	21.74	2[a]	40.00
Pen ch	7[a]	2.79	2[a,c]	28.57	4[a,b]	57.14	2[a]	50.00	3[a,c,d]	42.86	0[a]	0.00
Sac c	23[a,e]	9.16	7[a]	30.43	11[a]	47.83	4[a]	36.36	12[a]	52.17	3[a]	25.00

Note: * – $p < 0.05$, *** – $p < 0.001$: significant difference in sensitivity between adults and children.
a – significant difference relative to Alt a 1; b – significant difference relative to Mala s 6; c – significant difference relative to Mala s 11; d – significant difference relative to Sac c; e – significant difference relative to Asp f 1

of either Asp f 3 or Asp f 4 molecules showed equal prevalence, with each variant occurring in 4 patients (1.59 %). The next group consisted of allergens and their combinations occurring in 3 patients each (1.20 %): the Alt a 6 allergen and the Asp f 3 + Asp f 4 combination. Seven allergen combinations were observed in 2 patients each (0.80 %): Alt a 1 + Cla h 8; Alt a 1 + Cla h + Cla h 8; Alt a 1 + Sac c; isolated Cla h; isolated Cla h 8; Mala s 11 + Mala s 6; and isolated Pen ch. The remaining combinations were unique and were observed in only one patient each (**Fig. 1**).

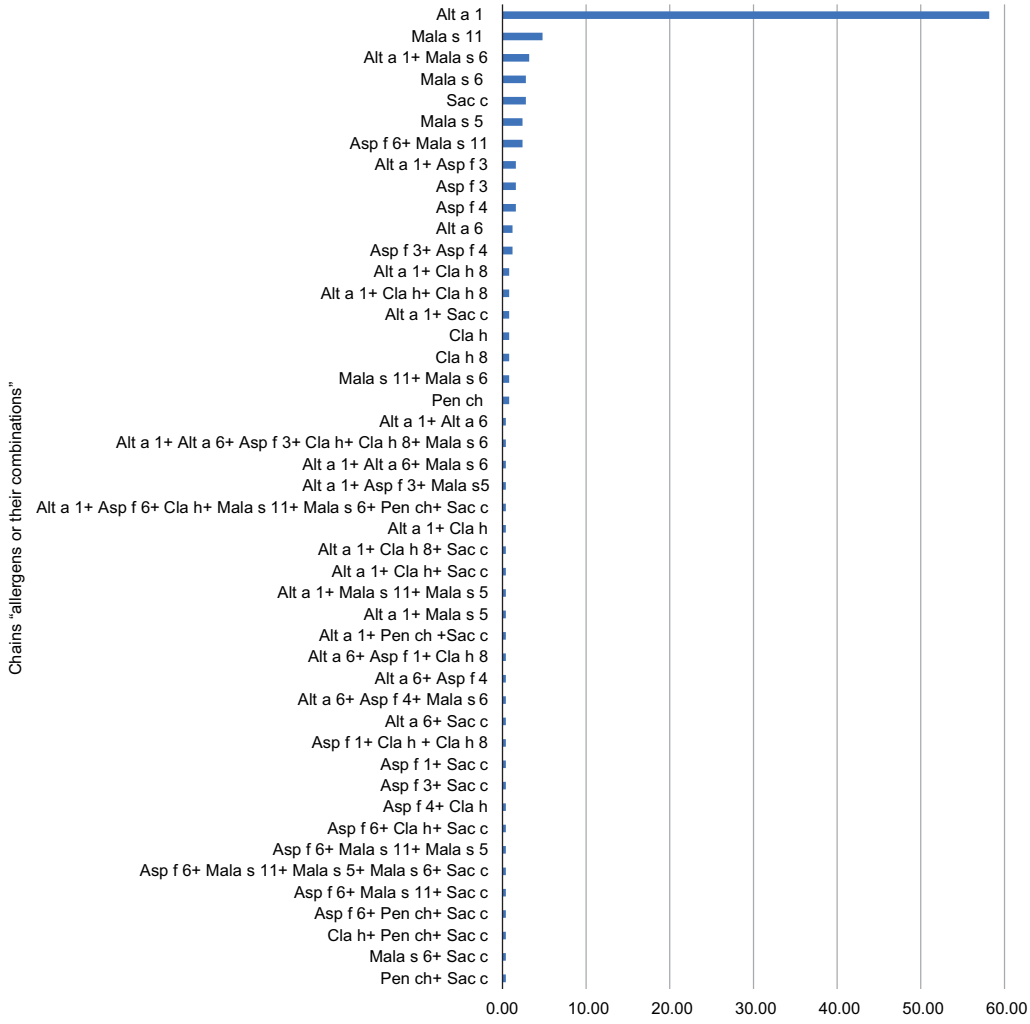

Fig. 1. Frequency of sensitization profiles in patients (%), categorized by specific combinations of allergenic components

The distribution of sensitivity to the most common allergens and their combinations revealed that adults predominated in only 3 out of the 10 most frequent profiles (Asp f 6 + Mala s 11, Asp f 3, and Asp f 4). Isolated sensitivity to Sac c was characterized by an almost equal distribution (4 children vs. 3 adults), whereas the combination of Alt a 1 + Asp f 3 was observed exclusively in children (**Fig. 2**).

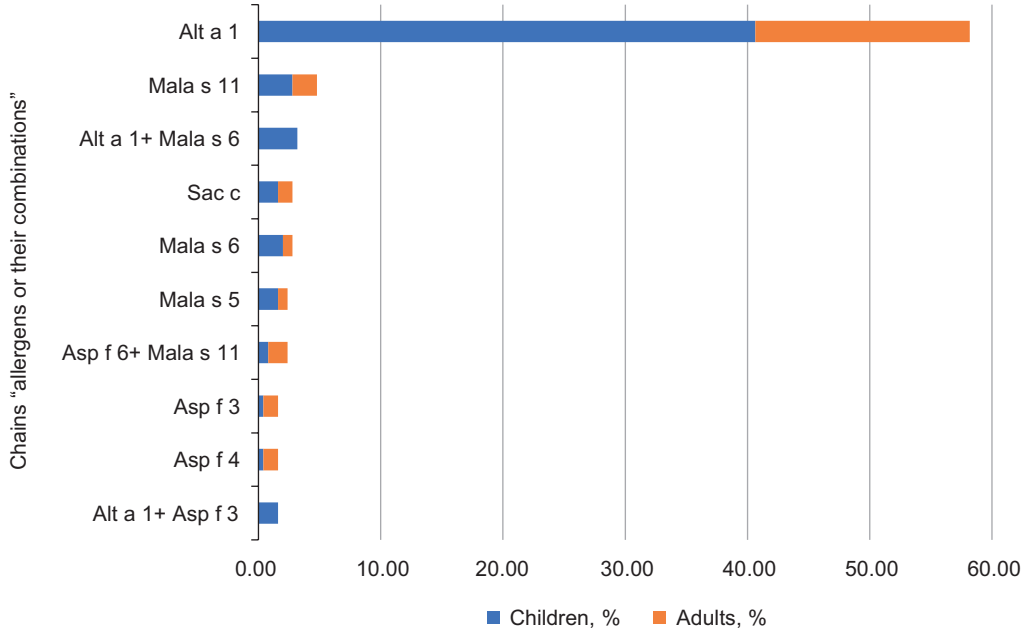

Fig. 2. Proportion of different age groups within the patient profiles sensitive to the most prevalent allergen combinations

Analysis of patient hypersensitivity levels to individual fungal allergen components. The analysis of specific IgE (sIgE) levels to various fungal allergens revealed significant differences in the strength of the immune response depending on both allergen type and patient age (**Table 2**). The magnitude of this response was evaluated according to the ALEX classification, which defines low (0.3–1.0 kU/L), moderate (1.0–5.0 kU/L), and high (> 5.0 kU/L) sIgE levels (Čelakovská *et al*., 2022).

In the present sample, the highest sIgE levels were characterized by sensitivity to Alt a 1. The median sIgE value for all individuals sensitive to Alt a 1 was 19.68 kU/L, indicating a strong immune response. Significant variability in individual reactions is highlighted by a substantial interquartile range (IQR) of 30.25.

The median sIgE value for Alt a 1 was significantly higher in children (25.64 kU/L) than in adults (7.87 kU/L) ($p < 0.001$). Notably, the sIgE level for Alt a 1 in adults remained the highest within this age group compared to sIgE levels for all other allergens.

Conversely, very low median sIgE values were observed for Alt a 6 (0.53 kU/L). The IQR was also small (0.71), indicating a less variable immune response intensity. Both indicators for Alt a 6 were lowest in adults (median: 0.45; IQR: 0.34), although this was not statistically significant ($p > 0.05$).

Apart from Alt a 1, significant levels of sensitivity were observed in both adults and children for Asp f 6 (4.16 kU/L) and Mala s 11 (3.24 kU/L); however, these remained below the threshold of sIgE = 5 kU/L, which corresponds to the high hypersensitivity level in the ALEX test. The median sIgE value for Asp f 1 (1.37 kU/L) reflected moderate sensitivity (1–5 kU/L). Median sIgE values for the remaining allergens did not exceed low levels (< 1 kU/L). However, sIgE levels for Asp f 3, Asp f 4, and Pen ch in children, as well as Cla h 8 in adults, did exceed the low-value threshold (**Table 2**).

Table 2. **Median sIgE values of patients sensitized to molecular fungal components**

Allergen component name	Median (kU/L)			IQR			Quartile 3			Quartile 1		
	total	children	adults	total	children	adults	total	children	adults	total	children	adults
Alt a 1	19.68	25.64	7.87***	30.25	32.24	19.38	35.27	39.77	23.62	5.02	7.54	4.24
Alt a 6	0.53	0.82[a]	0.45[a]	0.71	3.23	0.34	1.11	3.67	0.68	0.40	0.43	0.34
Asp f 1	1.37	0	1.37	15.03	0	15.03	15.03	0	15.91	0.89	0	0.89
Asp f 3	0.78	1.89[a]	0.56[a]	1.54	3.85	0.49	1.89	4.18	0.90	0.35	0.33	0.41
Asp f 4	0.77	1.53[a]	0.76[a]	2.10	7.01	0.94	2.52	7.44	1.39	0.42	0.43	0.45
Asp f 6	4.16	3.81[a]	4.51	6.95	2.57	12.38	7.73	4.88	13.07	0.78	2.31	0.69
Cla h	0.47	0.46[a]	0.48[a]	0.78	0.64	0.63	1.17	1.09	1.00	0.39	0.45	0.38
Cla h 8	0.64	0.45[a]	1.05[a]	0.51	0.25	2.78	0.97	0.67	3.39	0.46	0.42	0.61
Mala s 11	3.24	2.14[a]	4.48	5.06	3.76	20.68	5.64	4.31	21.78	0.58	0.56	1.10
Mala s 5	0.44	0.40[a]	0.73[a]	1.09	2.74	0.39	1.44	3.08	0.83	0.36	0.35	0.44
Mala s 6	0.68	0.68[a]	0.81[a]	0.61	0.61	0.38	0.99	0.99	0.83	0.39	0.38	0.45
Pen ch	0.43	1.09[a]	0.43[a]	0.85	1.64	0.11	1.21	1.98	0.51	0.36	0.34	0.41
Sac c	0.72	0.76[a]	0.53[a]	0.73	0.75	0.47	1.15	1.25	0.89	0.42	0.50	0.42

Note: *** – $p < 0.001$: significant difference between adults and children
a – significant difference relative to Alt a 1. sIgE values are expressed in kU/L
IQR – Interquartile Range

DISCUSSION

Despite the fact that the study provides for novel insights into fungal sensitization patterns in Central Ukraine, several limitations of this study should be acknowledged. First, the retrospective nature of the study and the fact that the study population consisted of patients who had already sought medical assistance for allergy-like symptoms may introduce a selection bias, meaning the results might not fully represent the general population. Second, while the ALEX2 multiplex test provides a comprehensive molecular profile, the clinical relevance of sensitization to certain components requires further correlation with specific clinical manifestations, which were not the aim of our study and were not detailed.

Third, the analysis was restricted to the fungal allergens included in the ALEX2 panel; therefore, sensitization to other locally relevant fungal species not represented in the test could have been overlooked. Finally, although Vinnytsia is representative of the forest-steppe zone of Ukraine, the results may not be directly generalizable to other climatic regions of the country, such as the southern areas or the northern forest zones, where different fungal spore concentrations and species diversity may prevail.

Nevertheless, our findings highlight a significant clinical burden, revealing a high prevalence of fungal sensitization in the Vinnytsia region: 9.57 % among 2,623 subjects, which corresponds to the upper limit of global estimates (3–10 %) (Fernández-Soto *et al*., 2018; López Couso *et al*., 2021).

The high frequency of hypersensitivity, particularly in children, aligns with local aerobiological conditions: in this region characterized by a humid temperate continental climate, with the concentrations of *Alternaria* and *Cladosporium* spores consistently exceeding clinical thresholds (Paredes Idiaquez et. al, 2025). Specifically, *Alternaria* levels exceeding 100 spores/m³ (Anees-Hill *et al*., 2022; Torres-Borrego *et al*., 2025) are observed for dozens of days per year (Kasprzyk *et al*., 2015). Unlike the short season of pollen allergy (Anderegg *et al*., 2021; Frisk *et al*., 2024; Zhang & Steiner, 2022), mold spores can persist indoors, ensuring nearly constant contact (Cervantes *et al*., 2025).

Alt a 1 from *Alternaria alternata* was identified as the most prevalent sensitizing component, which is consistent with data identifying *Alternaria* as one of the most potent airborne allergens (Sánchez *et al*., 2022; Wójcik-Kanach & Kasprzyk, 2025). Alt a 1 forms a distinct protein class found only in fungi, and is a key target for precision diagnostics and allergen immunotherapy (AIT) (Abel-Fernández *et al*., 2023; Izmailovich *et al*., 2023). The median sIgE value for Alt a 1 in children was three times higher than that of adults, which, according to the literature, is associated with a high risk of severe, often steroid-resistant asthma and hospitalizations (Arshad *et al*., 2024; Bush, 2020; Hughes *et al*., 2022; Klain *et al*., 2025).

Molecular diagnostics allowed for the differentiation between primary sensitizers (Alt a 1, Mala s 5, Mala s 11) and secondary/cross-reactive markers (Asp f 1, Asp f 6). For the latter, no cases of monosensitization were detected, confirming their role as indicators of polysensitization and a supposedly more severe clinical course (Wardlaw *et al*., 2021; Cacheiro-Llaguno *et al*., 2024; Patchett *et al*., 2023). The low frequency of sensitization to Asp f 1 (1.20 %) and its detection exclusively in adults aligns with the understanding of this component as a marker for complex chronic conditions, such as allergic bronchopulmonary aspergillosis (ABPA) and severe asthma with fungal sensitization (SAFS) (Asano *et al*., 2021; Chen *et al*., 2022; De Linares *et al*., 2023). Allergens Asp f 3 and Asp f 4, useful for ABPA diagnosis (Michel *et al*., 2022; Tiew *et al*., 2023), are associated with

the ability to damage the respiratory epithelium and enhance the penetration of other allergens (Kwong *et al*., 2023; Lukaszewicz *et al*., 2022; Wardlaw *et al*., 2021).

This aligns with evidence that *A. fumigatus* sensitization increases the risk of severe asthma by up to 3-fold, often as SAFS/ABPA with exacerbations and hospitalizations. Median sIgE for Asp f 6 (4.16 kU/L overall; 4.51 kU/L adults, **Table 2**) suggests persistent inflammation and steroid resistance (Mistry *et al*., 2021; Goh *et al*, 2017).

Components of *Malassezia sympodialis*, especially Mala s 11 (Mn-superoxide dismutase), demonstrated a high proportion of monosensitization and the ability to induce dendritic cell maturation and the release of TNF-α and IL-6 (Kader *et al*., 2021; Nowicka & Nawrot, 2019). Meanwhile, the Alt a 1 + Mala s 6 profile, found exclusively in children, suggests a link between skin barrier defects, atopic dermatitis, and respiratory allergy (Altrichter *et al*., 2020; Celakovska *et al*., 2021; Nowicka & Nawrot, 2019). The significant prevalence of sensitization to *Saccharomyces cerevisiae* (Sac c) and cases of monosensitization underscore its role as an independent allergen, important in food, respiratory (Belda *et al*., 2019; Parapouli *et al*., 2020), and occupational allergies with atypical clinical presentations (Alska *et al*., 2025; Dramburg *et al*., 2023; Xing *et al*., 2022).

In accordance with literature, Mala s profiles (e.g., Alt a 1 + Mala s 6 at 3.19 %; Mala s 6 + Sac c at 2.79 %) may indicate multi-system severity, especially in children. High monosensitization (48 % Mala s 11) and sIgE (3.24 kU/L Mala s 11) link to atopic dermatitis progression to asthma/rhinitis, may exacerbate skin dysfunction and flares. The pediatric-exclusive Alt a 1 + Mala s 6 (sIgE 25.64 kU/L for Alt a 1) may signal for a highly atopic phenotype, requiring integrated topical and respiratory management (Celakovská *et al*., 2018; Storz *et al*., 2024).

Sac c sensitization in polysensitization is known to amplify respiratory severity via food/occupational triggers and poorer lung function, highlighting avoidance and component-resolved diagnostics (Gueçamburu *et al*., 2025; Cacheiro-Llaguno *et al*., 2024).

Thus, polysensitization imposes a greater burden than monosensitization, with elevated uncontrolled symptoms and comorbidities. ALEX testing enables stratification for personalized AIT, targeting primaries while managing secondaries to mitigate severity in fungal-allergic patients.

These results demonstrate a high clinical value of multicomponent molecular diagnostics (Abel-Fernández *et al*., 2023; Hernandez-Ramirez *et al*., 2021) and are consistent with the rationale for personalized AIT (primarily targeting Alt a 1) (Izmailovich *et al*., 2023; López Couso *et al*., 2021; Rodríguez *et al*., 2021). Furthermore, they highlight the need for further research accounting for local aerobiological features and the impact of climate change on the seasonality of *Alternaria* (Rodinkova *et al*., 2024; Sztandera-Tymoczek & Szuster-Ciesielska, 2023; Ziska, 2021).

CONCLUSIONS

The molecular study of fungal sensitization in the Vinnytsia region revealed a significant level of hypersensitivity to fungal allergens among the population, particularly pronounced in the pediatric group, and allowed for the identification of key allergenic components with major clinical significance.

Specifically, the major allergen Alt a 1 from *Alternaria alternata* was identified as the primary etiological factor of fungal sensitization, with sensitivity detected in 70.12 % of the examined patients. Notably, for 82.95 % of all individuals sensitized to *Alternaria*, Alt a 1 was the sole fungal component of hypersensitivity. The median sIgE level for Alt a 1

in children was 3.26 times higher than in adults, which has been associated in previous studies with increased fungal-associated asthma severity in the pediatric population.

Multicomponent molecular allergy diagnostics enabled a precise differentiation of primary etiological sensitizers (Alt a 1, Mala s 5, Mala s 11) from secondary, cross-reactive markers of hypersensitivity (Asp f 1, Asp f 6). The absence of monosensitization to the latter in children highlights their secondary role. This precision confirms the clinical rationale for conducting Allergen Immunotherapy (AIT) aimed at reducing sensitivity to Alt a 1 in the sensitized cohort. Furthermore, the Alt a 1 + Mala s 6 co-sensitization profile, found exclusively in children, defines a highly atopic phenotype linking skin barrier dysfunction to respiratory allergy, necessitating an integrated clinical approach.

These findings can be utilized to better understand the characteristics of the immune response to fungal allergens across different age groups and to improve the diagnosis and treatment of relevant allergic conditions. Regional clinical recommendations for managing fungal-induced allergies should ensure the optimization of therapeutic outcomes, particularly through standardized, component-resolved AIT, to effectively control fungal allergy in Central Ukraine.

Future studies should further examine the observed fungal sensitization patterns using prospective study designs and independent patient cohorts. Additional research is needed to validate these profiles in clinical practice and to evaluate the effectiveness of AIT in sensitized patients. Combining molecular sensitization data with aerobiological monitoring may help to better interpret environmental exposure. From a clinical perspective, the findings may assist allergists in refining diagnostic approaches and in identifying patients who may be considered for further evaluation for AIT; however, these applications require confirmation in prospective studies.

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COMPLIANCE WITH ETHICAL STANDARDS

Conflict of Interest: the authors declare that they have no known competing financial interests or personal relationships that could have influenced the work presented in this article.

Animal Rights: this article does not contain any studies involving animals.

Human Rights: this article does not contain any studies involving human participants performed by any of the authors. [Note: If the data was derived from retrospective laboratory records, this standard statement is appropriate].

Ethical Approval: the study was approved by the Bioethics Committee of National Pirogov Memorial Medical University, Vinnytsia (Protocol No. 10, dated October 27, 2025).

AUTHOR CONTRIBUTIONS

Conceptualization: [R.Y.; Y.S.]; methodology: [R.V.; R.Y.]; validation: [R.Y.; R.V.; Y.S.; D.O.]; formal analysis: [R.Y.]; investigation: [Y.S.; R.Y.]; resources: [Y.S.; D.O.]; data curation: [Y.S.; R.V.]; writing – original draft preparation: [R.Y.; R.V.; D.O.]; writing – review & editing: [Y.S.; R.Y.; R.V.]; visualization: [R.Y.; R.V.]; supervision: [Y.S.; D.O.]; project administration: [Y.S.; R.V.]; funding acquisition: [–].

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ALTERNARIA ALTERNATA ЯК ПРОВІДНИЙ АЛЕРГЕН ГРИБІВ: МОЛЕКУЛЯРНИЙ АНАЛІЗ ЗАКОНОМІРНОСТЕЙ ГРИБКОВОЇ СЕНСИБІЛІЗАЦІЇ

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Вступ. Алергічні захворювання вражають значну частину населення світу і є однією з найпоширеніших проблем сучасної медицини. Серед агентів, що спричиняють респіраторні та шкірні патології, грибкові алергени відіграють значну роль, особливо в умовах підвищеної вологості й урбанізації. Попри це, епідеміологічна ситуація щодо грибкової сенсибілізації в Україні, зокрема, у Вінницькій області, залишається недостатньо вивченою. Натомість, її особливості, визначені на індивідуальному та популяційному рівнях, можуть стати вагомим підґрунтям ефективної профілактики алергії до грибів.

Мета. Метою нашого дослідження були оцінка поширеності й аналіз профілів чутливості до грибкових алергенів серед жителів Вінницької області на основі даних, отриманих за допомогою молекулярного тесту ALEX.

Матеріали та методи. Ми проаналізували профілі сенсибілізації до 13 грибкових алергенів у 2623 жителів Вінницької області, використовуючи мультокомпонентний тест ALEX. Ми врахували рівні sIgE для мажорних молекул і екстрактів *Alternaria*, *Aspergillus*, *Cladosporium*, *Malassezia*, *Penicillium* та *Saccharomyces* (поріг чутливості ≥ 0,31 kU/L). Вивчали загальну поширеність і випадки ізольованої сенсибілізації до окремих компонентів. Статистичний аналіз проведено за допомогою непараметричних методів (тест Манна–Вітні, χ^2 з поправкою Йейтса, $p < 0,05$) у програмах Statistica 8.0 та MS Excel.

Результати. Загальна поширеність сенсибілізації до грибкових алергенів становила 9,57 % від усіх 2623 протестованих. Гіперчутливість до грибів була в 1,85 разу частіше зареєстрована у дітей, ніж у дорослих. Провідним алергеном була *Alternaria alternata* (Alt a 1), чутливість до якої спостерігали у 70,12 % пацієнтів, сенсибілізованих до компонентів грибів. Серед дітей 82,93 % були сенсибілізовані лише до Alt a 1. Медіанне значення специфічного IgE (sIgE) до Alt a 1 у дітей (25,64 kU/L) було значно вищим, ніж у дорослих (7,87 kU/L), і було найвищим

серед sIgE до усіх грибкових компонентів. Алергени *Aspergillus fumigatus* (Asp f 1, Asp f 6) не продемонстрували ізольованої сенсибілізації, що вказує на їхню роль як вторинних алергенів у полісенсибілізованих пацієнтів, хоча їхні рівні sIgE були суттєвими. Дорослі частіше сенсибілізувалися до Asp f 1 і Asp f 6, тоді як чутливість до Alt a 1 і Mala s 6 була більш виражена у дітей.

Висновки. Отримані дані підтверджують високу поширеність грибкової сенсибілізації у Вінницькій області, особливо гіперчутливості до Alt a 1 серед дітей. Виявлені вікові й імунологічні особливості чутливості до алергенів є важливими для покращення діагностики та розробки ефективних, персоніфікованих програм алерген-специфічної імунотерапії.

Ключові слова: алергічні захворювання, грибкова сенсибілізація, багатокомпонентна молекулярна діагностика алергії, молекулярні профілі сенсибілізації, *Alternaria alternata*, *Malassezia sympodialis*, полісенсибілізація

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