

Original article

Evaluation of home allergen sampling devices

Background: Simple, inexpensive methods of sampling from allergen reservoirs are necessary for large-scale studies or low-cost householder-operated allergen measurement.

Methods: We tested two commercial devices: the Indoor Biotechnologies Mitest Dust Collector and the Dräger Bio-Check Allergen Control; two devices of our own design: the Electrostatic Cloth Sampler (ECS) and the Press Tape Sampler (PTS); and a Vacuum Sampler as used in many allergen studies (our Reference Method). Devices were used to collect dust mite allergen samples from 16 domestic carpets. Results were examined for correlations between the sampling methods.

Results: With mite allergen concentration expressed as $\mu\text{g/g}$, the Mitest, the ECS and the PTS correlated with the Reference Method but not with each other. When mite allergen concentration was expressed as $\mu\text{g/m}^2$ the Mitest and the ECS correlated with the Reference Method but the PTS did not. In the high allergen conditions of this study, the Dräger Bio-Check did not relate to any methods.

Conclusions: The Mitest Dust Collector, the ECS and the PTS show performance consistent with the Reference Method. Many techniques can be used to collect dust mite allergen samples. More investigation is needed to prove any method as superior for estimating allergen exposure.

J. K. Sercombe^{1,2}, D. Liu-Brennan^{1,2},
M. L. Garcia^{1,2}, E. R. Tovey^{1,2,3}

¹Woolcock Institute of Medical Research;

²Cooperative Research Centre for Asthma; ³The University of Sydney, Sydney, NSW, Australia

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Mr Jason Sercombe
Woolcock Institute of Medical Research
Rm 461 Blackburn Building D06
The University of Sydney
NSW 2006
Australia

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Exposure to indoor allergens, including those from the house dust mite, are an important factor in the occurrence of allergies and asthma (1, 2). Despite uncertainty defining allergen exposure in terms of asthma symptom risk (1), the combination of allergy and current allergen exposure is a major risk factor for severe asthma and for hospital admissions in adults and children (3, 4). In five controlled clinical trials where dust mite numbers or mite allergen concentrations were reduced by over 100-fold, asthma symptoms and airway hyper-responsiveness were improved (5). Although several recent trials using bedding covers as a means of allergen avoidance in adult asthmatics have shown no significant improvement in symptoms (6–9), some of these authors (6) as well as others in the field (10) stress that effective allergen avoidance should not be ruled out as a means of asthma management. To this end, the measurement of allergen in dust is necessary to monitor the effectiveness of, and to encourage compliance with, interventions to reduce allergen exposure (11, 12).

Reservoir dust samples are often collected as a proxy for chronic aeroallergen exposure and several methods have evolved to collect such samples. In research-based studies, such reservoir samples are usually collected by a technician visiting a patient's home and collecting samples using a modified vacuum cleaner (as reviewed by Platts-Mills and Chapman) (13). Allergen is then quantified using an enzyme-linked immunosorbent assay (ELISA) (14). Recently, kit-sets that enable the consumer to sample their own house dust and perform a rapid, 'pregnancy-test' type assay for mite allergen have become commercially available (15, 16). Such assays indicate allergen levels semi-quantitatively.

None of the allergen collection and assay systems in use have been standardized and a number of sources of variation are apparent. It has been demonstrated that the allergen content of dust collected with a vacuum cleaner may differ significantly over short periods of time (17) and between closely spaced sampling sites (17–20). Additional variables include the type of vacuum cleaner used (21), whether all the dust or just a 'fine' fraction was extracted for analysis (1, 22) and the extraction buffer and temperature used (23). While the convention for vacuum-collected samples is to express allergen content as a mass concentration ($\mu\text{g/g}$) of collected dust in observational studies (24, 25), the convention is to use an area

Abbreviations: BSA/PBST, 1% bovine serum albumin/phosphate-buffered saline 0.05% Tween; ELISA, enzyme-linked immunosorbent assay; ECS, Electrostatic Cloth Sampler; PTS, Press Tape Sampler.

concentration ($\mu\text{g}/\text{m}^2$) in interventional studies (6, 26). While all sampling methods provide valuable data, the systems in use are unstandardized and lack precision.

We have had previous experience using adhesive tape to collect dust from clothing (27) and using electrostatic cloths to remove dust from floors (28), and found both these methods simple to use. They also did not require an external source of energy or bulky equipment, such as a mains or battery-powered vacuum cleaner. Each of these methods was therefore applied in making simple collection devices that could be used to collect dust samples in homes with minimal instruction. For comparison we used a 'reference method', based on standardized collection with a vacuum cleaner, that we have used in previous epidemiological studies (29). We included the Mitest sampler used in one of the commercial rapid allergen assays (15), and also tested the Dräger Bio-Check which features an integrated sampler and a rapid immunoassay in a single unit (16).

Materials and methods

Carpets sampled

We sampled 16 carpets from homes located in Sydney, Australia. All carpets were at least 5 years old. The carpets used featured a wide range of pile heights, weave and materials (wool, synthetic and blends). From each carpet, an area was selected that, based on appearance, had been subject to a consistent degree of wear, soiling and traffic.

Dust sample collection

The same operator used all sampling devices and each area of carpet was sampled only once. Adjacent areas of carpet were sampled using each device, in a random pattern. The areas sampled and the sampling times are given for each device.

Mitest Dust Collector

This commercial sampling device fits to the nozzle of a vacuum cleaner and is supplied with the Indoor Biotechnologies Rapid Test for Mite Allergen (Indoor Biotechnologies Inc, Charlottesville, VA, USA) (30) and other assay kits (31). The device, which has an internal cylindrical filter of nylon mesh (pore size $40\ \mu\text{m}$) has been described elsewhere (15). Four sites, with a total area of $0.25\ \text{m}^2$ were sampled for a period of 2 min, according to the enclosed instructions (30). Following use, the instructions indicate capping each end of the tube, *in situ* extraction for 5 min using 10 ml of buffer, and then decanting the extract for assay. As we were interested in the Mitest's performance as a sampler, and to standardize with the assay used elsewhere, we removed, weighed and then extracted all of the dust in 15 ml of 1% bovine serum albumin/phosphate-buffered saline 0.05% Tween (BSA/PBST) buffer for 2 h with gentle mixing, followed by centrifugation at $800\ g$ for 10 min.

Electrostatic Cloth Sampler

The Electrostatic Cloth Sampler (ECS) evolved from our experience quantifying dust removal from floors by electrostatic cleaning cloths (28). The device (Fig. 1A) consists of a permanently electrostatically

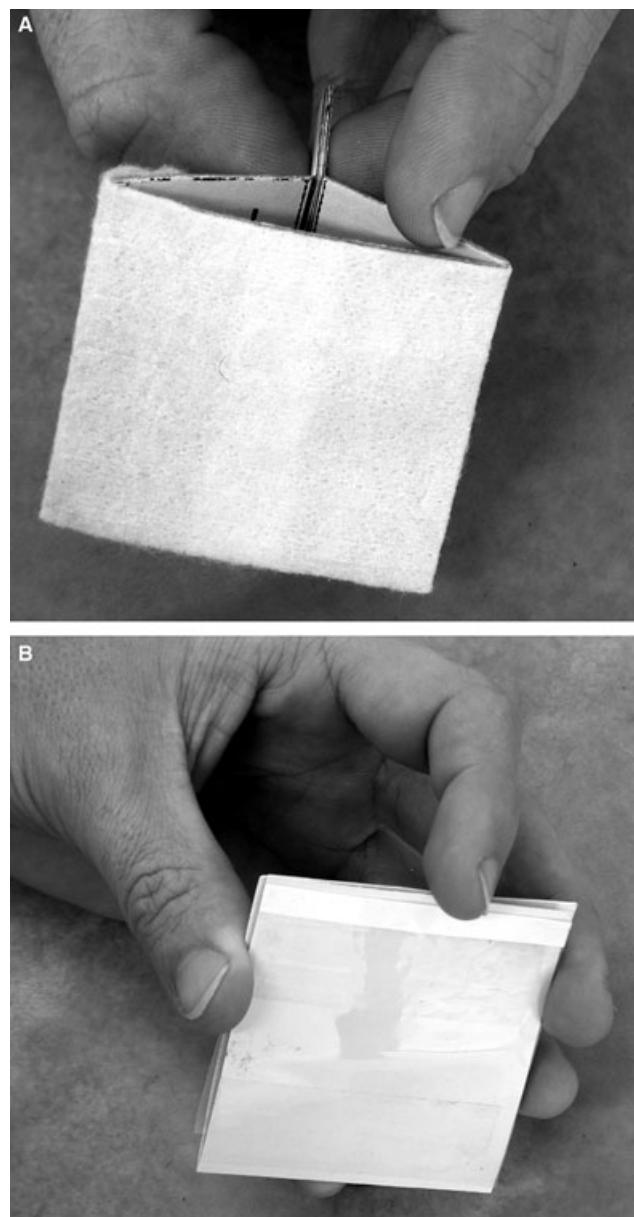


Figure 1. Photograph of the (A) Electrostatic Cloth Sampler and (B) Press Tape Sampler.

charged synthetic fabric (Pledge Grab-it Dry Cloths; SC Johnson & Son Inc, Racine, WI, USA), with a collection area $5 \times 5\ \text{cm}$, adhered to a stiff cardboard backing. The ECS was rubbed on a total of $0.25\ \text{m}^2$ of the surface to be sampled for 1 min. Prior to use the ECSs were dried at 40°C for 15 min to standardize their weight. After use they were re-dried and the weight of collected dust recorded. The cloth portion was then removed from the ECS and allergen extracted in 3 ml of BSA/PBST for 2 h with gentle mixing. The extract was recovered by centrifugation at $800\ g$ for 10 min using a plastic separator to isolate the recovered liquid from the cloth.

Press Tape Sampler

The Press Tape Sampler (PTS) evolved from our experience sampling allergenic dust from fabric surfaces (27). The device (Fig. 1B) consists

of a 5 cm × 5 cm piece of high-tack adhesive tape (Inhalix, Sydney, NSW, Australia) mounted on a cardboard backing. The adhesive is covered with a protective film prior to use, which is removed, and the tape pressed onto 10 different sites totalling a 0.025 m² sample area (i.e. 10 times 5 × 5 cm) within a 0.25 m² area of carpet. Again, the weight of dust collected was estimated by drying and weighing the device before and after use. The allergen was extracted from the adhesive in 2 ml of BSA/PBST for 2 h with gentle mixing, followed by centrifugation at 800 g for 10 min.

Reference method

The vacuum sampling method, used as our reference, uses a modified hand-held vacuum cleaner (Model 4071D; Makita Corp., Anjo Japan), fitted with two conical nylon bags, each of ~55 cm² area held in series in the nozzle. The first bag of 500 µm pore size nylon functions as a pre-filter, and fine dust is collected in the second bag of 25 µm pore size nylon. Four nonadjacent 0.25 m² areas were vacuumed so that an area of 1 m² was sampled over 1 min. The weight of fine dust collected in the second bag was measured, and 50 mg of this fine dust was eluted in 1 mL of BSA/PBST to extract allergen. Extracts were clarified by centrifugation at 800 g for 10 min.

ELISA

Extracts were assayed for mite allergen Der p 1 by a two-site monoclonal ELISA (32) (Indoor Biotechnologies Inc.). The Der p 1 content was calculated both as a mass concentration (µg of Der p 1/g of dust) and as an area concentration (µg of Der p 1/m²). Calculations were corrected for the extraction volumes used with each sampling device.

Dräger Bio-Check Allergen Control

The Dräger Bio-Check Allergen Control (Dräger Sicherheits-technik GmbH, Lübeck, Germany), which measures group 2 (Der p 2 and Der f 2) mite allergens, was used according to its enclosed instructions. Sampling was performed by rubbing the 0.3 cm porous pin of the sampler over an A4 sized area (0.0624 m²) for 60 s and the assay performed as directed. The results, read after 10 min, were classified as 'very high', 'high', 'slight amount' and 'no allergen detected', coded 3, 2, 1 and 0, respectively (33). Due to the construction of this device, neither dust mass estimation nor ELISA analysis was possible.

Statistical analysis

Results were examined pairwise for correlations between different sampling methods. Due to the non-normal distribution of the data, Spearman correlation analyses were used.

Results

All sampling devices recovered measurable quantities of dust and allergen from all carpets. Data for the Reference Method, Mitest, ECS and PTS were expressed both as a mass concentration (µg/g) and as an area concentration (µg/m²). The geometric mean, maximum and minimum Der p 1 concentrations, as determined by these four collection systems, are shown both as µg/g and µg/m² in Table 1. The total dust mass and the total amount of Der p 1 collected by each sampling system is also shown in Table 1.

All results for the Dräger Bio-Check were 1 or greater. As the minimum concentration for the reference method was 22 µg/g which exceeds the reported level for a class 3 reading from the Dräger Bio-Check device (16), all data from this device were excluded from the primary correlation analysis.

Spearman correlations for expression of results in units of Der p 1 mass concentration (µg/g) are shown in Table 2A. A significant correlation was found between the Reference Method and the other three devices; the Mitest, the ECS and the PTS. However, none of the other systems correlated with each other.

The Spearman correlations between the different sampling devices with data expressed as Der p 1 area concentration (µg/m²) are shown in Table 2B. Significant correlations were as follows: the Reference Method with the Mitest and ECS; the PTS with the Mitest and ECS; and the ECS with the Mitest.

In order to investigate the Dräger Bio-Check results we performed a secondary analysis following the method of Polzius et al. (16). Each Dräger Bio-Check result (read as 3, 2, 1 or 0) was used to place the corresponding Reference Method measurement of Der p 1 from the same location into one of four categories corresponding to the Dräger Bio-Check result. We then tested for significant differences in the median Reference Method readings between the categories. No significant differences were seen, indicating that the Dräger Bio-Check results were not significantly related to the Reference Method results.

Discussion

This study shows that four different methods can be used to collect samples of allergen from carpets and that many

Table 1. Results for Der p 1 mass concentration, Der p 1 area concentration, total dust mass and total Der p 1 collected for each sampling method

	Der p 1 mass concentration (µg/g)	Der p 1 area concentration (µg/m ²)	Total dust mass collected (g)	Total Der p 1 collected (ng)	Area sampled (m ²)
Reference Method	67.20 (22.27–621.35)	4.24 (0.68–27.59)	0.0631 (0.0215–0.1526)	4242.24 (678.94–27587.94)	1
Mitest	28.79 (6.00–138.82)	58.67 (11.43–322.43)	0.5095 (0.1520–1.0739)	14667.44 (2857.56–80606.40)	0.25
Electrostatic Cloth Sampler	26.52 (6.33–166.04)	1.44 (0.38–19.46)	0.0135 (0.0022–0.0422)	359.00 (94.48–4864.50)	0.25
Press Tape Sampler	2.78 (0.46–12.81)	13.92 (2.08–81.50)	0.0125 (0.0034–0.0265)	34.81 (5.21–203.75)	0.0025
Dräger Bio-Check					0.0624

Data given as geometric mean (minimum–maximum). The Dräger Bio-Check is read on an arbitrary ordinal scale.

Table 2. Spearman correlation results for Der p 1 (A) mass concentration ($\mu\text{g/g}$) and (B) area concentration ($\mu\text{g/m}^2$): r_s statistic (P -value). Significant correlations are in bold

	Reference Method	Mitest	Electrostatic Cloth Sampler	Press Tape Sampler
(A)				
Reference Method	1			
Mitest	0.74 (0.0010)	1		
Electrostatic Cloth Sampler	0.62 (0.0103)	0.37 (0.1577)	1	
Press Tape Sampler	0.55 (0.0283)	0.28 (0.2946)	0.37 (0.1612)	1
(B)				
Reference Method	1			
Mitest	0.64 (0.0071)	1		
Electrostatic Cloth Sampler	0.58 (0.0194)	0.52 (0.0374)	1	
Press Tape Sampler	0.36 (0.1723)	0.65 (0.0067)	0.70 (0.0027)	1

of the results correlate to some degree, although no two methods correlate completely.

It is recognized that mite allergen is widely distributed within the structure of carpets, with the greatest quantity, but the least concentration per weight of carpet fibers, at the base (34, 35). Mite allergens are associated with a broad range of particle sizes, at least in the aeroallergen fraction (36). Even the large particles and fibers present in dust, likely to be removed by sieving, are associated with considerable amounts of mite allergen (37). It is also recognized from earlier studies that repeated sampling using the same reference method, on the same and on different sites, involves considerable variance in measured allergen concentration. The different methods used here presumably increase variation by using different physical processes relating to the removal and retention of the particles carrying mite allergens. This variation is apparent both from the concentration of allergen per unit weight and per unit area.

The results for Der p 1 obtained by the Mitest and the Reference Method, which both involve the similar concept of vacuum sampling, differed markedly in magnitude, although they correlated. When data was expressed as a mass concentration, the Reference Method recorded the greater concentration, and when expressed as an area concentration, the Mitest recorded the greater concentration. In the Mitest sampler, all of the dust collected is extracted – no separation is made between fine and coarse dust. Hence, it would take only a few large non-allergen carrying particles to drastically reduce the Der p 1 mass concentration reported by the Mitest. The Mitest also uses a more powerful mains-powered vacuum cleaner, compared with the battery-powered cleaner of the Reference Method. This could have been the reason for the substantially higher readings seen when Der p 1 was expressed as an area concentration, presumably reflecting the greater harvesting of allergenic dust from deep in the carpet by the mains-powered vacuum cleaner. Both the Der p 1 area concentration and the total dust mass collected using the Mitest were around 10-fold greater

than those collected using the Reference Method. Consistent with our findings, Lewis and Breysse (21) found that a modified mains-powered vacuum cleaner collected four times as much mite allergen per area of carpet, and twice as much total dust, as a hand-held model.

In these experiments, the ECS collected a similar mass concentration of Der p 1 to the Mitest. Although this did not correlate with the Mitest, it did correlate with the Reference Method. Interestingly, the ECS reported the lowest area concentration of Der p 1 of the four quantifiable methods. Here a correlation was seen with the Reference Method, the Mitest and the Press Tape. Swabbing with a cotton cloth has previously been used as a means of collecting cockroach allergen in homes (38), and both moistened paper (39) and cotton gauze swabs (40) have been used to sample lead from carpets in houses. In a recent field trial by us using the ECS, where the devices and instructions were distributed by post, about 10% of the returned devices had lost weight due to removal of the ECS fibers when they were used too vigorously (A. van Overveld, personal communication, 2002), hence minor design modifications may be required. In the present study, it is likely that a combination of suboptimal allergen capture by the ECS and the loss of fibers from the ECS led to an inflated allergen mass concentration and a reduced allergen area concentration.

The PTS collected dust with the lowest mass concentration of Der p 1 and this probably reflects the disproportionate weight of adhered carpet fibers diluting the captured fine dust. As an area concentration, however, the PTS gave what appears to be reasonably high readings due to the small area from which the samples were taken with this device. The PTS did not correlate with the Reference Method in terms of an area concentration, but did correlate as a mass concentration. Other investigators have used adhesive materials to sample surfaces for lead, demonstrating better collection efficiencies than wipe samples when used on rough surfaces (41). When we used a similar device to sample cat allergen on clothes, numbers of allergenic particles recovered with adhesive tape correlated better with measurements of inhaled allergenic particles than did the allergen content of dust collected from the clothes by the Reference Method (27).

Our experience with the Dräger Biocheck does not agree with that of Polzius et al. (16) who reported that the readings from this device, using four categories of line intensity, were significantly related to the median of the corresponding Der p 2/f 2 and Fel d 1 ng/g values obtained using a modification of the Mitest procedure. These authors, however, also observed wide ranges of Mitest results corresponding to each Biocheck result category (16). Our evaluation of the Biocheck in an environment where mite allergen levels were much higher may have contributed to our finding of no agreement between reservoir levels and the Biocheck result. In our tests the Reference Method recovered a geometric mean of 67.2 $\mu\text{g/g}$ of Der p 1, compared with a geometric mean of

8 µg/g of Der p 2/f 2 recovered by the vacuum method in the highest category of Dräger Bio-Check results as reported by Polzius et al. (16). In addition, these authors used a sampling area of 0.1 m² and eluted in five drops of buffer, which differs from the instructions packaged with the devices. It is unlikely that our analysis would have been affected by our comparison of Der p 1 with the Biocheck Group 2 allergens, as Der p 1 and Der p 2 correlate significantly (15) and *D. farinae* is rare in Sydney.

At present there is no single validated method of sampling reservoir dust to maximize its relevance to clinical outcomes such as allergic symptoms or sensitization. Caution should be exercised in comparing data collected with different devices or using different units of expression. It is also clear that different results can occur with similar methods, and the ranking of the allergen content of sites changes depending on how data is expressed. For some purposes expression as area concentration (µg/m²) is favored (26). Indeed, where the mass or composition of sampled dust is known to have been changed by an intervention, µg/m² is most suitable (6). However, some investigations have supported the concept that in general, household allergen samples are best expressed as a mass concentration (µg/g) (25) and many studies use this measure.

When commercial devices are used to collect allergen samples to be processed in the home by a rapid immunoassay it is unlikely that the mass of dust collected will be recorded, therefore only an area concentration may be assessed. In addition to the Mitest Dust Collector there are at least three other commercially available home allergen sampling devices which use vacuum collection methods (42–44). None of these feature dust weight measurement as part of the assay process, although it could possibly be performed if samples were sent to a laboratory. The ECS and the PTS that we have developed both present the option of weight measurement if returned to a laboratory for processing. This may be an advantage in epidemiological studies where laboratory

processing of a large number of subject-performed samples is achievable.

The effect of reservoir allergen concentration on airborne exposure has also not been established. More than 30 studies have included an examination of the relationship between reservoir allergen and airborne allergen (reviewed by O'Meara and Tovey) (45). Overall such relationships rarely exist and when they do, are weak. Recently, Gore et al. published a study indicating a weak correlation between the number of allergen-carrying particles inhaled on a series of nights in bed and the reservoir allergen content of each bed expressed as µg/g (46). Extensions of this type of experiment could provide more information on the relationship between different measures of reservoir allergen and clinically relevant exposure.

In the estimation of allergen exposure, the availability of simple, inexpensive measures of reservoir concentration increases the scope of research projects by facilitating the collection of dust samples by patients, and can increase patient education and compliance (12, 16, 47, 48). Currently, both commercial at-home kits and laboratory-based methods of sample collection and assay for allergens are available. Results obtained using many of these methods may be comparable to varying degrees. If the requirement is simply to be able to distinguish high from low allergen levels at a site, then all of the methods are acceptable. If great importance is to be placed on small differences, then no method has been proven to be more reliable. Where cost, convenience and transport of samples is important, then self-collection of dust by subjects using small hand-held samplers, as described here, has practical advantages.

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