Indoor Dust Allergen Levels in the Homes of Patients with Childhood Asthma: An Experience From Southwestern Iran

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ABSTRACT

Exposure to indoor allergens plays an important role in the etiology of asthma. This study was designed to quantify indoor allergens from homes of families that had at least one case of childhood asthma at home in a southwestern city of Iran. The relationship between the indoor allergen levels and home characteristics was also investigated.

Dust samples were collected from the bedrooms and the kitchens of 35 homes where children with persistent asthma were living. The levels of indoor allergens were measured by enzyme linked immunosorbent assay (ELISA).

Detectable amounts of mite, mouse and cockroach allergens were found in all evaluated places. None of our patients were exposed to a threshold concentration of indoor allergen for sensitizing at home. Regarding of mite allergens, the levels of Der f1 were significantly higher than Der p1 and a direct correlation was observed between living in an apartment and Der f1 levels. Moreover, Fel d1 (cat) and Bla g1 (cockroach) allergens were found in the children’s bedrooms more frequently than those in the kitchens. In this study, direct associations were obtained between Bla g1 allergen and the duration of occupancy and between Fel d1 and average home size. A total of 34.2% of the patients showed positive skin reactions to at least one of the tested allergens as 17.1% of them showed reactivity to D. pteronyssinus.

Proper controlling of cockroaches and mice by public health officials would be a practical approach to avoid inducing asthma or worsening the symptoms.

Keywords: Allergens; Asthma; House dust; Immunologic sensitization

INTRODUCTION

Sources of indoor allergens including house dust
mites, pets, mice, cockroaches and molds are the main risk factors in the pathogenesis of asthma.\textsuperscript{1,2} Pyroglyphid house-dust mites including Dermatophagoides pteronyssinus, Dermatophagoides farinae and Euroglyphus maynei are the major sources of allergens in homes. Mites are small arthropods that feed on human skin scales and are found in large numbers in beds, mattresses, carpets and furniture. Mite fecal pellets are a major source of allergens, which are classified into group 1 (Der p1 and Der f1) and group 2 (Der p2 and Der f2). Mites are very sensitive to low humidity and extreme temperatures.\textsuperscript{3,4}

The major cat allergen, Fel d1, is found in the saliva, skin and fur. This glycoprotein has charged polar groups that adhere to surfaces such as walls, carpets and furniture. This allergen can also readily be carried into homes without cats on cloths and shoes, and may persist for a time even after removing the cats; thus it is not easy to completely get rid of this allergen.\textsuperscript{5,6}

The house mouse is also a potential source of indoor allergens. A major protein present in mouse urine, Mus m1, becomes airborne and highly allergenic once the urine dries.\textsuperscript{7} Cockroaches can be found usually in kitchens with a sustained heating. Blattella germanica, producing Bla g1 and Bla g2 allergens, is one the most common cockroach species in homes.\textsuperscript{8} Although molds are primarily considered outdoor allergens, Aspergillus species can be detected in warm and humid indoor places. Asp f1, derived from Aspergillus fumigatus, is one of the common indoor allergens.\textsuperscript{9}

Indoor allergens are important factors in development and exacerbation of asthma, but there are a few reports from northern and southern coastal areas in Iran just focusing on house dust mite allergen levels.\textsuperscript{10,11} This study was designed to quantify the indoor allergens in the homes of families with at least one child suffering from persistent asthma in Shiraz (southwestern Iran). The relationship between the levels of each indoor allergen and home characteristics as well as the rate of patient sensitization to each allergen was also investigated.

MATERIALS AND METHODS

Thirty-five families who were living in Shiraz (a city in southwestern Iran) with at least one child with persistent asthma according to expert panel report 3 criteria\textsuperscript{12} were enrolled in this study. Written informed consents were provided by participants. The volunteer families were referred to the allergy clinic at Ali-Asgar Hospital, Shiraz University of Medical Sciences. The study protocol was approved by the Ethics Committee of the university (approval number: 3836).

Sampling was conducted in the main area of the city which was divided to five subdomains, each contains seven houses. A home environment questionnaire was filled to obtain information about the type of dwelling, age of the building, average size of home, damp stains, number of householders, number of rooms, length of residence, use of carpets in the bedroom and finding cockroaches during the previous year.

During winter 2013, two separate dust samples were collected from the children’s bedrooms (pillows, mattress, sheets, bedroom floors and carpets) and the kitchen (floor, around the cabinets and refrigerators).

Participants were asked not to clean the child’s bedroom or the kitchen and not to change the child’s bed sheets for one week prior to dust collection. House dust allergens were extracted by previously published protocol.\textsuperscript{13} The concentrations of allergens were determined in the extracted mixtures by commercial enzyme linked immunosorbent assay (ELISA) kits (Indoor Biotechnologies, Clwyd, UK) according to the manufacturer’s instructions. To calculate the allergen concentrations per gram of dust, the standard curve, was plotted from double dilutions of each pure allergen. The lower detection limit of the ELISA assay was 2 ng/mL for Der p1 and Der f1, 0.8 ng/mL for Fel d1, 0.32 ng/mL for Asp f1, 0.08 ng/mL for Bla g1 and 0.2 ng/mL for Mus m1.

Skin prick tests were performed for each patient with standard commercial extracts of dust mites (D. pteronyssinus and D. farinae), cat, mouse, cockroach and A. fumigatus (Greer, Lenoir, NC, USA). Histamine (10 mg/mL) and saline were used as positive and negative controls, respectively. The results of the skin tests were examined after 15 min and considered positive when the wheal was 3 mm greater in diameter than the negative control.

Statistical Analysis: Normal distribution of data was verified with the Kolmogorov-Smirnov test. The mean levels of each allergen in the child’s bedroom and kitchen were compared with the nonparametric Mann-
Whitney U test. Spearman’s rank correlation coefficient was used to find the strength of association between home characteristics or reactivity of skin prick test and allergen levels. All statistical analyses were done with SPSS 15.0, and \( p \leq 0.05 \) was considered statistically significant.

RESULTS

A total of 35 homes of families with a child who had asthma were selected for dust collection. Twenty-two children (63%) were living in apartments and the remaining 13 were living (37%) in houses. Detectable levels of mite, mouse and cockroach allergens were found in all homes. Cat allergen was found in 94.2% and A. fumigatus in 85.7% of homes. Table 1 shows the levels of six indoor allergens in samples from the child’s bedroom and the kitchen. The levels of Der f1 were significantly higher than Der p1 both in the kitchens and in the child’s bedrooms. The levels of Fel d1 and Bla g1 allergens in the child’s bedrooms were significantly higher than those in the kitchens \( (p<0.0001 \) and \( p=0.009 \), respectively).

Direct correlations were observed between living in an apartment and Der f1 levels \( (p=0.04) \). Table 2 shows the correlation between indoor allergen levels and home characteristics.

The presence of cockroaches during the previous year was reported by 50% of the families. Dampness was not a significant factor to increase the level of Asp f1 at homes.

A total of 34.2% of the patients showed positive skin reactions to at least one of the tested allergens as 17.1% of them showed reactivity to D. pteronyssinus, 14.2% to D. farinae, 14.2% to mouse, 11.4% to cat, 5.7% to cockroach and 2.8% to A. fumigatus.

Table 1. Indoor dust allergen levels in homes of families with a patient with childhood asthma (n=35)

<table>
<thead>
<tr>
<th>Topics</th>
<th>Der f1 (ng/g)</th>
<th>Der p1 (ng/g)</th>
<th>Mus m1 (ng/g)</th>
<th>Fel d1 (ng/g)</th>
<th>Bla g1 (ng/g)</th>
<th>Asp f1 (mg/g)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Kitchen</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Range</td>
<td>6.8-112</td>
<td>0.0-11.4</td>
<td>0.0-65.4</td>
<td>0.0-19</td>
<td>0.0-676.2</td>
<td>0.0-21.8</td>
</tr>
<tr>
<td>Mean ± SD</td>
<td>19.3±17.37</td>
<td>5.0±2.77</td>
<td>16.6±12.35</td>
<td>1.2±3.65</td>
<td>205.5±139.43</td>
<td>4.5±6.33</td>
</tr>
<tr>
<td><strong>Child’s bedroom</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Range</td>
<td>2.4-87</td>
<td>0.0-11.8</td>
<td>2.2-78.6</td>
<td>0.0-19.6</td>
<td>63.4-1232.8</td>
<td>0.0-26.4</td>
</tr>
<tr>
<td>Mean ± SD</td>
<td>16.9±20.47</td>
<td>3.7±2.61</td>
<td>19.5±14.85</td>
<td>5.3±5.58</td>
<td>285.3±207.37</td>
<td>5.2±6.49</td>
</tr>
</tbody>
</table>

Table 2. Correlation between home characteristics and indoor dust allergen levels

<table>
<thead>
<tr>
<th>Home characteristics</th>
<th>R &amp; p-value</th>
<th>Der f1</th>
<th>Der p1</th>
<th>Mus m1</th>
<th>Fel d1</th>
<th>Bla g1</th>
<th>Asp f1</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Home age</strong></td>
<td>R</td>
<td>0.325</td>
<td>-0.351</td>
<td>-0.186</td>
<td>0.054</td>
<td>0.251</td>
<td>-0.298</td>
</tr>
<tr>
<td></td>
<td>p-value</td>
<td>0.061</td>
<td><strong>0.039</strong></td>
<td>0.284</td>
<td>0.76</td>
<td>0.145</td>
<td>0.082</td>
</tr>
<tr>
<td><strong>Occupancy years</strong></td>
<td>R</td>
<td>-0.08</td>
<td>-0.221</td>
<td>-0.045</td>
<td>0.328</td>
<td>0.338</td>
<td>-0.304</td>
</tr>
<tr>
<td></td>
<td>p-value</td>
<td>0.655</td>
<td>0.201</td>
<td>0.798</td>
<td>0.055</td>
<td><strong>0.047</strong></td>
<td>0.076</td>
</tr>
<tr>
<td><strong>Number of rooms in the home</strong></td>
<td>R</td>
<td>-0.012</td>
<td>0.022</td>
<td>0.164</td>
<td>-0.011</td>
<td>-0.311</td>
<td>-0.001</td>
</tr>
<tr>
<td></td>
<td>p-value</td>
<td>0.946</td>
<td>0.902</td>
<td>0.347</td>
<td>0.95</td>
<td>0.069</td>
<td>0.997</td>
</tr>
<tr>
<td><strong>Average home size</strong></td>
<td>R</td>
<td>0.096</td>
<td>-0.002</td>
<td>0.019</td>
<td>0.334</td>
<td>-0.308</td>
<td>0.069</td>
</tr>
<tr>
<td></td>
<td>p-value</td>
<td>0.591</td>
<td>0.989</td>
<td>0.913</td>
<td><strong>0.05</strong></td>
<td>0.072</td>
<td>0.692</td>
</tr>
<tr>
<td><strong>Number of householders in the home</strong></td>
<td>R</td>
<td>0.09</td>
<td>-0.132</td>
<td>-0.022</td>
<td>0.172</td>
<td>-0.16</td>
<td>-0.04</td>
</tr>
<tr>
<td></td>
<td>p-value</td>
<td>0.612</td>
<td>0.449</td>
<td>0.898</td>
<td>0.323</td>
<td>0.358</td>
<td>0.821</td>
</tr>
</tbody>
</table>

R, Spearman’s rank correlation coefficient. Statistically significant associations \( (p<0.05) \) are shown in boldface.

DISCUSSION

Exposure to multiple indoor allergens is common everywhere in the world. Different levels of house dust mites from 85% to 95% have been detected in the homes of patients with asthma.\(^{14,15}\) In our study, detectable levels of house dust mite allergens were found in all evaluated places; which is in contrast to the
data reported by Fereidouni et al., who found house dust mites only in northern Iran.\textsuperscript{13} Despite the warm-dry climate in Shiraz, moisture from sweating during the night may be sufficient for surviving the mites. Moreover, this study was conducted during the winter, when heat and humidity were enough to allow mites to grow. None of the studied homes had dust mite levels greater than 200 ng/g which was considered as a threshold concentration for allergic sensitization.\textsuperscript{16} Nevertheless, 20% of our patients were sensitive to dust mites according to their skin tests, so it appears that sensitization may occur at much lower concentrations of these allergens.

Carpets as the main reservoir of house dust mites were found in all of the studied homes, thus this factor was not included in our evaluation of mite allergen levels.

Many epidemiologic studies just focused on the levels of Der p1 in house dust; in this study the levels of both Der f1 and Der p1 were determined. The results showed that Der f1 was more common than Der p1, which is consistent with reports from northern Iran, South Korea and Southern China.\textsuperscript{13, 15, 17} The prevalence of Der f1 may therefore be higher in certain climates, but because of not being able to monitor temperature and humidity accurately, it could not be said whether the conditions in the homes studied were more suitable for Der f1-producing mites than other mites.

The results showed that Der f1 levels were significantly greater in apartments than in houses. Because of the smaller size of the dwelling or the use of centralized heating systems, heat uniformly in apartments were probably easier than houses, and it may provide more favorable conditions for the growth of mites.

In contrast to previous reports,\textsuperscript{14, 18} the level of Der p1 was higher in modern homes than older ones in the present study. Foam insulation and modern air conditioning systems, which are used routinely for new buildings in Shiraz, may help to retain humidity and support mite growth. Although the level of mite allergens in kitchens was higher than those in the children’s bedrooms, the difference was not statistically significant. It appears that both places were suitable for growing mites.

Detectable levels of Fel d1 were found in 94.2% of the homes studied, in at least one of the two provided samples. Feral cats are abundant in Shiraz and indirect contact with cat’s allergens is frequent. Although exposure to 100 ng/g of Fel d1 has been reported to be associated with increase in the risk of sensitization,\textsuperscript{19} continuous or direct exposures to cat allergens was not reported by any of our patients. The highest level of Fel d1 detected in this study was one-fifth of the sensitization threshold, while 11.4% of our patients showed sensitization to cat allergens in the skin tests. The levels of Fel d1 were associated with the square meter of living places area, and were higher in children’s bedrooms than in kitchens. This may reflect the persistence of cat allergens on clothing, which is more frequent contact with bedding materials, whereas this allergen can be easily removed from the kitchen by frequent cleaning, which is a common activity at Iranian homes.

The results showed detectable levels of mouse allergen in all homes. The prevalence of home-derived mouse allergens has been reported to be from 82% to 95%.\textsuperscript{20, 21} The critical level of this allergen for mouse sensitization is 1600 ng/g.\textsuperscript{20} and none of the patients were exposed to this level of mouse allergen in their homes however, 14.2% had reactive skin tests to mouse allergens. The levels of Mus m1 did not differ between kitchens and bedrooms. Mice are attracted to the food debris in kitchens and their airborne mouse allergens can be transported indirectly into other rooms.\textsuperscript{21} Our results are in consistent with the study by Coleman et al. who did not find any significant relationship between home characteristics and Mus m1.\textsuperscript{22}

In our study, a detectable level of cockroach allergen was found in all places confirming the earlier reports on the importance of allergen-producing cockroaches in urban settings.\textsuperscript{23} In contrast to the previous findings,\textsuperscript{24, 25} we found remarkably higher levels of this allergen in children’s bedrooms than in kitchens. This reflects that frequent cleaning in the kitchen is effective to decrease the level of cockroach allergen. On the other hand, taking food into the bedroom may provide favorable conditions for gathering cockroaches in there.

Our results also indicated that aspergillus allergen was common in the homes we studied and 85.7% of the dust samples had detectable levels of Asp f1. The rate of skin test sensitization to aspergillus was previously reported to be 11% among 230 children with asthma in southern Iran.\textsuperscript{26} Higher levels of Asp f1 in dust from homes with the evidence of dampness compared to other homes were not found; however, the presence of aspergillus spores could not be ruled out because levels
of air-borne allergens could not be measured in the field.

To minimize the confounding effects of outdoor allergens due to shared epitopes with some indoor allergens, this study was performed in winter although the levels of house dust mite, cockroach and aspergillus allergens may be higher in the summer. 27 The results would be more comprehensive if it were possible to collect dust samples monthly for 1 year or even seasonally.

The concentrations of all studied allergens were less than threshold concentrations of allergic sensitization, it seems that, our patients may be sensitized by exposure to these allergens in their kindergartens/schools. A comparison of allergen levels in kindergartens/schools and the living areas of these children would have shed further light on the role of indoor allergens in the induction and exacerbation of asthma.

Although we did not find any relationship between the concentrations of allergens at home and the rate of sensitization to each allergen, exposure to indoor allergens was shown as a crucial step for the induction of asthma. Therefore public health officials have an important role in proper controlling of urban indoor allergens like cockroaches and mice, as well as predisposing factors for asthma that are gradually rising in the areas like air pollutants 28 and fine particles. 29 This would be practical approaches to avoid inducing asthma or worsening the asthma symptoms.

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