

# Clinical aspects of allergic disease

## Indoor allergen levels in day nurseries

A. Dornelas de Andrade, MSc,<sup>a</sup> D. Charpin, MD,<sup>a</sup> J. Birnbaum, MD,<sup>a</sup>

A. Lanteaume, MSc,<sup>a</sup> M. Chapman, PhD,<sup>b</sup> and D. Vervloet, MD<sup>a</sup>

Marseilles, France, Charlottesville, Va., and Pernambuco, Brazil

**Background:** Because allergic sensitization seems to occur especially during infancy, we decided to evaluate such an exposure in day nurseries.

**Methods:** Thirty day nurseries in Marseilles, which were selected at random, were visited during 2 weeks in April 1993. Routine cleaning includes daily cleaning of smooth floors, weekly laundering of sheets, and monthly cleaning of soft toys. Mattresses are encased in synthetic covers. Dust samples were collected from four settings: infants' mattresses and pillows, smooth floors, and soft toys. Levels of mite, cockroach, cat, and dog allergens were analyzed with a monoclonal antibody-based ELISA.

**Results:** Mite allergen levels were lower than the proposed threshold level for sensitization (2 µg/gm of dust) on 94% of mattresses and soft toys and on 100% of floors and pillows. Cat allergen levels in mattresses ranged from less than 0.1 to 4.5 µg/gm dust. On floors, cat allergen levels ranged from less than 0.1 to 2.4 µg/gm dust. Only 10% of pillows and soft toys had levels greater than 2 µg/gm of dust. Fel d I levels were significantly higher ( $p < 0.03$ ) in mattresses from nurseries with curtains and were correlated with the percentage of children with a cat at home. In almost all day nurseries, cockroach allergen (Bla g I and Bla g II) levels were very low. Only three samples from mattresses had dog allergen levels greater than 2 µg of Can f I allergen per gram of dust. On floors the level was always lower than 2 µg/gm.

**Conclusions:** These data clearly show that indoor allergen levels are much lower in day nurseries than in most houses. Most samples contain allergen levels below threshold levels for sensitization. Thus children of atopic parents are less likely to become sensitized to indoor allergens in day nurseries than in their own homes. In addition, this study emphasizes the efficacy of avoidance measures such as use of synthetic protective mattress covers, frequent washing of sheets and soft toys, and avoidance of carpets and curtains. (*J ALLERGY CLIN IMMUNOL* 1995;95:1158-63.)

**Key words:** House dust mite, cat, dog, cockroach, day nurseries, environment, allergy, housing

Several reports<sup>1-4</sup> have shown that early exposure to indoor allergens, especially house dust mites is a strong risk factor for sensitization and development of asthma. Avoidance programs fo-

### Abbreviation used

mAb: Monoclonal antibody

From <sup>a</sup>Département des Maladies Respiratoires, Hôpital Sainte Marguerite, Marseilles, France; and <sup>b</sup>Division of Allergy and Clinical Immunology, Health Sciences Center, Charlottesville.

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Reprint requests: Daniel Vervloet, Service de Pneumo-Allergologie, Département des Maladies Respiratoires, Hôpital Sainte Marguerite, BP 29, 13277 Marseille Cedex 9, France.

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cus on dwellings. However, infants (aged 3 months to 3 years) may spend 30% of their time in day nurseries. Unlike schools, day nurseries mimic homes because they include a kitchen and mattresses. This study evaluated exposure to indoor allergens (mite, cat, dog, and cockroach) in day nurseries in Marseilles, France and examined the relationship between housing characteristics and cleaning habits on the one hand and concentration of major allergens in dust on the other hand.

## METHODS

### Environmental sampling

Thirty day nurseries were selected at random from the 60 day nurseries in the town of Marseilles, France. They were visited by one of the investigators during a 2-week period in April 1993. Because of possible seasonal variations, a second sampling was performed in October in a subgroup of 10 day nurseries.

During each visit, the director of the day nursery filled out a questionnaire about arrangements and routine cleaning of his or her day nursery. A second questionnaire, related to infants' and staff members' exposure to pets, was given to the director and collected a few days later.

Dust samples were collected in each day nursery from three different areas: infant section (3 months to 1 year of age), second-year section, and third-year section. In each section dust was vacuumed from three mattresses, three pillows, three soft toys, and the smooth floor of the room where mattresses were kept. In the analysis, dust samples from the three different sections were considered together. Samples were collected according to the recommendations of the First International Workshop on Dust Allergens and Asthma.<sup>5</sup> During the study period, directors were advised not to change routine cleaning methods. Altogether, four dust samples were collected in each day nursery (i.e., one sample each from mattresses, pillows, soft toys, and smooth floors); thus, 120 samples were studied. The fine dust was weighed; then a 100 mg aliquot was extracted in 2 ml of borate-buffered saline (pH 8.2), incubated overnight, and centrifuged. The supernatant was stored at  $-20^{\circ}\text{C}$ . Of the 120 dust samples collected, 44 did not contain enough dust and were discarded.

### ELISA

Levels of mite (group I = *Der p I* + *Der f I*), cat (*Fel d I*), and cockroach (*Bla g I* and *Bla g II*) allergens were analyzed in 76 samples, and dog allergen (*Can f I*) in 46 samples with monoclonal antibodies (mAbs) and an ELISA method. Details of the assays have been reported elsewhere.<sup>6-10</sup> The detection limits for the assays were 0.1  $\mu\text{g/gm}$  for *Der p I* and *Der f I*, 0.1  $\mu\text{g/gm}$  for *Fel d I*, and 0.6 U/gm for cockroach (*Bla g I* and *Bla g II*) allergens. Two dilutions of each sample were assayed, and each dilution was assayed twice. Thus each result was the mean of these four determinations. The concentration of *Fel d I* was calculated assuming 1 U *Fel d I* to be equivalent to 4  $\mu\text{g}$  of protein.

For cockroach allergen, two mAbs were used, mAb 10A6 for *Bla g I* and mAb 8 F4 for *Bla g II*. The ELISAs for *Bla g I* and *Bla g II* were performed according to methods previously described,<sup>8</sup> that is, with polyclonal rabbit anti-cockroach antibodies. The reference (UVA 89/01) contained 500 U/ml *Bla g I* and 300 U/ml *Bla g II*. The concentration of *Can f I* protein was calculated assuming 1 U of *Can f I* to be equivalent to 1 ng of *Can f I* protein (Dr. R. Aalberse; personal communication).

TABLE I. Routine measures for allergen reduction in day nurseries ( $n = 30$ )

Measure	Percent of nurseries
Synthetic mattress covers used	100
Sheets washed at least weekly	100
Smooth floors cleaned daily	100
Soft toys washed monthly	80
Synthetic mattress covers washed at least monthly	60

### Statistical analysis

Analysis of variance was used to compare mean values, Spearman's rank correlation to comparatively assess rank in two series, and simple correlation to compare levels of two quantitative variables.

## RESULTS

### Characteristics of day nurseries

**Building characteristics and arrangements.** In the group of 30 day nurseries, seven were built between 1901 and 1957, 15 between 1960 and 1978, and eight between 1980 and 1992. Smooth floors were present in 100% of day nurseries, and central heating was present in 80%. There were no visible signs of molds, humidity, or water infiltration in 83%, 73%, and 67% of the day nurseries, respectively.

**Routine cleaning.** Table I, which is based on the standardized questionnaire, shows the routine cleaning in the day nurseries.

### Indoor allergen levels

**Mite allergens.** Group I mite allergen levels (*Der p I* + *Der f I*) ranged from less than 0.1 (below the detection limit of the ELISA assay) to 5.3  $\mu\text{g/gm}$  on mattress dust, from less than 0.1 to 1.4  $\mu\text{g/gm}$  on floors, from less than 0.1 to 0.4  $\mu\text{g/gm}$  on pillows, and from less than 0.1 to 2.3  $\mu\text{g/gm}$  on soft toys. Concentration of mite allergens was lower than the proposed threshold level for sensitization ( $<2 \mu\text{g/gm}$  of dust) on 94% of mattresses and soft toys and on 100% of floors and pillows (Fig. 1).

In samples collected in October, levels of mite allergens were somewhat higher than those in samples collected in April. Levels ranged from less than 0.1 to 3.4  $\mu\text{g/gm}$  in mattress dust (geometric mean = 0.4  $\mu\text{g/gm}$ ) and from less than 0.1 to 0.6  $\mu\text{g/gm}$  in floor dust (geometric mean = 0.2  $\mu\text{g/gm}$ ) but were still lower than the proposed threshold level for sensitization.

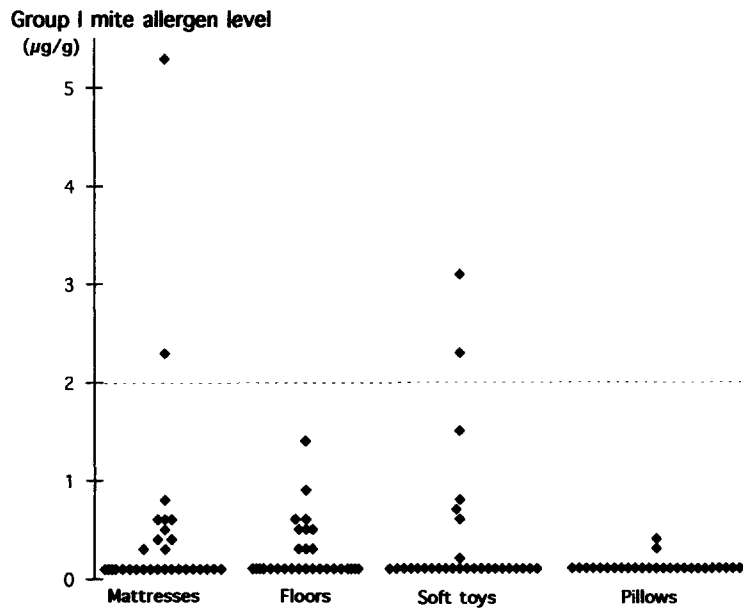


FIG. 1. Group I (*Der p 1* + *Der f 1*) mite allergen levels (micrograms per gram of dust) from various sites of 30 day nurseries. Dashed line indicates threshold level for sensitization.

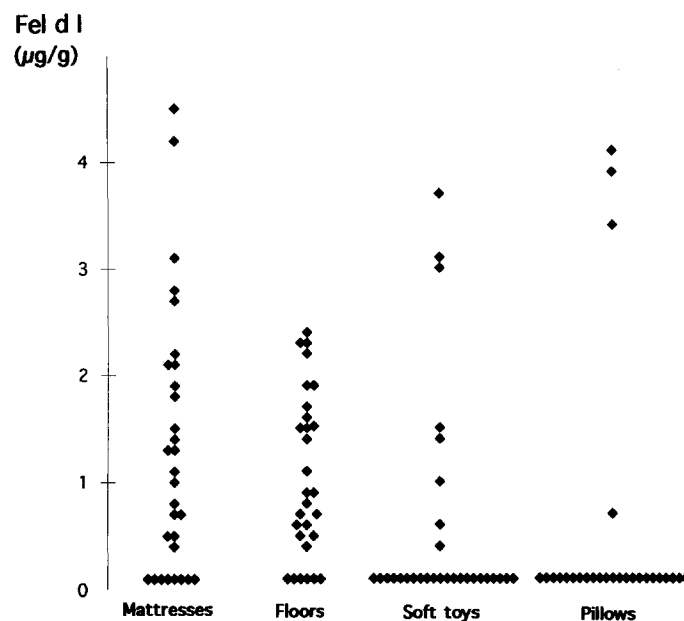
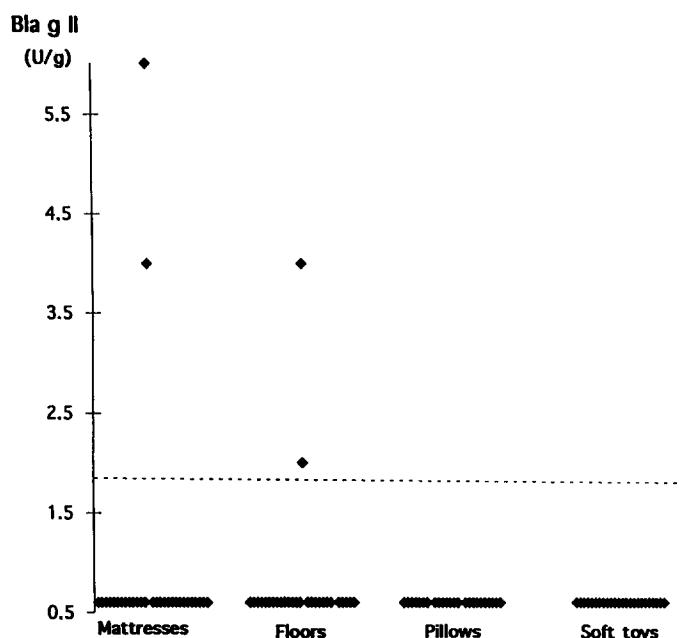


FIG. 2. Levels (micrograms per gram of dust) of cat allergen (*Fel d 1*) from various sites of 30 day nurseries.

**Cat allergens.** Cat allergen (*Fel d 1*) levels were higher than mite allergen levels. On mattresses, they ranged from less than 0.1 (below the detection limit of the ELISA) to 4.5 µg/gm of dust (27% >2 µg of dust). On floors, they ranged from less than 0.1 to 2.4 µg/gm of dust (13% >2 µg/gm). On pillows cat allergen levels ranged from less than 0.1 to 4.1 µg/gm and on soft toys, from less than 0.1 to

3.7 µg/gm with only 10% greater than 2 µg/gm of dust. Fig. 2 shows the distribution of *Fel d 1* in different samples. *Fel d 1* levels were significantly higher ( $p < 0.03$ ) on mattresses from nurseries with curtains. *Fel d 1* levels were somewhat higher on the mattresses (geometric mean = 0.9 µg/gm dust) and floors (0.8 µg/gm of dust) from nurseries with foam carpets than from day nurseries without



**FIG. 3.** Levels (units per gram of dust) of cockroach allergens (*Bla g II*) from various sites of 30 day nurseries.

such carpets (geometric mean on mattresses and floors = 0.4 and 0.5  $\mu\text{g/gm}$  of dust, respectively), but this difference did not reach statistical significance. *Fel d I* levels on mattresses and floors were correlated ( $r = 0.2, p < 0.01$ ), as well as *Fel d I* and mite allergen levels on mattresses ( $r = 0.3, p < 0.001$ ). *Fel d I* levels on mattresses correlated ( $r = 0.14, p < 0.05$ ) with the percentage of children with a cat at home. Such a relationship with the staff members who have cats could not be demonstrated.

**Cockroach allergens.** Cockroach allergen (*Bla g I* and *Bla g II*) levels were very low in almost all day nurseries. Cockroach (*Bla g I*) allergen was found in samples from five day nursery floors, ranging from 2 U/ml to 14 U/ml. In the other 25 day nurseries, the level was below the detection limit of the ELISA. On mattresses we found 2 U/gm of dust in only four day nurseries. On pillows and soft toys, only three showed 2 U/gm of dust of cockroach (*Bla g I*) allergens (data not shown). *Bla g II* allergen was detected on mattresses from two day nurseries (4.0 and 6 U/gm of dust, respectively) and on floors from three nurseries, in the range of 2 to 4 U/gm. On pillows and soft toys, *Bla g II* antigen levels were below the detection limit of the ELISA method (Fig. 3). No visible evidence of cockroach infestation was seen in any day nursery. In nurseries with visible signs of humidity (infiltration of water and presence of molds) mean level of

cockroach allergens on mattresses (*Bla g I* and *Bla g II*) was significantly higher ( $p = 0.003$ ) than in day nurseries without such signs.

**Dog allergens.** Forty-six samples were evaluated for dog allergen levels. The level was greater than 2  $\mu\text{g}$  of *Can f I* allergen per gram of mattress dust in only three samples (2.3, 4.1, and 4.5  $\mu\text{g/gm}$ , respectively). In all others the level was very low, ranging from less than 0.2 to 1.8  $\mu\text{g/gm}$ . On floors the level was much lower, ranging from less than 0.2 to 1.2  $\mu\text{g/gm}$  (Fig. 4). There was no correlation between the number of children and staff members with a dog at home and *Can f I* allergen levels.

## DISCUSSION

These findings suggest that day nurseries are protected against mite, cat, dog, and cockroach allergens.

In our study we found a level of mite allergens greater than 2  $\mu\text{g/gm}$  of dust in only two of 30 (6%) day nurseries. In addition, the highest level found was 5.3  $\mu\text{g/gm}$  of dust. Such low levels are most likely related to the presence of smooth floors,<sup>11, 12</sup> use of synthetic mattress covers,<sup>13, 14</sup> and at least weekly washing of sheets in all day nurseries studied. Only three day nurseries reported visible signs of humidity, molds, and water infiltration. Among them, two had mite allergen levels higher than 2  $\mu\text{g/gm}$  dust. The low level of mite allergens was confirmed in samples collected in October,

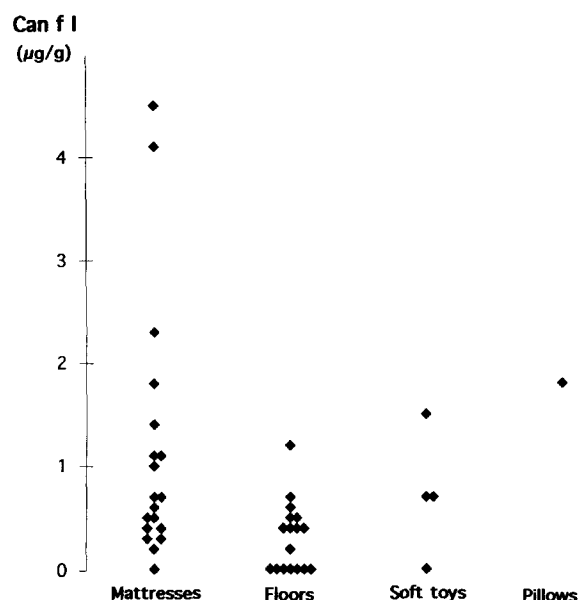


FIG. 4. Distribution of *Can f I* allergen levels (micrograms per gram of dust). Each point represents the individual value for the 30 different day nurseries.

which is known to be the high mite level season. In any case, the mite allergen levels were lower than the proposed threshold for sensitization.<sup>1</sup> Our data support the results of other studies,<sup>11,15</sup> which demonstrated such low mite allergen levels in schools and public places.

Cats are an important source of indoor allergens, but results are more difficult to interpret because there is no defined threshold level for sensitization. Pollart et al.<sup>16</sup> have suggested that exposure to more than 10 μg of *Fel d I* per gram of dust could be a risk factor for acute asthma attacks and that exposure to more than 2 μg of *Fel d I* per gram of dust is a risk factor for sensitization. In contrast, Call et al.<sup>17</sup> proposed 8 μg/gm as a threshold level for sensitization. Gelber et al.,<sup>18</sup> in a recent report, showed that 8 μg/gm of *Fel d I* allergen is the level at which almost all patients who are allergic to cats experience symptoms and is the minimal level found in a house with a cat. According to these latter thresholds, none of the day nurseries studied have sufficient allergen levels to induce sensitization or to cause symptoms. Levels measured in day nurseries are similar to those in houses where a cat has never been kept.<sup>19</sup> Cat allergens in day nurseries probably originate from shoes and clothes of persons who keep a cat at home.<sup>20,21</sup> Indeed, we found a correlation between the percentage of children who have cats and the *Fel d I* levels in mattresses. For the staff members, there was no such correlation with the

allergen level, probably because they change clothes and shoes before entering day nurseries.

Levels of cockroach allergen were low. They could have been higher if samples had been collected in the kitchen. Pollart et al.<sup>8</sup> showed that dust from kitchen floors contained 50 times more allergen than dust from bedrooms. In our study we collected dust from bedrooms and playrooms but not from kitchens because the children are not allowed to stay there. Among the 30 day nurseries, only six had measurable cockroach allergen levels (*Bla g I* + *Bla g II*) on mattresses in association with visible signs of humidity (infiltration of water and molds).

Our study shows that dog allergen levels in day nurseries are low, similar to or lower than those in houses without dogs. Schou et al.<sup>10</sup> found a mean *Can f I* level of 3 μg/gm of dust in houses without dogs. A *Can f I* level of greater than 10 μg/gm of dust has been found in 95% of houses with a dog.<sup>22</sup> This value has been proposed to be the threshold for dog sensitization. In the day nurseries, the median value was lower than 1 μg/gm of dust. *Can f I* concentrations in our day nurseries were much lower than those recently reported by Munir et al.<sup>23</sup> in Swedish schools. In this study *Can f I* levels were remarkably high, ranging from 1.7 to 28.2 μg/gm on chairs and floors. This might be so because in day nurseries, unlike schools, floors are cleaned by wet sweeping at least once a day.

## Conclusion

This study evaluated indoor allergen exposure levels (mite, dog, cat, cockroach) in day nurseries in Marseilles, France. Allergen levels in day nurseries were generally low, in most cases below the proposed thresholds for IgE-mediated sensitization. The results suggest that (at least in the Marseilles area) indoor allergen exposure in nurseries is unlikely to be a significant cause of sensitization or symptoms in infants. In addition, this study emphasizes the efficacy of such avoidance measures as covering mattresses; regularly washing sheets and soft toys; avoiding carpets, curtains, and rugs; and installing smooth floors.

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