

Mouldy houses influence symptoms of asthma among atopic individuals

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Summary

Background The influence of current levels of indoor fungi on asthma is a controversial issue that needs to be resolved in order to advise patients appropriately.

Objective To assess the seasonal variation in indoor fungal levels and the impact of these levels on asthma among mould-sensitized individuals.

Methods Thirty-five young adults with current asthma and sensitization to fungi were visited four times over 1 year. At each home visit a questionnaire was administered and samples of dust and air were collected. Participants also recorded information on symptoms, peak expiratory flows (PEF) and medication use. Dust samples were analysed for house dust mite allergen (Der p 1) and total fungal biomass (ergosterol). Total and genus-specific fungal propagules were identified in air samples. Seasonal variation in allergen levels and significant independent effects of fungal levels on peak flow variability (PFV) were identified by repeated measures analysis of variance.

Results Significant seasonal variations were observed in viable airborne fungi, ergosterol levels in the floor dust and PFV. PFV correlated significantly with symptom scores and the dose of reliever medication. PFV was also significantly associated with smoking and visible mould. The association between visible mould and PFV was independent of season, smoking and the dose of reliever medication. However, there was no association between total fungi, specific fungi or ergosterol and PFV. Der p 1 levels had no significant influence on asthma, even in HDM-sensitized individuals.

Conclusions Mouldy homes adversely influence asthma in asthmatics sensitized to fungi.

Keywords ergosterol, fungi, house dust mites, indoor allergens, indoor environment

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Introduction

There is accumulating evidence that exposure to mould is associated with asthma. Damp conditions in homes have repeatedly been shown to be associated with respiratory symptoms [1, 2], although the exact causal pathway is yet to be established. Dampness encourages fungal growth and it may be this fungal exposure that causes respiratory symptoms. Visible mould, which is a predictor of levels of fungi in the environment [3], has been found to be associated with respiratory symptoms [2] and a dose–response relationship has been described between the number of mouldy sites and the risk of respiratory symptoms. Only a few studies have combined aero biological and clinical data to assess the impact of fungi on respiratory health. Among them, some have shown positive results [4–6], while the others have not [7, 8].

Many methodological problems prevent firm conclusions being derived from the available studies. The majority of the studies were cross-sectional studies or longitudinal studies carried out over a short time, and the results could have been confounded by the season. Each season features different atmospheric conditions and changes in other potential covariates, which are difficult to adequately measure and take into account in the analysis, other than by repeated data collection over all four seasons. This would allow for the seasonal variation in asthma [9–12] and fungi [13–16] to be taken into account when examining the association between them. Confounding effects caused by the seasonal variation in other allergens such as house dust mites (HDM) is another issue that has not yet been examined. Most of the studies used questionnaires rather than objective environmental data as the sole method of assessment of exposure and outcome, possibly biasing the results through misclassification.

We have previously found viable airborne fungal propagules and dust ergosterol levels, which are an indicator of the cumulative exposure to fungi, to be significantly associated with features of asthma in a cross-sectional study that was carried out on a community sample in Melbourne, Australia [17].

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Season was a significant predictor of both ergosterol and viable airborne fungal propagules in this study, hence it may have confounded the association between indicators of fungal exposure and asthma. Therefore, we carried out a cohort study over 1 year to more closely examine the association between fungal exposure and asthma among sensitized subjects. Our aims were:

- To confirm the seasonal variation of indoor fungi in Melbourne homes.
- To determine whether current fungal exposures were related to manifestations of asthma in sensitized individuals once the confounding effects due to season and HDM allergen levels were taken into account.

Methods

Subjects

This was a follow-up study to the European Community Respiratory Health Survey (ECRHS) in Melbourne, for which the sampling procedures have already been published [18]. All participants with current asthma and sensitization to fungi in 1996 were invited to participate in this longitudinal study.

Definitions

Sensitization to an allergen was defined as having a maximum weal diameter of 3 mm or more to the allergen in the skin prick test (SPT), with a negative reaction to saline (control). The allergen extracts used were *Dermatophagoides pteronyssinus*, *Cladosporium herbarum*, *Aspergillus fumigatus*, *Alternaria tenuis*, *Epicoecum nigrum* and *Penicillium*, with positive (histamine) and negative (saline) controls.

Current asthma was defined as having experienced wheeze during the last 12 months, plus having bronchial hyper-reactivity (BHR) (19) in the 1996 follow-up study. BHR was defined as a $PD_{20} FEV_1 < 2$ mg methacholine in an inhalation bronchial challenge test.

Occupations of the participants were classified according to the Australian Standard Classification for Occupations (ASCO). Managers, administrators (ASCO group 1) and professionals (ASCO group 2) were reclassified for the study as class 1 and the remaining ASCO groups as class 2.

Data collection

All participants in the study ($n = 39$) were visited at home four times over a period of 1 year in 1997/1998. Floor dust and air samples were collected from bedrooms at each visit. A validated home visit report [20] and respiratory questionnaire were administered to gather information on residential characteristics, smoking and respiratory symptoms within each season. Bedroom air temperature and relative humidity were recorded at the same visit using a digital thermo-hygrometer (Type THG-388, RS Components, China) and a standardized procedure [21]. Absolute humidity for each house was calculated using the measurements of temperature and relative humidity. The participants were requested to complete diary cards for 14 days after each home visit and to post these back to the study centre.

Diary cards

These were self-administered forms on which participants recorded peak expiratory flows (PEF), symptoms, asthma

medication use and time spent at home. Symptoms of asthma, i.e. wheeze, cough, sleep and restriction of activities were assessed on a scale from zero (asymptomatic) to three (severe). These scores were added together to derive the daily total symptom score for each day for each individual.

Participants were provided with PEF meters (Allersearch, Melbourne, Australia) and trained to record the highest of three peak expiratory flow rates twice a day on waking each morning and at bedtime. Daily peak flow variability (PFV) was defined as the difference between morning and evening PEF on each day, expressed as a percentage of the higher figure [22].

Two clinical indices were calculated from the information extracted from the diary cards. They were the average daily total symptom score and the average daily PFV for each 14-day period.

Collection of air and dust samples from the bedroom floor

Samples of dust and air were collected from the bedroom floor of the study subjects' homes using a standard protocol [21, 23].

Measurement of levels of HDM allergens in dust

Dust samples were analysed for the major HDM allergen Der p 1 [24] using a standard enzyme-linked immunosorbent assay (ELISA) and a standardized procedure, for which the details have already been published [21]. Allergen levels were expressed as mcg/g of fine dust.

Measurement of cumulative fungal levels in floor dust

The fungal membrane lipid ergosterol was assessed as an indicator of total fungal biomass in dust samples, using a modification of the technique of Martin et al. [25], the details of which we have already published [3]. Results were expressed as mcg ergosterol/g of fine dust. Ergosterol assays were carried out in two batches. The lower limit of detection of the assay for ergosterol was 0.5 mcg/g, and approximately 10% of dust samples fell below this level in the first batch (i.e. first two seasons of the study: winter and spring). However, in the second batch that included the dust samples collected during summer and autumn, ergosterol levels below 10 mcg/g were not resolved, and approximately 50% of dust samples fell below the limit of detection due to equipment failure. Re-examination of the remaining samples from the previous batch showed the values above 10 mcg/g to be well correlated with the previous findings. Hence, we are confident that the measurements of ergosterol are valid, except that the minimum detection level changed between the two time points. In order to overcome the problem of missing data, the mid-point between zero and minimum detection level for the specific season was assigned for the samples with ergosterol levels below the detection level, when ergosterol levels were treated as an explanatory variable, but censored regression was used when ergosterol levels were treated as an outcome (see analysis section below).

Measurement of total and specific fungal levels in the air

The identity and abundance of viable fungal propagules suspended in the air at the time of sampling were estimated using a two-stage Andersen sampler (Model 10-800, Graseby/Andersen, Atlanta, GA, USA), utilizing potato dextrose agar plates and a standardized procedure [3]. Five genera, which have previously been identified as allergenic, i.e.

Cladosporium, *Alternaria*, *Epicoccum*, *Penicillium* and *Aspergillus*, were identified based on colony and spore morphology. All the other colonies were aggregated into one group as 'others'. Levels of airborne fungi were expressed as number of colony forming units per cubic meter of air (CFU/m³).

Analysis

The Statistical Analytical System package [26] and STATA [27] were used to analyse the data. The distributions of viable airborne fungal propagule counts, ergosterol levels, Der p 1 levels, PFV and symptom scores were positively skewed. Log transformation normalized the distributions of total fungal propagules, *Cladosporium*, other fungal propagules, ergosterol, Der p 1 and PFV.

The levels of viable airborne fungi, ergosterol and visible mould were used as indicators of fungal exposure. Levels of viable fungal propagules and ergosterol were analysed as continuous variables to examine their influence on the PFV.

Temperature, relative and absolute humidity, Der p 1 levels, presence of a cat in the household, smoking status and medication use were considered as possible confounders. The factors associated with PFV ($P < 0.2$) were controlled when examining the relevant associations.

To assess seasonal variation in indoor fungi, repeated measures analyses were performed to take into account the multiple and correlated measurements per household. Due to the frequency of non-detects for ergosterol, the assessment of seasonal variation in ergosterol levels used repeated measure analysis of variance for left censored (log) normal data using the xtintreg procedure of STATA.

Continuous outcomes, which were normally distributed when log transformed, were summarized by season with the geometric means and geometric standard deviations. These geometric means were compared across seasons using the SAS MIXED procedure incorporating season as a factor, allowing an unstructured correlation matrix between the repeated measurements, and applying an *F*-test. Binary outcomes were similarly modelled using the SAS GLIMMIX macro and applying a Wald test. Where outcomes could not be transformed to normality, Friedman's non-parametric two-way ANOVA with factors of season and household was applied. Assessment of

the relationships between fungal exposure and manifestations of asthma (and in particular PFV) were initially performed cross-sectionally for each season by linear regression and reported as regression coefficients representing dose-response slopes for continuous exposures or as ratios of geometric means for binary exposures. These relationships adjusting for the potentially confounding effects of season were assessed using repeated measures models as above, including the season effect and other potential confounders. Assessment of homogeneity of the cross-sectional relationships across seasons were performed by including the appropriate interaction terms in the repeated measures models.

Ethics

The study was approved by the Ethics Review Committee at The Alfred Hospital. Written informed consent was obtained from all participants.

Results

Characteristics of the study sample

A total of 39 current asthmatics who were sensitized to fungi participated in the study, which commenced in June 1997. The relevant characteristics of the 35 participants, who completed the study over four seasons, and their homes, are described in Table 1.

Eighty five per cent of the subjects were also sensitized to HDM. Few of the participants smoked (5.7%) and this did not change over the study period. Passive smoking exposure at home varied slightly across seasons but this variation was not statistically significant (Table 1). As expected, temperature, relative humidity and absolute humidity were clearly and significantly related to the season (Table 1), with both relative humidity and temperature higher in summer and autumn than winter or spring.

Forty per cent of the subjects claimed to have seasonal asthma. The asthma symptoms of participants were indeed more severe in autumn and winter (Table 2, Fig. 1). PFV and the use of preventer medication were greatest in winter. Wheeze, cough at night and exertional dyspnoea seemed to have similar

Table 1. Characteristics of the study subjects and their bedrooms

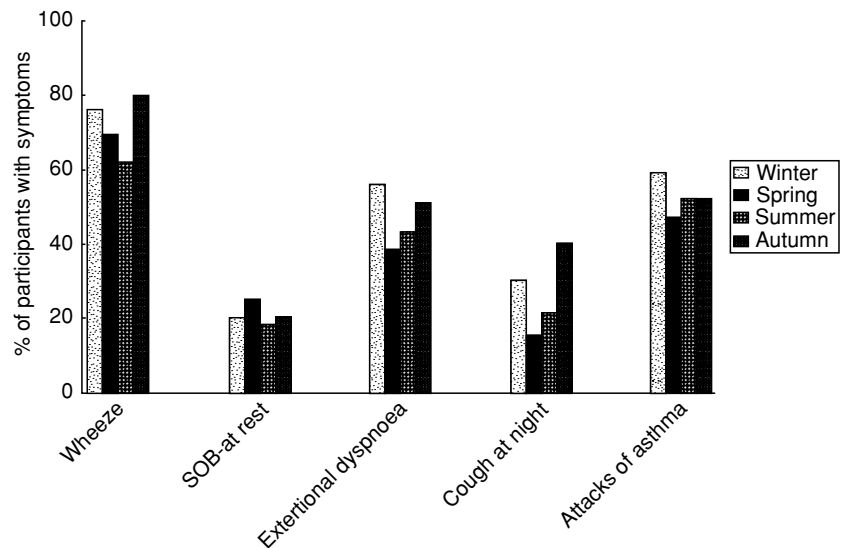
	Winter (<i>n</i> = 35)	Spring (<i>n</i> = 35)	Summer (<i>n</i> = 35)	Autumn (<i>n</i> = 35)	<i>P</i> *
Mean age (SD)†	38 (6.5)				
Gender (% male)	53.8%				
Australian born	82%				
Occupational class 1	53%				
Sensitized to HDM	85%				
Sensitized to cat	82%				
Prevalence of cats	22%	22%	20%	20%	0.91
Current smoker	5.7%	5.7%	5.7%	5.7%	1.02
Passive smoker: home	17.1%	14.3%	22.9%	20.0%	0.82
Temperature: bedroom (°C)	17	21	24	22	< 0.001
Relative humidity: bedroom (%)	54	50	58	57	< 0.001
Absolute humidity: bedroom (g/m ³)	6.4	7.8	10.8	9.4	< 0.001

*As described in the Analysis section, different methods of repeated measures analysis were used to derive the *P*-values depending on the distribution of the data. †SD: standard deviation.

Table 2. Seasonal variations in markers of asthma

	Winter (n = 35)	Spring (n = 35)	Summer (n = 35)	Autumn (n = 35)	P*
PFV%: GM (GSD)†	6.0 (1.8)	4.7 (2.2)	5.3 (1.9)	5.0 (2)	0.05
Night symptom score per day: M (R)‡	0 (0–0.7)	0 (0–1.2)	0 (0–0.4)	0 (0–0.6)	0.32
Day symptom score per day: M (R)	0.2 (0–1.9)	0.2 (0–2)	0.2 (0–2)	0.1 (0–0.6)	0.83
Total symptom score per day: M (R)	0.2 (0–2.6)	0.2 (0–3.2)	0.2 (0–2.2)	0.1 (0–2)	0.72
Use of preventer medication for asthma	29%	18%	24%	18%	0.71
Use of reliever medication for asthma	49%	49%	53%	49%	0.92

*As described in the Analysis section, different methods of repeated measures analysis were used to derive the P-values depending on the distribution of the data. †GM (GSD): geometric mean (geometric standard deviation). ‡M (R): median (range).

**Fig. 1.** Prevalence of asthma symptoms as reported in the respiratory questionnaire.

seasonal variation. The prevalence of these symptoms was higher in the autumn and winter than spring or summer. The prevalence of 'attacks of asthma' was highest in winter (Fig. 1). However, only the seasonal variation in PFV was statistically significant ($P = 0.05$). Paradoxically, there was hardly any variation in the use of reliever medication for asthma across seasons. We then examined these associations while accounting for any interaction between season and the self-reported seasonal asthma. There was no change in the above findings.

PFV was significantly correlated with the total symptom score ($r = 0.3$, $P < 0.001$) and frequency of dosing of reliever medication ($r = 0.3$, $P < 0.001$) but not with frequency of dosing of preventer medication. Total symptom scores were correlated with frequency of dosing of both reliever medication ($r = 0.7$, $P < 0.001$) and preventer medication ($r = 0.2$, $P = 0.02$). The correlation between the frequency of dosing of preventer and reliever medication was also statistically significant. ($r = 0.4$, $P < 0.001$).

The PFV among current smokers was twice (12%) that of non-smokers (6%) ($P = 0.02$). Temperature, relative humidity, absolute humidity and cat ownership were not related to PFV.

Seasonal variation in fungi and Der p 1

Over the year, the numbers of indoor total viable airborne fungal propagules ranged from 0 to 6105 CFU/m³, with a median level of 421 CFU/m³. Among the specific airborne

fungi investigated in this study, the most common genera in all four sampling periods were *Cladosporium* and *Penicillium*. Ergosterol levels ranged from 0 to 165 mcg/g of dust with one outlier of 1346 mcg/g of dust and a median ergosterol level of 15.8 cg/g of dust.

Table 3 describes the seasonal variation in fungi and Der p 1. Seasonal variations in the levels of total fungi and all the specific fungi except *Aspergillus* were observed to be statistically significant. Levels of total fungal propagules, propagules of *Cladosporium* and *Penicillium* were highest in summer, being 1.8-, 2.6- and 4-fold, respectively, higher than the lowest levels observed, which were in winter for all three indices. Levels of *Aspergillus* and *Alternaria* propagules were highest in autumn. The highest average ergosterol level was observed in winter, which was about 2.4-fold compared with the lowest level in spring.

A reduction in the proportion of houses with current visible mould was observed over the study period but this variation was not statistically significant. Seventeen participants (48%) admitted to having ever observed mould in their bedrooms when asked at the beginning of the study but two of these subsequently denied it.

Der p 1 levels also varied across seasons, with the highest level being in autumn, but was only 1.4-fold higher than the lowest level observed in winter (Table 3). The association between Der p 1 levels and season was not statistically significant.

Table 3. Seasonal variations in fungi, Der p 1 and asthma

	Winter	Spring	Summer	Autumn	P*
Total fungi CFU/m ³ : GM (GSD)†	392 (96)	562 (71)	655 (70)	448 (67)	< 0.001
Cladosporium CFU/m ³ : GM (GSD)	133 (92)	263 (92)	346 (91)	185 (92)	< 0.001
Penicillium CFU/m ³ : M (R)‡	37	75	150	37	0.01
<i>Aspergillus</i> percentage§	17%	18%	25%	40%	0.08
<i>Alternaria</i> percentage§	2.5%	2.5%	20%	35%	< 0.001
Other fungi CFU/m ³ : GM (GSD)	112	187	0	75	< 0.001
Ergosterol mcg/g of dust: GM (GSD)	8.0 (6.5)	3.3 (3.3)	5.2 (12.5)	7.4 (5.5)	0.04
Current visible mould	25.7%	14.3%	17.1%	8.6%	0.32
Der p 1 mcg/g of dust: GM (GSD)	12.8 (5)	13.4 (5)	14.6 (6)	17.0 (5)	0.41

*As described in the Analysis section, different methods of repeated measures analysis were used to derive the *P*-values depending on the distribution of the data. †GM (GSD): geometric mean (geometric standard deviation). ‡M (R): median (range). §Median levels of *Aspergillus* and *Alternaria* in all four seasons were 0.

Some significant correlations were observed between the levels of different fungi and ergosterol. The levels of *Penicillium* were positively correlated with levels of *Cladosporium* ($r = 0.4$, $P = 0.01$) and ergosterol ($r = 0.5$, $P = 0.004$) in spring. *Aspergillus* was positively correlated with ergosterol ($r = 0.3$, $P = 0.04$) in summer. *Aspergillus* was negatively correlated with *Cladosporium* ($r = -0.3$, $P = 0.01$) in autumn. Viable fungal propagules and ergosterol levels were not correlated with current visible mould, visible mould during the past 12 months or visible mould ever, and these findings were consistent across the seasons.

The influence of indoor fungi on the clinical activity of asthma

Visible mould was associated with a 1.5-fold increase in the PFV, which was statistically significant (95% CI = 1.03–2.3, $P = 0.02$). This association remained significant after accounting for the confounding effects of season, smoking status and the frequency of dosing of reliever medications. Season of measurement remained a significant explanatory factor for the PFV with all these factors in the model ($P = 0.01$).

The levels of total fungi, specific genera or ergosterol did not have any significant relationship to PFV (Table 4). Der p 1 levels

were not related to PFV, symptom scores or medication use, even among people who were sensitized to house dust mites.

The above associations were examined while accounting for the interactions between season and fungal indicators. The interactions were not significant and findings remained the same.

Discussion

We found significant seasonal variation in indoor viable airborne fungi, and ergosterol levels in floor dust, in this sample of Melbourne homes. Statistically significant seasonal variation in PFV was also observed. Visible mould was significantly associated with PFV but the levels of ergosterol, viable airborne fungi and Der p 1 were not associated with PFV.

The most commonly found indoor fungal genera in the homes of these asthmatics sensitized to fungi were *Cladosporium* and *Penicillium*. The prevalence and the seasonal variation of viable airborne fungi were similar to our previous findings in a cross-sectional study of a community sample of Melbourne homes, from which this sample was recruited [3]. Except in winter, almost half of the houses had levels of total viable fungal propagules above the threshold that has been suggested as hazardous to health by the World Health Organization [28]. The seasonal variation of ergosterol was also similar to our previous study, but the ergosterol levels were much higher in this sample of homes with asthmatics sensitized to fungi when compared with the entire sample. Higher levels of ergosterol have been observed in homes of people with sensitization to fungi compared with homes of people without sensitization [17].

The results of this study on seasonal variation in indoor fungi confirm the findings of a similar study carried out in the Latrobe Valley, which is located in a rural area of Victoria, 200 km south-east of Melbourne [29]. However, the levels of indoor viable airborne fungi were somewhat higher in the Latrobe Valley, which supports the hypothesis that homes in rural areas harbour higher levels of airborne fungal propagules than those in urban areas [30]. Another discrepancy between the two studies was the significant seasonal variation in levels of *Aspergillus* observed in Latrobe Valley, but not in the present study. This is probably related to the sample size being inadequate to detect a small variation when compared with the

Table 4. The association between indoor fungi and peak flow variability

Measurements of indoor fungi	Proportionate change in GM† of PFV (<i>P</i> *)	Proportionate change in GM† of PFV adjusted for smoking, reliever medication and season (<i>P</i> *)
Presence of visible mould	1.5 (0.02)	1.4 (0.02)
Ergosterol‡	1.002 (0.31)	1.003 (0.52)
Total fungi‡	0.99 (0.63)	0.99 (0.71)
<i>Cladosporium</i> ‡	1.2 (0.72)	1.1 (0.74)
<i>Penicillium</i> ‡	1.01 (0.53)	1.01 (0.62)
<i>Aspergillus</i> ‡	0.8 (0.21)	1.08 (0.73)
<i>Alternaria</i> ‡	0.5 (0.22)	0.61 (0.41)
Others‡	0.90 (0.14)	0.90 (0.12)

*As described in the Analysis section, different methods of repeated measures analysis were used to derive the *P*-values depending on the distribution of the data. †GM: geometric mean. ‡Increase with 10 units in the allergen levels.

larger study carried out in Latrobe Valley, which involved 80 households. Alternatively, this may also be related to the differences between the two areas in home construction or proximity to substrate on which fungi can grow.

The severity of asthma was greater in autumn and winter in this sample of current asthmatics, but only the association between PFV and season was statistically significant. Clinically significant magnitude of changes in PFV is not well defined, but 6% PFV has been suggested to correspond to mild asthma, with an increased PFV correlating with increased severity [31]. It should be noted that the PFV was highest in winter, which was the first study period and the training/learning effect causing higher variability in the peak flow rates may have partly contributed to this finding.

The lack of seasonal variation in the symptom score was surprising. This may be either related to the limited sensitivity of the symptom scale or to the increased use of preventer medication in winter. A peak in hospital admissions for asthma has been observed in autumn and winter in a number of studies [9–12]. The seasonal variation in asthma has been partly attributed to the meteorological conditions associated with each season [32]. Results of the present study did not support this hypothesis. None of the physical factors such as indoor temperature, relative humidity or absolute humidity were related to the PFV. However, the findings related to relative humidity, temperature and season were limited by the short duration of the measurement. The likely alternative hypothesis of the importance of respiratory viral infections was not specifically examined.

Reported past visible mould was associated with the PFV in this study. In contrast, current visible mould has been associated with asthma in adults [1, 33] and children [7, 8, 34]. In our study, the proportion of houses with current visible mould fell over the study period, which may be either related to measures taken to reduce visible mould or reporting bias as a result of being involved in the study. Hence, the lack of association between current visible mould and PFV may be partly explained by misclassification of exposure status.

We did not observe any association between viable indoor airborne fungal propagules or ergosterol levels in dust and PFV in this sample of young adults. In contrast to these findings, we have previously observed high levels of viable airborne fungi to be a risk factor for bronchial hyper-reactivity to methacholine and high levels of ergosterol to be a risk factor for wheeze within the last 12 months [17]. Similarly, physician-diagnosed asthma in children aged 7 to 14 years has previously been associated with levels of indoor *Penicillium* in winter [5]. In the same study, respiratory symptoms were higher with increased exposure to *Cladosporium* and total airborne spores. Outdoor fungal spores have also been observed to be positively related to asthma symptom scores and medication use in a small follow-up study of asthmatics aged 9 to 12 years [35]. Asthma is a heterogeneous condition that comprises distinct phenotypes with different clinical expressions in different age groups and each of these phenotypes may have different risk factors [36]. Hence, the discrepancies observed between studies with respect to the influence of viable airborne fungal propagules on different clinical expressions of asthma are perhaps not surprising. On the other hand, air sampling was carried out only over a 1-min period and one time point per season. Hence the high random variation in fungal propagules would have led to non-differential misclassification of the exposure status, which may have contributed to

the observed lack of association. Also, the lack of association between ergosterol levels and PFV may partly be related to the non-differential misclassification of the exposure status due to the assignment of an arbitrary value.

The seasonal variation in BHR among allergic asthmatic individuals has been attributed to the variation in allergen exposure [37]. Variation in indoor fungi and house dust mite allergen levels did not fully explain the seasonal variation in PFV observed in this study. The unexplained variation may be related to increased respiratory tract infections in winter but we did not have data to test this hypothesis.

There was no association between Der p 1 levels and PFV, even among subjects who were sensitized to house dust mites. Evidence is accumulating on the lack of importance of Der p 1 levels as a cross-sectional risk factor for clinical activity of asthma, especially in areas with very high Der p 1 levels [38]. Paradoxically, current Der p 1 levels have been found to significantly influence PD₂₀, log dose–response slope and peak expiratory flow variability in HDM-sensitive subjects reactive to methacholine [39] and to be significantly higher in the bedroom floors of severe compared with mild sensitized asthmatics [40]. However, these samples were recruited from asthma clinics and represented the severe end of the spectrum of disease in contrast to our community-based sample. Furthermore, although these patients lived in areas with quite high HDM exposure, their exposure was not nearly as high as in the studies that have not observed a clinical relationship with HDM exposure, including the current one. Hence, these findings may indicate that the severity of the disease modifies the association between current Der p 1 levels and asthma. Current exposure to Der p 1 may be important in clinical activity of asthma among severe asthmatics, but not at the community level. Alternatively, the almost uniform exposure to Der p 1 levels at very high levels as in our study may be sufficient to maintain maximum asthma symptoms in all susceptible asthmatics, so that a dose–response relationship may not be observed.

In conclusion, this study has demonstrated an association between visible mould and PFV in a group of asthmatics sensitized to fungi. Direct observation of growth of mould seems superior to any other available measures of environmental biomass. There is a need for further studies, especially of interventions to reduce the mould exposures. From a public health perspective, we would encourage asthmatics with sensitization to fungi to take measures to reduce their exposure to fungi. Frequent airing, regular use of kitchen exhaust fans and bedroom ceiling fans, removal of old wall-to-wall carpets, exclusion of pets, frequent vacuuming and cleaning visible mould patches have been suggested as potentially effective measures to reduce mould exposure [3].

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