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Original Article

Mould Sensitisation among Bakers and Farmers with Work-related Respiratory Symptoms

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Abstract: Fungi belong to common allergens, which can be found both in occupational and non-occupational environment. The aim of the study was to determine the frequency and factors associated with mould allergy among bakers and farmers reporting work-related respiratory symptoms. The study group comprised 117 bakers and 83 farmers reporting work-related respiratory symptoms. Skin prick tests (SPT) with common, fungal and occupational allergens, estimation of serum total IgE level, spirometry, histamine test and specific inhalative challenge test with occupational allergens were performed in all subjects. The prevalence of hypersensitivity to fungal species was higher among farmers (32.5%) than bakers (16.2%). Positive SPT with mould allergens were found among 10.8% farmers and 6.8% bakers with occupational allergy. The fungi allergens giving positive SPT results most frequently were *Candida albicans*, *Levures mélanges, Aspergillus mix.* and *Charbons cerealiers* in both groups. Among mould allergens, hypersensitivity to *Aspergillus* genus was the most common in farmers and bakers group, while among *Saccharomyces − Candida albicans* sensitization was the most frequently detected. Mould hypersensitivity is related to occupational respiratory allergy, especially to asthma and rhinitis in farmers, and occupational asthma in bakers. Although sensitization to *Saccharomyces* was more frequent, similar correlation was not observed.

Key words: Bakers, Farmers, Mould allergy, Work-related symptoms

Introduction

Fungi belong to common allergens, which can be found both in occupational and non-occupational environment. The term “moulds” is a popular name used to identify filamentous fungi. They are part of air pollution and dust. Exposure to fungi can cause allergic diseases such as rhinitis and bronchial asthma, allergic bronchopulmonary mycoses as well as allergic alveolitis[1)]. All activities related to working in the land, like mowing, plowing, weeding, harvesting, gardening, etc. are associated with exposure to fungi allergens, which grow in the soil. Moulds are also the component of grain dust that is present at the workplace of farmers, grain elevator workers, millers, bakers and grain reloading workers. Undoubtedly, bakers and farmers are at risk of allergy to moulds. Lack of epidemiological data regarding occurrence of sensitisation due to occupational exposure to moulds justifies the need to carry out research among subjects exposed to those microorganisms. The aim of this study was to determine the frequency of fungal allergy, as well as factors associated with hypersensitivity to moulds among bakers and farmers reporting work-related respiratory symptoms.

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Subjects and Methods

The study group comprised 200 patients reporting work-related respiratory symptoms (upper airway symptoms: rhinitis, itching or nasal blockage, the lower airway symptoms: cough, dyspnoea or whistles were considered to be work-related if they were reported to worsen during the work shift and improve when away from work). The 117 bakers and 83 farmers were examined at the Department of Occupational Diseases. For the purpose of further analysis bakers and farmers were divided into two groups depending on the result of specific challenge test with occupational allergens. The first group comprised 63 bakers and 22 farmers with occupational allergic respiratory disease (rhinitis and/or asthma). The second group consisted of 54 bakers and 61 farmers reporting work-related respiratory symptoms but with negative specific challenge test in whom work-exacerbated asthma and/or rhinitis were recognized.

The Regional Bioethical Committee approved the study protocol. All the participants gave their consent prior to the study.

Questionnaire

The subjects were administered a validated questionnaire. It included a history of respiratory and skin symptoms, personal and family history of atopy, exposure to pet allergens at home, medication use and smoking habits.

Skin prick tests (SPT)

SPT were performed on the volar part of the forearm with a standard, commercially available battery of common allergens including tree and grass pollens, *Dermatophagoides pteronyssinus*, *Dermatophagoides farinae*, *Acarus siro*, *Thyrophagus putrescentiae*, *Lepidoglyphus destructor*, *Euroglyphus manei*, feathers and fungal series: Moulds I (*Alternaria tenuis*, *Botrytis cinerea*, *Cladosporium herbarum*, *Culvularia lunata*, *Helminthosporium*, *Fusarium moniliforme)*, Moulds II (*Aspergillus fumigatus*, *Mucor mucedo*, *Penicillium notatum*, *Pullularia pullulans*, *Rhizopus nigricans*, *Serpula lacrimans*), *Alternaria tenuis*, *Aspergillus fumigatus*, *Botrytis cinerea*, *Candida albicans*, *Trichophyton mentagrophytes*, *Cladosporium herbarum*, *Fusarium moniliforme*, *Helminthosporium halodes*, *Mucor mucedo*, *Penicillium notatum*, *Pullularia pullulans*, *Rhizopus nigricans*, *Serpula lacrymans*, *Curvularia lunata*, *Phoma betae*, *Neurospora sitophila*, *Alternaria sp.*, *Aspergillus mix.*, *Cladosporium sp.*, *Penicillium mix.*, *Levures melangees*, *Charbons cerealiers* (Allergopharma, Germany; Stallergrnes, France). The bakery series [oatmeal, wheat, corn, barley and rye flour (Allergopharma, Reinbek, Germany)] and farmer occupational allergens [mixture of grain, hay, horse epithelium, swine epithelium, sheep, goat, rabbit epithelium, fur, barley, corn, oat, rye, wheat, cereals, straw (Allergopharma, Germany; Stallergens, France)] were also tested.

The negative control was a diluted allergen and the positive control was a 1 mg/ml histamine dihydrochloride solution (Allergopharma, Germany). The largest wheal diameter was assessed after 15 min. A weal diameter of 3 mm or more and equal or greater than half of that formed by histamine was defined as positive, indicating sensitization. Subjects avoided antihistamines or antidepressants for at least 72 h before testing.

Total IgE

Total serum IgE was evaluated using the ImmunoCap-100 System (Phadia, Uppsala, Sweden). Total IgE level > 100 KU/L was considered as elevated.

Inhalation challenge tests

Specific inhalative challenge tests (SICT) with wheat flour, rye flour and improvers (mixed for the SICT) among bakers and with mixed farmer's material [grain, hay, straw, animal fur (pig, cow), bird feathers (chickens, geese)] among farmers were performed in a work-site simulation setting (room space 6 m^2 with temperature 22–25°C) using the patient's own samples. Baker's materials were delivered by the employer, while farmers brought their samples individually. The mycological contamination of test materials used for the SICT was not performed.

The patient was sifting approximately 500 g of materials for 30 min or until asthmatic reaction symptoms appeared. Potato flour was used as placebo, the test was performed on the previous day before specific challenge.

The patients who did not show significant (≥20%) fall in FEV_1 during the challenge test underwent a repeated challenge test for two hours on the next day. In patients with changes in FEV_1 ranging between 10 and 20% after that challenge, the exposure was prolonged up to three hours.

The study participants did not receive any systemic or local medication. Inhaled short-acting β_2-agonists was stopped at least 6 h before the study, inhaled long-acting β_2-agonists − 48 h, inhaled steroids − 5 d, systemic steroids − 14 d. Antihistamine medications 7 − 42 d before challenge test depending on the time of acting. Patient's oral statement determined complying with such recom-

mendations.

Symptom score

The number of sneezes and the degree of mucosal oedema, rhinorrhea and itching were evaluated. Total symptom score (SS) ranged from 0 to 8 and represented the sum of the scores for sneezing (0 sneezes − 0 points, 1–4 sneezes − 1 point, >4 sneezes − 2 points), rhinorrhea (none − 0 points, mild − 1 point, abundant − 2 points), mucosal oedema (none − 0 points, mild − 1 point, nasal block − 2 points) and itching (none − 0 points, itching of the nose or throat − 1 point, itching of the nose and throat − 2 points).

Pulmonary function

Resting spirometry (Vicatest 2A, Mijnhardt, The Netherlands) was performed in all subjects reporting chest symptoms. Bronchial response was measured by serial monitoring of Forced Expiratory Volume in one second (FEV_1) and Peak Expiratory Flow Rate (PEFR) before and 5 min, 30 min, 1 h, 2 h, 4 h, 6 h and 24 h after the provocation.

Histamine challenge was performed according to Cockroft[2]. Bronchial response was measured by FEV_1 monitoring. The non-specific bronchial hyperreactivity was evaluated on the day before the SICT and 24 h after the test.

Nasal lavage and challenge procedure

All the procedures were performed according to the 'nasal pool' method. Nasal washings were collected before the SICT and 4 and 24 h afterwards, the washings processing is described in detail elsewhere[3].

Diagnostic criteria

Occupational allergic rhinitis was recognized in subjects reporting work-related nasal symptoms with positive nasal response to specific challenge test i.e. total score of more than 3 points and significant increase in total count and proportion of eosinophils (twofold increase and eosinophilia at least 5% after challenge) in nasal lavage fluid.

Occupational asthma was recognized in subjects reporting work-related chest symptoms, for whom a SICT induced significant bronchial response (at least a 20% decrease in FEV_1) − early or dual asthmatic reaction, or a threefold increase in non-specific bronchial hyperreactivity accompanied by increase sputum eosinophilia (at least 3% after challenge)[4].

Statistical analysis

Continuous variables were expressed as mean values ± standard deviations (SD) while the nominal variables, as numbers and percentages. To identify the associated factors of hypersensitivity to fungi, mould and *Saccharomyces* allergens, the odds ratios (OR) and their 95% confidence intervals (CI) were calculated (Statistica 10). This analysis concerned the two groups of subjects who were sensitized and non-sensitized to at least one fungi allergen. The p value below 0.05 was adopted as the reference for selecting significant associated factors.

Results

Among farmers in 20 cases (24.1%) occupational asthma and in 17 cases (20.5%) occupational rhinitis were recognized whereas amid bakers in 46 subjects (39.3%) occupational asthma and in 54 cases (46.2%) occupational rhinitis were diagnosed. The characteristic of the study group is presented in Table 1. The average duration of symptoms among farmers was 11.2 ± 9.7 yr, while in bakers 6.1 ± 6.9 yr. First symptoms appeared after 16.1 ± 9.8 and 11 ± 8.7 yr of professional work, respectively.

The results of SPT to common, occupational and fungal allergens are presented in Tables 2, 3 and 4. A total of 55.4% of farmers and 64.9% of bakers had at least one positive allergy skin test response to common allergens, while sensitization to occupational allergens was detected among 33.7% farmers and 63.2% bakers. The prevalence of hypersensitivity to all tested fungal species was higher among farmers (32.5%) than in bakers group (16.2%). Positive SPT with mould allergens was found among 9 farmers (10.8%) and 8 bakers (6.8%) with occupational allergy. The most frequent fungi allergens giving positive SPT results were *Candida albicans*, *Levures mélanges*, *Aspergillus mix*. and *Charbons cerealiers* in the group of farmers as well as bakers.

The average total IgE level was elevated in both baker's and farmer's groups [182.8 ± 158.2 (min. 10, max. 871) KU/l and 215.8 ± 248 (min. 10, max. 1,000) KU/l respectively] and what is more it was higher among subjects with occupational allergy [283.3 ± 203.3 (min. 11, max. 871) KU/l and 236.7 ± 264.9 (min. 10, max. 1,000) KU/l, respectively] (Not shown in table).

Occupational respiratory allergy among farmers (OR=7.75) including asthma (OR=6.33) as well as rhinitis (OR=5.9) was more likely to be found in farmers sensitized to moulds (the univariate analysis, Table 5), while only occupational asthma was associated with hypersen-

Table 1. The characteristic of farmers (n=83) and bakers (n=117) with work-related respiratory symptoms participating in the study

	Farmers						Bakers					
	Occupational respiratory allergy n=22			Work-exacerbated respiratory symptoms n=61			Occupational respiratory allergy n=63			Work-exacerbated respiratory symptoms n=54		
	Mean	SD	n (%)	Mean	SD	n (%)	Mean	SD	N (%)	Mean	SD	N (%)
Age (yr)	46.1	7.9		48.7	8.4		34.9	9.9		39.7	13.4	
Gender: male			14 (63.6)			32 (52.5)			54 (85.7)			42 (65.4)
Gender: female			8 (36.4)			29 (47.5)			9 (14.3)			12 (34.6)
Currently smoking			3 (13.6)			8 (13.1)			9 (14.3)			13 (30.8)
Smoking in the past			8 (36.4)			16 (26.2)			28 (44.4)			19 (23.1)
Family history of atopy			5 (22.7)			20 (32.8)			18 (28.6)			13 (19.2)
Duration of symptoms (yr)	11.3	8.6		11.2	10.1		5.6	5.1		6.8	8.6	
Latency period till symptoms onset (yr)	16.1	10.4		16.6	9.7		9.7	7.2		12.6	10.1	
Cough			16 (72.7)			53 (86.9)			45 (71.4)			41 (76)
Dyspnoea			22 (100)			57 (93.4)			46 (73)			50 (92.6)
Rhinitis			17 (77.3)			48 (78.7)			54 (85.7)			40 (74.1)
Skin symptoms			7 (31.8)			13 (21.3)			14 (22.2)			16 (29.6)

Table 2. The results of skin prick tests to common allergens in the study groups of farmers (n=83) and bakers (n=117) with work-related respiratory symptoms

Positive skin prick tests with:	Farmers		Bakers	
	Occupational respiratory allergy n=22	Work-exacerbated respiratory symptoms n=61	Occupational respiratory allergy n=63	Work-exacerbated respiratory symptoms n=54
	n (%)	n (%)	n (%)	n (%)
At least one common allergen	16 (72.7)	30 (49.2)	49 (77.8)	27 (61.5)
Dermatophagoides farina	10 (45.5)	14 (23)	21 (33.3)	10 (30.8)
Dermatophagoides pt.	11 (50)	14 (23)	25 (39.7)	11 (23.1)
Feathers	1 (4.5)	1 (1.6)	1 (1.6)	–
Grass pollen	4 (18.2)	2 (3.3)	18 (28.6)	3 (11.5)
Tree pollen I[a]	3 (13.6)	2 (3.3)	17 (26.9)	3 (7.7)
Tree pollen II[b]	4 (18.2)	1 (1.6)	16 (25.4)	5 (11.5)
Weeds	9 (40.9)	3 (4.9)	15 (23.8)	8 (19.2)
Acarus siro	9 (40.9)	17 (27.9)	18 (28.6)	8 (23.1)
Thyrophagus putrescentiae	10 (45.5)	14 (23)	18 (28.6)	8 (15.4)
Lepidogryfus destructor	10 (45.5)	15 (24.6)	17 (26.9)	8 (23.1)
Euroglyphus mannei	4 (18.2)	6 (9.8)	8 (12.7)	–

[a] alder, hazel, poplar, elm, willow. [b] bird, beech, oak, plane.

sitivity to mould allergens among bakers (OR = 4.77). Similar results were obtained after analysis for all fungal species, however for single *Saccharomyces* allergens the association was not confirmed.

Discussion

Allergy to moulds, especially the occupational one, is insufficiently investigated. Occupational exposure to fungi occurs in bakeries during the production of bread, as well as in agriculture. Farmers are an occupational group that is especially affected by inhalation large amounts of micro-

Table 3. The skin prick test results to occupational and fungi allergens in the group of farmers (N=83)

Positive skin prick tests with:	Work-related respiratory symptoms n=83 (100%)	Occupational respiratory allergy n=22 (100%)	Work-exacerbated respiratory symptoms n=61 (100%)
	n (%)	n (%)	n (%)
Occupational allergens	28 (33.7)	12 (54.5)	16 (26.2)
Mixture of grain	6 (7.2)	4 (18.2)	2 (3.3)
Hay	10 (12.1)	7 (31.8)	3 (4.9)
Horse epithelium	3 (3.6)	3 (13.6)	0
Swine epithelium	5 (6)	5 (22.7)	0
Sheep	3 (3.6)	2 (9.1)	1 (1.6)
Goat	3 (3.6)	2 (9.1)	1 (1.6)
Rabbit epithelium	3 (3.6)	2 (9.1)	1 (1.6)
Fur	4 (4.8)	3 (13.6)	1 (1.6)
Barley	10 (12)	5 (22.7)	5 (8.2)
Corn	1 (1.2)	1 (4.5)	0
Oat	11 (13.3)	5 (22.7)	6 (9.8)
Rye	5 (6)	3 (13.6)	2 (3.3)
Wheat	8 (9.6)	5 (22.7)	3 (4.9)
Cereals	10 (12)	6 (27.3)	4 (6.6)
Straw	9 (10.8)	6 (27.3)	3 (4.9)
With mould allergens	**14 (16.9)**	**9 (40.9)**	**5 (8.2)**
Moulds I*	7 (8.4)	5 (22.7)	2 (3.3)
Moulds II**	8 (9.6)	4 (18.2)	4 (6.6)
Alternaria tenuis	3 (3.6)	2 (9.1)	1 (1.6)
Aspergillus fumigates	2 (2.4)	2 (9.1)	0
Botrytis cinerea	1 (1.2)	1 (4.5)	0
Trichophyton mentagrophytes	1 (1.2)	1 (4.5)	0
Cladosporium herbarum	1 (1.2)	1 (4.5)	0
Fusarium moniliforme	0	0	0
Helminthosporium halodes	1 (1.2)	1 (4.5)	0
Mucor mucedo	2 (2.4)	2 (9.1)	0
Penicillium notatum	2 (2.4)	2 (9.1)	0
Pullularia pullulans	2 (2.4)	2 (9.1)	0
Rhizopus nigricans	2 (2.4)	2 (9.1)	0
Phoma betae	1 (1.2)	1 (4.5)	0
Alternaria sp.	2 (2.4)	1 (4.5)	1 (1.6)
Aspergillus mix.	4 (4.8)	2 (9.1)	2 (3.3)
Cladosporium sp.	2 (2.4)	1 (4.5)	1 (1.6)
Penicillium mix.	2 (2.4)	2 (9.1)	0
Saccharomyces	**16 (19.3)**	**7 (31.8)**	**9 (14.8)**
Candida albicans	13 (15.7)	6 (27.3)	7 (11.5)
Levures melanges	7 (8.4)	3 (13.6)	4 (6.6)
Basidiomycota	**5 (6)**	**3 (13.6)**	**2 (3.3)**
Serpula lacrymans	0	0	0
Curvularia lunata	1 (1.2)	1 (4.5)	0
Neurospora sitophila	1 (1.2)	1 (4.5)	0
Charbons cerealiers	4 (4.8)	2 (9.1)	2 (3.3)

* Alternaria tenuis, Botrytis cinerea, Cladosporium herbarum, Culvularia lunata, Helminthosporium, Fusarium moniliforme. ** Aspergillus fumigatus, Mucor mucedo, Penicillium notatum, Pullularia pullulans, Rhizopus nigricans, Serpula lacrimans.

Table 4. The skin prick test results to occupational and fungi allergens in the group of bakers (N=117)

Positive skin prick tests with:	Work-related respiratory symptoms n=117 (100%)	Occupational respiratory allergy n=63 (100%)	Work-exacerbated respiratory symptoms n=54 (100%)
	n (%)	n (%)	n (%)
Occupational allergens	74 (63.2)	55 (87.3)	19 (35.2)
Wheat flour	52 (44.4)	42 (66.7)	10 (18.5)
Rye flour	51 (43.6)	43 (68.3)	8 (14.8)
Corn flour	14 (11.9)	13 (20.6)	1 (1.9)
Oat flour	10 (8.5)	10 (15.9)	0
Barley flour	19 (16.2)	16 (25.4)	3 (5.6)
α-amylase	10 (8.5)	6 (9.5)	4 (7.4)
With mould allergens	**11 (9.4)**	**8 (12.7)**	**3 (5.6)**
Moulds I*	3 (2.6)	3 (4.8)	0
Moulds II**	2 (1.7)	1 (1.6)	1 (1.9)
Alternaria tenuis	1 (0.9)	1 (1.6)	0
Aspergillus fumigates	1 (0.9)	0	1 (1.9)
Botrytis cinerea	0	0	0
Trichophyton mentagrophytes	1 (0.9)	0	1 (1.9)
Cladosporium herbarum	1 (0.9)	1 (1.6)	0
Fusarium moniliforme	1 (0.9)	1 (1.6)	0
Helminthosporium halodes	0	0	0
Mucor mucedo	1 (0.9)	1 (1.6)	0
Penicillium notatum	1 (0.9)	1 (1.6)	0
Pullularia pullulans	0	0	0
Rhizopus nigricans	0	0	0
Phoma betae	0	0	0
Alternaria sp.	1 (0.9)	1 (1.6)	0
Aspergillus mix.	3 (2.6)	3 (4.8)	0
Cladosporium sp.	2 (1.7)	2 (3.2)	0
Penicillium mix.	2 (1.7)	1 (1.6)	1 (1.9)
Saccharomyces	**13 (11.1)**	**8 (12.7)**	**5 (9.3)**
Candida albicans	10 (8.5)	6 (9.5)	4 (7.4)
Levures melanges	5 (4.3)	4 (6.3)	1 (1.9)
Basidiomycota	**3 (2.6)**	**3 (4.8)**	**0**
Serpula lacrymans	0	0	0
Curvularia lunata	0	0	0
Neurospora sitophila	0	0	0
Charbons cerealiers	3 (2.6)	3 (4.8)	0

* Alternaria tenuis, Botrytis cinerea, Cladosporium herbarum, Culvularia lunata, Helminthosporium, Fusarium moniliforme. ** Aspergillus fumigatus, Mucor mucedo, Penicillium notatum, Pullularia pullulans, Rhizopus nigricans, Serpula lacrimans.

organisms, including many species of moulds. Describing their work environment and exposure to moulds, the so-called "*field fungi*" − developing on plants in the outdoor environment (mainly *Alternaria sp.* and *Cladosporium sp.*) and "*storage fungi*", which grow on indoor stored raw plant and animal materials in the conditions of high temperature and humidity (mostly *Aspergillus sp.* and *Penicillium sp.*) could be distinguished. The dominant species isolated from samples of grain and grain dust were *Alternaria alternate, Penicillium* spp. and *Mucor mucedo*[5].

Bakery and pastry employees also have occupational contact with various species of fungi. The *Aspergillus candidus, A. niger, A. oryzae, Candida albicans, C. tropicalis*, *Trichophyton sp.* and *Puccinia graminis* are the most frequently detected. Klaustermeyer et al. described a case of baker's asthma caused by *Alternaria* and *Aspergillus*

species found in a bakery[6]. Additionally, in bakeries all kinds of fungal enzymes considered as occupational allergens are used (in order to increase the viscosity of flour, to improve its quality and modify the baking process). Also *Candida albicans* species may occur in the bakery humid microclimate.

The chronic respiratory diseases caused by inhaled exposure to flour and grain dust are the earliest known occupational diseases. It has been estimated that the incidence of occupational allergic rhinitis among bakers ranges from 18 to 29%, and bronchial asthma from 4.9 to 7%[7, 8]. Also the incidence of respiratory diseases among farmers is still considered to be high[9–11]. There is no published data about the relationship of hypersensitivity to moulds and occupational respiratory allergy among bakers and farmers. During our study we applied questionnaire to estimate the presence of allergic symptoms. Due to low specificity of the questionnaire survey in diagnosing occupational respiratory allergy, the obtained results have been clinically verified by performing SICT. Additionally, the diagnosis of mould allergy was based on the positive results of SPT.

Over 32% of farmers and 16% bakers with work-related respiratory symptoms in this study were sensitized to one or more fungal allergens. Generally the incidence of mould allergy ranges from 2 to 10% in the population[12, 13]. It is higher among atopic patients (44%) and extends to 80% among asthmatics[14–16].

In our study, 40.9% farmers and 12.7% bakers with occupational respiratory allergy were sensitised to mould allergens, compared to 8.2% farmers and 5.6% bakers with work-exacerbated respiratory allergy. Both among farmers and bakers positive SPT to *Aspergillus mix*. with mould series had been found the most frequently. On the contrary, in a study performed by Corey *et al.*, atopic subjects with rhinitis were most frequently sensitised to *Alternaria* (30%), *Helminthosporium* (28%), *Aspergillus* (21%) and *Penicillium* (10%)[14]. Also Mari *et al.* showed that 19% patients with respiratory symptoms reacted to at least one fungal extract, while the incidence of sensitization to *A. alternata* and *C. herbarum* was found respectively in 66% and 13% subjects[17]. Moreover, the adult chronic rhinitis patients with diagnosed bronchial asthma were most frequently allergic to *Alternaria* (47.1%) and *Cladosporium* (30.8%)[18]. Generally, the incidence of *A. alternata* sensitization within atopic patients varies between 3.6 and 39.4%[1].

There are few data concerning occupational mould sensitisation. In Prior *et al.* study about 12% young farmers had positive SPT to mould allergens. Positive SPT results to *Cladosporium* and *Aspergillus* were found respectively in 0.7% and 2.2% farmers[19]. Contrary to our data, in this group the hypersensitivity to moulds was found more frequently among asymptomatic farmers than in those with asthma or asthma and rhinoconjunctivitis[20]. Skjold *et al.* studied a cohort of apprentices bakers and found a positive SPT to moulds in 1.1% of apprentices[21], whereas in the group of 287 Polish apprentice bakers 3.1% were sensitized to moulds I and 4.2% to moulds II[22]. The prevalence of sensitization to mould mix (*Cladosporium*, *A. alternate*, *Fusarium*) and *Aspergillus* fumigatus among supermarket bakery workers was established to be between 7 and 3% respectively[23]. In our study, the increasing prevalence of fungal hypersensitivity among both farmers and bakers with occupational allergy was observed whereas in the groups without occupational allergic diseases the incidence ranges about describable in general population[12, 13].

It is worth noting that the hypersensitivity to *Saccharomyces* was also frequent in our patients (19.3% farmers and 11.1% bakers), in contrast to *Basidiomycota* allergy. In the report by Corey *et al.* 22% subjects were sensitized to *Candida* and 14% to *Curvularia*[14], while, in the Helbling *et al.* study, 9.8% of atopic subjects were sensitized to at least one *Basidiomycete* species[24].

It is important to note that diagnosis of fungal allergy entailed difficulties. First, there is a lack of standardized extracts, as even commercially available products show high variations in the allergen content. Secondly, the presence of cross-reactive allergens throughout to the close phylogenetic relationship of some fungal species has been described[1, 25]. There are various reasons for the insufficient quality of fungal extracts for skin prick testing. They vary considerably in their protein composition depending on sources used, on the other hand, growth conditions, protein extraction methods and storage conditions are critical to the quantity and even existence of individual allergens[1].

While there is a lot of clinical evidence of the higher prevalence of bronchial asthma in mould-sensitive patients, there is no evidence to support those observations among subjects with occupational respiratory allergy[18, 26]. Moulds such as *Alternaria*, *Aspergillus*, *Cladosporium, Helminthosporium, Epicoccum, Aureobasidium* and *Penicillium* have frequently been implicated in allergic asthma[28]. Bogacka *et al.* concluded that mould allergy may be a risk factor for bronchial asthma development[18]. In another study, the frequency of sensitization to the mould allergens was much higher among patients with asthma alone (45.8%) or both asthma and rhinitis (28.3%) than

Table 5. The analysis of the associated factors of hypersensitivity to fungi allergens in the study group (the univariate analysis)

Factor	Farmers sensitised to:					
	moulds		fungi		Saccharomyces	
	OR	95% CI	OR	95% CI	OR	95% CI
Work-related respiratory symptoms - Rhinitis	1.01	0.25–4.14	1.25	0.35; 4.5	1.53	0.29; 7.81
Work-related respiratory symptoms - Asthma (cough and/or dyspnoea)	0.8	0.08–8.02	0.69	0.17; 2.74	0.95	0.18; 5.11
Occupational respiratory allergy	**7.75*** **	**2.18–27.54**	**3.68****	**1.30; 10.38**	2.69	0.85; 8.60
Occupational rhinitis	**5.90****	**1.67–20.85**	3.0	0.99; 9.12	3.05	0.90; 10.34
Occupational asthma	**6.33****	**1.82–22.03**	**3.59***	**1.24; 10.4**	2.27	0.69; 7.46
Occupational asthma without occupational rhinitis	7.4	0.96–57.89	4.60	0.68; 31.31	1.44	0.13; 15.10
Occupational rhinitis without occupational asthma	11.2	0.57–220	3.07	0.17; 55.16	5.78	0.31; 106.98
Occupational asthma and occupational rhinitis	**5.0****	**1.38–18.14**	1.14	0.05; 28.30	0.50	0.02; 12.67
Factor	Bakers sensitised to:					
	Moulds		fungi		Saccharomyces	
	OR	95% CI	OR	95% CI	OR	95% CI
Work-related respiratory symptoms - Rhinitis	0.38	0.10; 1.46	0.90	0.27; 3.07	1.39	0.28; 6.88
Work-related respiratory symptoms - Asthma (cough and/or dyspnoea)	..	..	2.04	0.43; 9.78	2.86	0.34; 23.79
Occupational respiratory allergy	2.47	0.61; 9.98	2.08	0.72; 5.98	1.43	0.43; 4.71
Occupational rhinitis	1.45	0.41; 5.11	1.76	0.64; 4.80	2.02	0.61; 6.66
Occupational asthma	**4.77***	**1.18; 19.35**	2.48	0.90; 6.80	1.94	0.60; 6.28
Occupational asthma without occupational rhinitis	4.86	0.66; 35.72	2.29	0.37; 14.14	..	..
Occupational rhinitis without occupational asthma	..	..	1.07	0.19; 6.03	0.61	0.06; 5.87
Occupational asthma and occupational rhinitis	2.9	0.81; 10.30	2.41	0.44; 13.13	3.73	0.40–34.83

* $p<0.05$, ** $p<0.01$ and *** $p<0.001$.

those with rhinitis alone (11.8%). Mould sensitization was clearly associated with asthma rather than rhinitis[26]. Also Li *et al.* found an increase in prevalence of sensitization to moulds in patients with asthma alone, and those with both asthma and rhinitis[27].

The results of our study revealed that subjects sensitized to mould allergens developed statistically more frequently occupational respiratory allergy. Additionally, *Aspergillus sp.* was the species most frequently causing mould allergy. This relationship could be explained through the occurrence of moulds in materials used for conducting specific provocation challenge test especially in the farmer's one. Therefore, among subjects sensitized to moulds more often the positive results of SICT were found. This study has suggested an important role for mould sensitization in the development of occupational respiratory allergy but on the other hand, it could mean that the bronchial inflammation of asthma predisposes for sensitization to inhaled mould spores[26]. Moreover, similar dependence was not found for *Saccharomyces* allergens.

Association between work related symptoms and skin sensitisation to common allergens was confirmed in many studies; atopy plays an important role in the development of allergic diseases especially those resulting from exposure to high molecular weight allergens[29]. Positive SPT to common allergens including moulds was found to be a significant risk factor of occupational allergic rhinitis and bronchial asthma among apprentice bakers[22]. In Droste *et al.* study atopy was independent risk factors for work-related symptoms[30].

The limitation of the present study is that the variables found to be significant in the univariate analysis were not included in the logistic regression model as the sub-groups were not numerous. Furthermore, during the study, the mould allergy was confirmed by skin prick testing only.

Conclusions

Results of our study indicate that 32.5% of farmers and 16.2% bakers with work-related respiratory symptoms are sensitised to fungal allergens. Among mould allergens, hypersensitivity to *Aspergillus* genus, especially *A. fumigatus* was the most common in farmers and bakers group, while among *Saccharomyces* − *Candida albicans* sensitisation was the most frequently detected. Our recent findings indicate that mould hypersensitivity is related to occupational

respiratory allergy, especially to asthma and rhinitis among farmers, and occupational asthma in bakers. Although sensitisation to *Saccharomyces* was more frequent, similar correlation was not observed.

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