

Prevalence of immunoglobulin E for fungi in atopic children

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Summary

Background The prevalence of sensitization to fungi in young atopic patients in relation to age and clinical importance is largely unknown.

Objective The aim of this study was to investigate the prevalence of sensitization to different fungi in atopic children in relation to age and other aeroallergens.

Methods A total of 137 atopic children (male 62%, female 38%; mean age 5 years and 9 months, range 5 months–14 years) were studied. Sera of all patients were routinely tested for total IgE and specific IgE against aeroallergens and milk. Positive sera were also tested for IgE against *Alternaria alternata*, *Aspergillus fumigatus*, *Cladosporium herbarum* and *Penicillium chrysogenum*, using the Pharmacia Enzyme CAP procedure.

Results In this study in atopic children total IgE showed a significant linear relation with age, whereas specific IgE against outdoor fungi, indoor fungi and house dust mite showed significant non-linearity with age. Prevalence of specific IgE for *Cladosporium* ranked first, followed closely by *Aspergillus* and *Alternaria*. Calculation of the sensitization of indoor and outdoor fungi showed maximum prevalence at 7.8 years, followed by lower values at higher ages. A similar significant relation was also found for *Alternaria*, while this relation was not significant for the other individual fungi. Specific IgE for indoor and outdoor fungi was associated with the presence of specific IgE for aeroallergen and milk. We found that all children aged 4 years and older showed IgE for house dust mite that did not decline with increasing age.

Conclusions Sensitization to fungi is prevalent in childhood, with an age-dependent distribution reaching maximum values at 7.7–7.8 years, followed by a decline for all fungal sensitization with increasing age. The importance and relative contribution of fungal sensitization to airway disease, compared with the other allergens, remains to be established.

Keywords: fungi, children, sensitization, house dust mite, age

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Introduction

Sensitization to inhaled allergens has been studied in young children and may show elevated values for IgE against

environmental inhalant allergens already during the first years of life [1]. Allergy to house dust mite in childhood is the most prominent allergy in western Europe, followed by allergy for cat and dog dander [2,3]. The role of sensitization to fungi in relation to the prevalence and clinical importance in young-aged atopic patients remains largely unknown.

Fungi have been recognized as a cause of allergic airways disease since Blackley firstly inhaled *Penicillium glaucum* in

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1856, inducing 'bronchial catarrh'. Fungi are ubiquitous in nature; identification and classification of fungi as a cause of airway-induced atopic disease has evolved slowly [4]. Detailed information about the presence of sensitizing fungal spores in the outdoor air is available in the Netherlands [5–7].

Information with respect to different fungi is still lacking in the paediatric age-group, although literature suggests an age-dependency of fungal sensitization [2,8,9].

Interest in fungal sensitization has increased after the observations that sensitization to *Alternaria* was predictive for the development of asthma [10,11] and appeared to be a risk factor for severe asthma [12]. Furthermore, studies of IgE antibodies against different fungal extracts showed sensitization in children, reaching 30% [13]. Similarly, a prevalence of positive skin testing with *Cladosporium herbarum* of 40% was found in 4-year-old children, but lower IgE titres were shown in older-aged groups [2]. In a group of adults with obstructive airway disease we found an age-dependent sensitization (skin test) with high values in the younger age-group (16–19 years) and lower values at higher ages [9].

The aim of this study was to investigate the prevalence of sensitization to different fungi in atopic children. We studied overall prevalence of allergy against fungi, as well as age-dependency and relation to clinical atopic symptoms and other airborne allergens, in a group of children with atopic manifestations visiting the Paediatric Department of Pulmonology.

Materials and methods

Patients

Children aged 0–15 years visiting the outpatient Department of Paediatric Pulmonology for the first time in 1996 and 1997 were recruited. The inclusion criterion was the presence of atopic disease (including upper and/or lower respiratory symptoms, asthma and/or rhinitis, and/or eczema). Asthma was diagnosed if they fulfilled the criteria described in the 'international consensus report on diagnosis and treatment of asthma' [13]. Rhinitis and eczema were diagnosed when the answers were positive to the particular questions of the British Medical Research Council [14].

IgE measurement

All patients were routinely screened for total IgE and specific IgE antibodies against house dust mite, grass and tree pollen, cat and dog dander and milk. The sera that were positive for specific IgE against one or more inhalant allergen were subsequently quantified for IgE antibodies against *Alternaria alternata*, *Aspergillus fumigatus*, *Cladosporium herbarum* and *Penicillium chrysogenum*, using the Pharmacia Enzyme CAP procedure. The results are expressed in kU/L. For determination

of IgE titres the classification according to Pharmacia (Uppsala, Sweden) was used. The four fungi were analysed separately or grouped according to indoor fungi (*Penicillium* and *Aspergillus*) and outdoor fungi (*Cladosporium* and *Alternaria*).

Medical information

Information about asthma, eczema, rhinitis and 'other allergies', such as allergy against flowers, plants, drugs or animals, was obtained using the Dutch version of the standardized questionnaire of the British Medical Research Council [14].

Statistical analysis

Restricted cubic spline functions were used in order to test linearity or to describe the non-linear relationships of age with the various outcome variables (IgE sensitization and prevalence, respectively) in multivariable linear or logistic regression models. Interaction between age and sex were added to the model to assess a possible different relationship with age for males and females. Estimates are shown in graphs expressed as probability for sensitization (total IgE and specific IgE to fungi and house dust mite) together with 95% confidence intervals. Chi-square test for trend in binomial proportions (two-sided test) was used to study the trend for the proportion of fungal sensitization for increasing classes of airborne allergens and milk.

Results

In this cross-sectional study, 137 children, 85 males (62%) and 52 females (38%), were included and six children were excluded due to lack of information. The ages ranged from 5 months to 14 years, mean age was 5 years and 9 months.

Distribution of total IgE against age is shown in Fig. 1. Total IgE shows a significant linear relation with age, which is significantly different for males and females.

In Table 1 the class distribution to the numbers of children for the total sensitization (classes 1–6) to *Cladosporium*, *Alternaria*, *Aspergillus* and *Penicillium* and house dust mite (*Dermatophagoides pteronissimus*) allergen are shown, respectively.

The highest prevalence for specific IgE to the different fungi was for *Cladosporium* (18.2%), followed by *Aspergillus* (15.3%), *Alternaria* (14.6%) and *Penicillium* (7.3%).

Figure 2(a and b) shows the expected probability for having specific IgE against the combinations of outdoor fungi and indoor fungi against age. These probabilities were significantly associated (curves were tested for non-linearity), maximal values for probability were found at 7.8 and 7.7 years, respectively. Males show apparently lower probabilities in comparison with females, but these differences are not significant (not shown). Similar

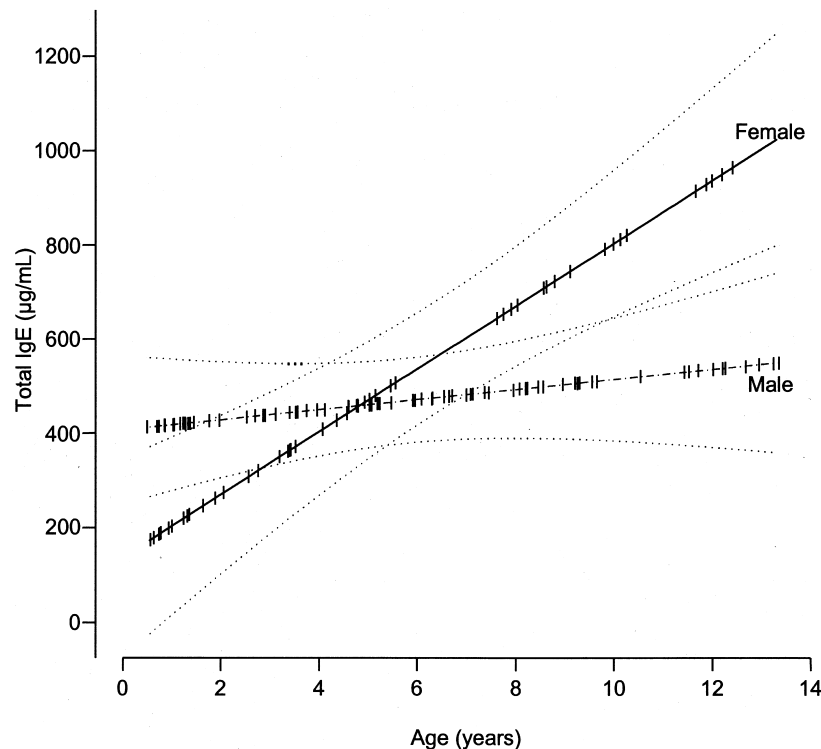


Fig. 1. Distribution of total IgE against age. The prevalences are shown for males and females. Distribution of individuals is indicated in the figure (small vertical lines at top and graph).

age-dependent associations are found for the IgE levels against the four individual fungi, which are significant for both the age-dependency and non-linearity for *Alternaria* only (not shown). Expressing IgE values in kU/L instead of classes showed a slight change in the maximum of the probabilities to lower age values (maximum at 7 years, not shown).

For comparison with the sensitization to fungal allergens, the age-dependent distribution of IgE against the most important inhalant allergen in the Netherlands, the house dust mite *Dermatophagoides pteronissinus*, is shown in Fig. 3. The probability for IgE to house dust mite shows non-linearity due to reaching the maximum of 100% in this group of patients at the age of 4 years without decline with

increasing age. Study of the distribution of IgE in kU/L shows increasing concentrations of IgE to house dust mite allergen, reaching maximal concentrations between 5 and 6 years of age without changes in quantities of IgE at higher ages (not shown). No differences in the distribution of IgE were found between males and females.

The probability for having asthma against age is shown in Fig. 4. Significantly higher values for asthma are found with increasing age, while the curve is significantly non-linear due to the fact that above the age of 3 years all children had developed asthma, according to the clinical criteria used.

We studied the association of having IgE against fungi as a fraction of having IgE against the aeroallergens and milk allergen. It is found that the fraction of positive IgE against outdoor fungi and indoor fungi was significantly associated with increasing class values for IgE for these allergens (not shown). Fungal IgE is most strongly associated with IgE against dog, cat, tree and grasses (chi-square indoor fungi, respectively, 26.17, 16.16, 18.35 and 16.68; chi-square outdoor fungi 33.06, 20.95, 16.13 and 21.52). The association with sensitization to house dust mite and milk allergens, shows lower chi-square values (chi-square indoor fungi, respectively, 7.14 and 12.42; chi-square outdoor fungi 8.39 and 7.28).

Table 1. Numbers of patients positive for IgE against fungal extracts and house dust mite (HDM) in children

Class	<i>Cladosporium</i>	<i>Alternaria</i>	<i>Aspergillus</i>	<i>Penicillium</i>	HDM
0	108	112	49	114	23
1	4	5	14	13	5
2	15	11	16	10	17
3	4	8	5		17
4	4	1			26
5	2				24
6					26
Total	137	137	137	137	137

Discussion

In this study in atopic children, total IgE shows a significant

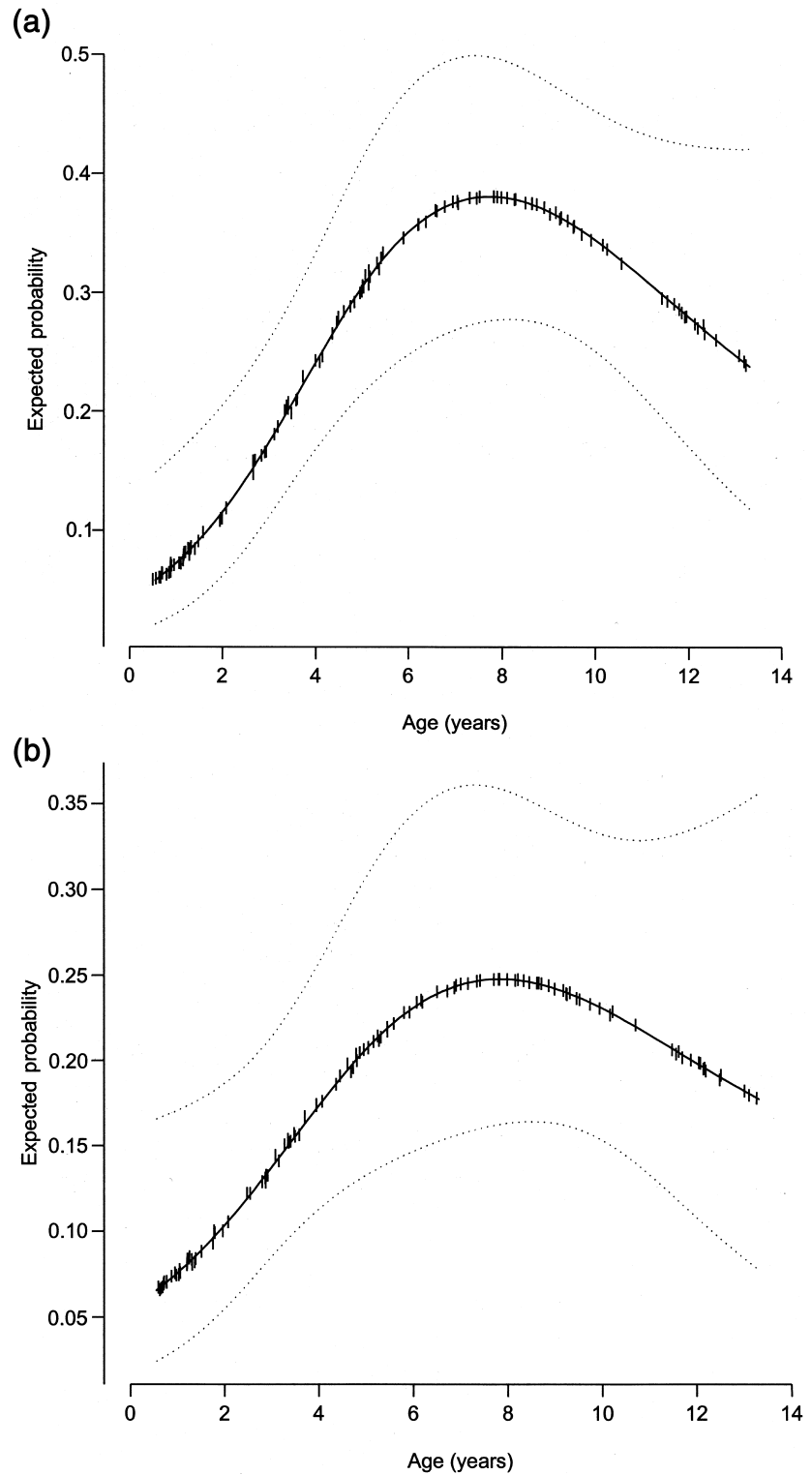


Fig. 2. (a) Expected probability for having specific IgE against age for outside fungi (*Cladosporium* and *Alternaria*). Distribution of individuals is shown as vertical lines. (b) Expected probability for having specific IgE against age for indoor fungi (*Aspergillus* and *Penicillium*). Distribution of individuals is shown as vertical lines.

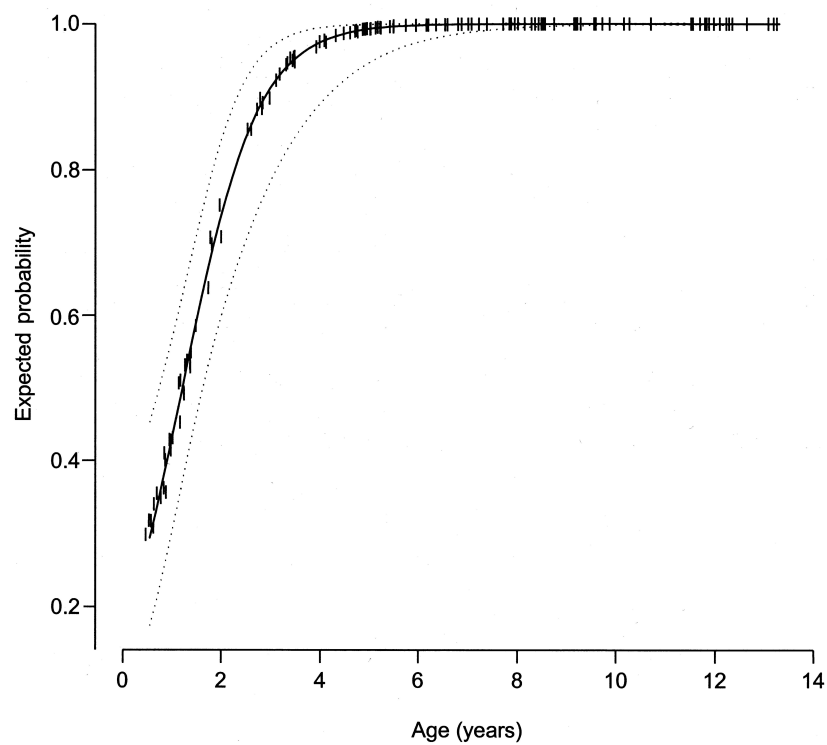


Fig. 3. Age-dependent distribution of specific IgE against age for house-dust mite. Distribution of individuals is shown as vertical lines.

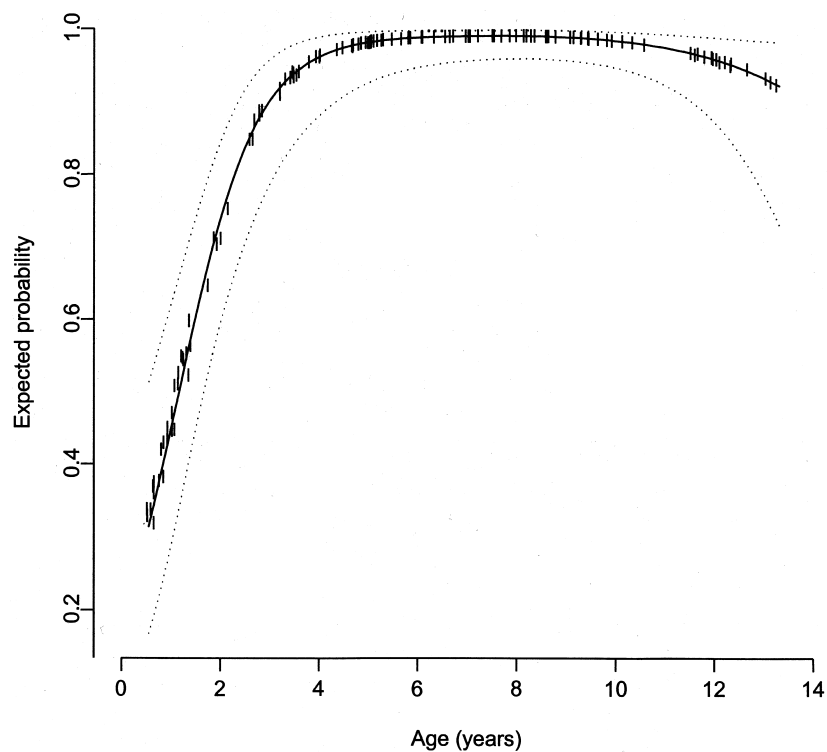


Fig. 4. Age-dependent distribution of the probability of having asthma. Distribution of individuals is shown as vertical lines.

linear relation with age, whereas specific IgE values against outdoor fungi, indoor fungi and house dust mite show significant non-linearity with age. Prevalence of specific IgE for *Cladosporium* ranked first, followed closely by *Aspergillus* and *Alternaria*. Specific IgE for indoor and outdoor fungi was related to the presence of specific IgE for aeroallergen and milk. We found that all children from the age of 4 years and older had allergy for house dust mite.

The high prevalence of specific IgE for fungi was also suggested in the study from Niemeijer; 10–40% prevalence of *Cladosporium* by skin testing during childhood as compared with older aged groups (0–5%) [2]. A previous study in adult patients showed age-dependent prevalence of skin test-positive for fungal allergy, with higher values in younger age-groups [9]. One study showed the prevalence of skin prick test positivity to *Alternaria* and *Cladosporium* in subjects with nasal and/or bronchial symptoms of suspected allergic cause in several European countries. The variation was between 3% and 20%, with an age range of 5–60 years [15].

Recently an age-dependent distribution for various aeroallergens, including moulds, was also reported for a desert environment (Kuwait) [16], showing high frequencies for fungal sensitization in these age-groups (3–17 years) followed by lower values at increasing age. Our observations indicate that high fungal sensitization is not unique for desert environments but is found, with similar frequencies, in milder and more humid climates. Furthermore, this study shows that sensitization to fungi reaches a maximum at 7.7–7.8 years, followed by a strong decline with age not found for the sensitization to house dust mite. Sensitization to house dust mite in this area shows high values till around 20–30 years of age, followed by gradual decline with age [9,16]. The clinical relevance of the higher sensitization rates to fungi at a younger age is in accordance with Tariq *et al.* [17], who found that in 4-year-old children, sensitization to *Cladosporium* and *Alternaria* (6%) correlated with the clinical diagnosis of asthma, eczema and rhinitis, respectively, and ranked third as the most common source for sensitization after house dust mite (11.9%) and grass pollen (7.8%).

The relationship between allergy for fungi and asthma had been reported earlier [18]. Sensitization to *Alternaria* allergens appeared to be significantly associated with asthma [19]. Sensitization to house dust mite and possibly fungi allergen played an important role in the relation between living in a damp home and childhood respiratory symptoms [20]. Recently factors associated with airborne fungal spore concentrations and health outcome in children were investigated. Indoor exposure to certain fungal genera in winter was a risk factor for asthma, atopy and respiratory symptoms in children [21]. Others concluded that peak flow variability in atopic children was associated with reported

fungi in the home [22]. The period prevalence of cough and upper respiratory symptoms was significantly higher in children living in houses with reported fungi. All these data suggest that sensitization to fungal allergens is of great importance in a young population. Fungi-sensitive patients should avoid exposure to areas where these micro-organisms may develop, such as fallen leaves, basements, cut grass, barns and wet-wooded areas. Since high humidity is the main factor in indoor fungal growth, it is important to avoid high levels of humidity indoors, i.e. more than 40% [23].

It is interesting that the prevalence of antibodies against *Alternaria* (14.6%) was almost similar to *Cladosporium* (18.2%). In several studies in the Netherlands it has been shown that *Cladosporium* is by far the most abundant in the outdoor air. Numbers of spores of *Alternaria* are generally much lower than those of *Cladosporium* (respectively 7500 and 600 000 spores per m³) [23]. This pattern of differences in numbers of fungal spores was consistent during 10 years in the Netherlands [7]. These observations on number of spores, together with corresponding prevalence of sensitization, suggest that *Alternaria* species have a strong capacity for sensitization compared with *Cladosporium*. This suggestion is in accordance with epidemiological clinical observations, which concluded that exposure to the aeroallergen *Alternaria* and environmental fungi was a risk factor for respiratory arrest in children and young adults with asthma [24,25]. *Alternaria* is the major allergen associated with the development of asthma in children raised in a semi-arid environment [10], while in a recent population-based study it was found that *Alternaria* was a risk factor for severe asthma [12]. The strong capacity of proteases of *Alternaria* in activating airway epithelial cells may be one of the factors favouring sensitization and clinical reactivity to this fungus [26].

The association between sensitization to aeroallergens and IgE to fungi appears to reflect the general (genetic) predisposition of the atopic airways for sensitization to multiple aeroallergens. Currently there is no explanation for the lower values for sensitization to fungal allergens above the age of 8 years, while sensitization to house dust mites remains high. Decline of exposure to fungal spores is certainly not a factor of importance. It can be speculated that the continuous high level of exposure to fungal spores may induce an increasing efficiency of the general defence against micro-organisms (bacteria, viruses and fungi) during development of the immunological response of the airways. An increased immunological defence with age may result in a more effective and rapid elimination of micro-organisms, thereby limiting the antigenic stimulation. Alternatively, it cannot be excluded that the lower values of IgE for fungi are the result of an induction of tolerance for abundantly inhaled fungal spores. It will be important to know if such lower values may be found in longitudinal studies, possibly reflecting natural

desensitizing mechanisms in atopic patients for fungal allergens. Considering the individual fungi, the correlation appears mainly due to the outdoor fungi. Exposure to any fungi indoors could have the potential to increase the risk of allergic sensitization to any allergen.

We did not find any significant difference for specific IgE against fungi between males and females, except for total IgE and age. This observation may be due to the selection bias: we studied an already sensitized population, thereby excluding the general observed different sensitization rates for males and females. Since this was a cross-sectional study, no definitive conclusions can be drawn about the influence of ageing on the prevalence of positive IgE test. Longitudinal studies are needed to show that our results are not caused by factors (environmental influence) other than age. Results as obtained from this study are only affected by criteria of having a positive IgE against one of the aeroallergens that were routinely tested, since only six children were excluded from this study.

In conclusion this study shows that sensitization to fungi is prevalent in childhood, with an age-dependent distribution reaching maximum values at 7.7–7.8 years, followed by a decline for all fungal sensitization with increasing age. The importance and relative contribution of fungal-induced manifestation of airway diseases, compared with the other allergens, remains to be established.

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