

Comparison of vacuuming procedures for reservoir dust mite allergen on carpeted floors

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Allergen concentration in dust collected from a single square meter of carpet has been widely used as a proxy for aeroallergen exposure in a room. However, little is known about the horizontal distribution of allergens in carpeting or whether sampling several smaller sites might better approximate the average level. Eight bedrooms and 4 lounge rooms were divided into squares of 0.25 m², and the vacuumed dust was assayed for mite allergen (Der p 1). The proximity of each square to windows, doors, and beds was determined. In each room, the total allergen for each of 3 randomly chosen sets of 4 adjacent squares (forming a 1-m² area) was compared with that for each of 3 randomly chosen sets of 4 nonadjacent squares. The reliability of measuring allergen concentration from a single 0.25-m² area was modest (intraclass correlation coefficient [ICC] = 0.52), indicating wide-ranging allergen concentrations across short distances within each room. The reliability of the measurements based on 4 nonadjacent squares (ICC = 0.782) was higher than that of the measurements based on 4 adjacent squares (ICC = 0.67). Der p 1 concentrations decreased with distance from doors (paired partial correlation coefficient, -0.1217; *P* = 0.03). We recommend sampling 4 nonadjacent 0.25-m² areas that are not equidistant from the door when reservoir concentrations of Der p 1 are being measured. (*J Allergy Clin Immunol* 2002;109:122-4.)

Key words: Der p 1, exposure, horizontal distribution, reservoir

Much of our understanding of the relationship between allergic disease and exposure to a domestic allergen is based on measurement of the allergen content of house dust as a proxy for that exposure. The recommended protocol for sampling reservoir dust involves vacuuming a single area of 1 m² of carpet.¹ Despite the wide adoption of this protocol, only 3 studies have examined the distribution of Der p 1 allergen within rooms.²⁻⁴

Abbreviation used

ICC: Intraclass correlation coefficient

These studies showed massive variation between sites among separate samples collected concurrently from the same area. In a 1995 study done in Sydney, Australia,² the 95% ranges of single determination were 1.91-fold and 5.11-fold for bedroom and lounge room (living room) floor areas, respectively; in a German study,³ the ranges were 12.4-fold and 14.8-fold for beds and floor areas, respectively. In a study done in Manchester, United Kingdom,⁴ the corresponding 95% ranges, estimated from data presented in the paper, were approximately 23.7-fold and 12.9-fold for bedroom floor and lounge room floor areas, respectively. Such variation raises the question of whether a different sampling strategy of collecting dust from several smaller sites having the same total area in a room might better represent the overall allergen concentration of the room. Such a protocol has recently been used, though without validation.⁵

It is also unknown whether the distribution of allergen within a carpet site is related to its proximity to different items in the room. One could hypothesize that proximity to a doorway might increase allergen via greater human traffic or, alternatively, might reduce it as a result of dilution of allergen with trafficked sand; proximity to a bed might be associated with more settled aeroallergen from the bed, and proximity to a window might be associated with a modified microclimate and therefore with local production of dust mite allergen.

In this study, we examined the distribution of dust mite allergen concentration in 12 carpeted rooms by analyzing the allergen concentration in 0.25-m² squares (termed *subsquares*). To explore the effect of sampling at 4 distributed subsquares in comparison with sampling at a single larger site, the intraclass correlation coefficient (ICC) reliability of measuring total allergen concentration in sets of 4 nonadjacent subsquares chosen at random was compared with that of measuring the concentration in sets of 4 adjacent subsquares. These 2 sam-

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pling strategies were compared with sampling 1 subsquare in each room.

To study the effect of location within a room, the allergen concentration in subsquares was examined as a function of distance from beds, doorways, and windows. The distance from a window or bed was measured directly between the two, whereas the distance from a doorway was measured as the shortest walking route.

METHODS

Sampling procedure

The study was conducted in 8 carpeted (wall-to-wall) bedrooms and 4 loungerooms in 8 homes in Sydney. With masking tape, the available floor space in each room was divided into subsquares of 0.25 m². A diagrammatic representation of one of the rooms has been published elsewhere.⁶ Distances were measured to each square from each window, doorway, and bed.

Allergen quantification

Dust was collected through use of a hand-held vacuum cleaner (Makita Electric Works Ltd, Anjo, Japan) fitted with a 25- μ m nylon filter bag for each sample. Each subsquare was vacuumed for 30 seconds.

The general assay procedure has been described previously.⁷ Briefly, each sample was sieved (particle diameter, <0.4 mm), and 100 mg of the fine dust was extracted in 2 mL 1% BSA/PBS/0.5% Tween 20 overnight at room temperature and then centrifuged; the supernatant was stored at -20°C before assay. The amount of dust mite allergen (Der p 1) was quantified by means of ELISA kits (Indoor Biotechnologies Ltd, Cardiff, United Kingdom) and expressed in micrograms of Der p 1 per gram of fine dust through use of the allergen standard supplied with the kit.

Statistical analyses

To randomly select 4 adjacent squares forming 1 m², each 0.25-m² subsquare ($n = 14$ to 39; median, 26 per room) was assigned a number and one of the subsquares was selected by random number generation (Excel 2000 for Windows, Microsoft, Redmond, Wash). Every subsquare combination with an area of 1 m² that included the selected subsquare was assigned a number from 1 to 4, and the final 1-m² area was selected by random number generation. The second and third 1-m² areas were selected by the same process as the first. Selection of the available subsquares was performed without replacement.

Three sets of 4 nonadjacent subsquares, each representing a total area of 1 m², were selected from each room. This was achieved for each room by selecting 12 subsquares at random, without replacement. The first 4 selected subsquares formed the first set, the next 4 the second set, and the remaining 4 the third set.

The Der p 1 concentration value assigned to each set of 4 adjacent or nonadjacent subsquares was the geometric mean of the 4 subsquares' values. Data were log₁₀-transformed to normalize the distribution.

The reliability of each of these 2 sampling strategies and of the alternative based on individual subsquares was assessed by calculating the ICCs⁸ for the resulting units of observation—ie, for sets of 4 adjacent subsquares, sets of 4 nonadjacent subsquares, and individual subsquares. These sampling techniques were compared for all of the subsquares in the room through use of ICCs.⁸ Because the analysis required an equal number of samples (subsquare concentrations) per subject (room) and because the smallest number of subsquares in a room was 14, a total of 14 subsquares of each room were selected by random number generation to represent all of the subsquares in the room.

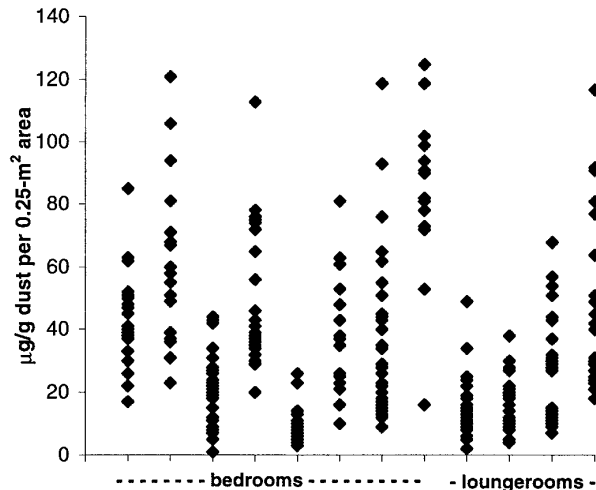


FIG 1. Concentration of Der p 1 allergen in each 0.25-m² area ($n = 14$ to 39 per room) in 12 carpeted rooms.

The range of single determinations were calculated according to the method of Marks et al.² These values indicate the “fold” difference of a single estimate within which the true value would be expected in 95% of cases. Briefly, through use of log data, the SD of the single estimate is calculated as the square root of the residual mean square from an ANOVA table. The range of single determinations is calculated as the anti-log of $1.96 \times$ the SD.

The relationships between Der p 1 concentration and distance from beds, windows and doorways were examined by means of SASS statistical software (SAS Institute Inc, Cary, NC) through use of paired Pearson's correlation coefficients with adjustment for the effect of differences between the rooms (partial correlation coefficient).

RESULTS

The allergen concentrations in the 0.25-m² subsquares are shown for each room in Fig 1. The smallest range within a room was 3 to 26 μ g/g of dust, and the largest range within a room was 9 to 119 μ g/g of dust—a difference of 13-fold. The mean subsquare concentration of Der p 1 in the rooms varied between 10.4 and 83.9 μ g/g of dust.

Geometric mean values for the 3 sets of adjacent and nonadjacent subsquares per room are shown in Fig 2. The reliability of sampling a 1-m² area as 4 nonadjacent subsquares was higher (ICC = 0.812) than that of sampling a 1-m² area as 4 adjacent subsquares (ICC = 0.686) or as individual 0.25-m² subsquares (ICC = 0.36).

The “fold” difference of a single estimate within which the true value would be expected in 95% of cases was 1.77-fold for nonadjacent subsquares, 2.20-fold for adjacent subsquares, and 3.17-fold for all subsquares.

Within rooms, there was a significant negative correlation between distance from doorway and mite Der p 1 concentration (partial correlation coefficient, -0.117; $P = .03$), such that concentration decreased with walking distance from the door. There was no significant correlation ($P > .05$) between Der p 1 concentration and distance from beds or windows.

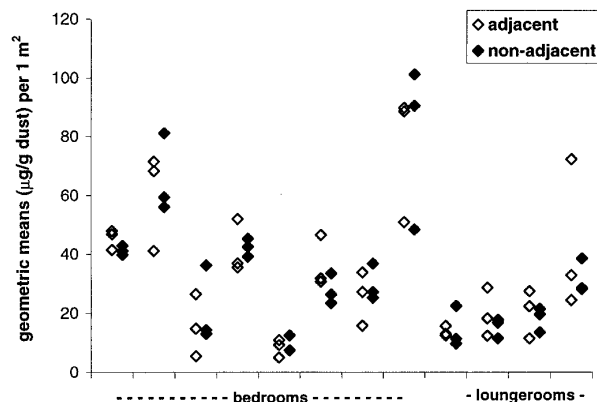


FIG 2. Geometric mean concentrations of Der p 1 in 1-m² areas when sampled as either 4 adjacent or 4 non-adjacent 0.25-m² areas. Three 1-m² areas were sampled per method per room in 12 carpeted rooms.

DISCUSSION

This study compared 2 methods of collecting reservoir dust mite allergen from carpeted floors and examined potential factors that might have influenced the distribution of dust mite allergen across a carpet. Our findings showed that sampling 4 distributed (nonadjacent) 0.25-m² areas was a less variable estimate of allergen concentration in a room than sampling a single 1-m² area; it thus better represented the distribution of allergen across the room. A negative gradient of Der p 1 concentration was shown with distance from doorway.

In this study, the reproducibility of single estimates of Der p 1 collected from nonadjacent subsquares was higher than that for single subsquares and adjacent subsquares and also higher than all values detected in previous investigations of 1-m² areas.²⁻⁴ These findings provide further evidence that sampling 4 nonadjacent subsquares is the optimal measure of sampling Der p 1 from floors.

The factors responsible for the patterns of variation detected within a room are not known. The distribution of dust mite food in settling dander, variations in microclimate within the carpet, the settling pattern of dust in areas with different air currents, the proximity of aeroallergen sources, the effects of cleaning, and the dilution of allergen by trafficked dust would all potentially contribute. In addition, any measurement of allergen itself seems to be associated with some nonsystematic sampling error. The association with trafficked areas, such as doorways, would suggest that food supply might be a more important factor in general.

From the findings in this and other studies, it is understood that allergen concentration in carpets changes with time,³ between rooms, with the presence of insulation, with the presence of rooms under the flooring (eg, a garage), across the carpeting (ie, horizontally), with thickness of the carpet underlay,⁹ and between types and length of carpet fibers^{3,10}; studies in progress in our lab-

oratory have also suggested vertical differences within the pile. However, it remains unclear how much of this allergen becomes airborne and inhalable and whether vacuum sampling for dust mite allergen accurately reflects what residents inhale over time.

In conclusion, we recommend sampling 4 nonadjacent 0.25-m² areas that are not equidistant from doorways to reliably measure the allergen across a carpeted floor.

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