

Mite allergen (Der p 1) is not only carried on mite feces

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Exposure to allergens derived from house dust mites (eg, *Dermatophagoides pteronyssinus*) is considered an important factor in the development and exacerbation of asthma. In cultures, more than 95% of mite allergen Der p 1 was associated with mite feces (mean diameter of $22 \pm 6 \mu\text{m}$; range, 10 to $40 \mu\text{m}$).¹ Domestic air sampling in bedrooms during dust disturbance showed that greater than 80% of detectable Der p 1 was associated with particles larger than $10 \mu\text{m}$ and only a small proportion with particles smaller than $5 \mu\text{m}$,^{2,3} the identity of which is not known. To explain how exposure to such large particles ($>10 \mu\text{m}$) could cause asthma, it was proposed that a low proportion of these large fecal particles would enter the lower airways, and the effects of high local allergen concentrations from each particle, on chronic exposure, might provide sufficient stimulus to maintain or produce hyperreactivity.² There is also some direct evidence of mite allergen, albeit in low concentrations, in the lungs of asthmatic subjects.⁴ We investigated the sources of Der p 1 aeroallergen by using a new technique, which enables individual particles containing allergen to be visualized.

METHODS

Airborne particles were sampled with intranasal air samplers (Institute of Respiratory Medicine, Sydney, Australia), which collect particles larger than $5 \mu\text{m}$. Samples were collected in parallel with a lapel-mounted filter (Institute of Occupational Medicine [IOM], Edinburgh, UK) and a vacuum pump, which achieved a flow rate of 1.8 L/min with the filter in place.

Each sample was collected for 10-minute periods, and 4 nasal samplers and 6 filter samples were collected while brushing the carpet once every 30 seconds. Within the nasal sampler, particles were collected onto a polyvinylidene difluoride membrane and coated with a thin film of an aqueous-based adhesive that was later overlaid with a thin layer of agarose to fix captured particles in position. In the IOM sampler, particles were collected by suction onto a dry polyvinylidene difluoride membrane (pore size, $0.45 \mu\text{m}$), which was later overlaid with transparent adhesive tape to prevent particle movement. Particles captured for both IOM and nasal air samplers

Abbreviation used

IOM: Institute of Occupational Medicine

were immunostained with Der p 1-specific mAb (Indoor Biotechnologies, University of Virginia) followed by an anti-mouse alkaline phosphatase conjugate and developed by using 5-bromo-4-chloro-3-indolyl phosphate/nitro blue tetrazolium substrate. The total number of particles and those containing Der p 1 were counted, and their sizes were measured manually by using an eye piece graticule. On the basis of their appearance, allergen-containing particles were described as being either fecal (spherical, rough, dense appearance), fibers (threadlike), or flakes (flat, translucent particles).

RESULTS

A total of 242 particles containing Der p 1 were collected on the nasal samplers over 40 minutes (55% of the total number of particles captured) (Fig 1, A). The type of particle, frequency, and largest diameter were as follows: feces, 36% (range, 15 to $40 \mu\text{m}$) (Fig 1, B); fibers, 34% (range, 10 to $150 \mu\text{m}$) (Fig 1, C); and flakes, 30% (range, 5 to $50 \mu\text{m}$) (Fig 1, D). The median size of all particles containing mite allergen was $25 \mu\text{m}$. The reservoir concentration of Der p 1 in the fine carpet dust was $152 \mu\text{g/g}$ dust. By using the IOM sampler, a total of 71 particles that contained Der p 1 were collected over 60 minutes. The nature and size of the airborne particles carrying Der p 1 were similar, although the distribution differed as follows: feces, 45% (range, 10 to $40 \mu\text{m}$); fibers, 11% (range, 10 to $85 \mu\text{m}$); and flakes, 44% (range, 3 to $80 \mu\text{m}$). Judged by the intensity of immunostaining, feces contained the most allergen, fibers intermediate amounts, and flakes the least.

DISCUSSION

The results of these experiments suggest that mite aeroallergen is carried on a variety of particles, including small particles, and not exclusively on mite feces. The deposition of particles in the lung is determined by their aerodynamic diameter. For spherical particles, such as mite feces, their aerodynamic diameter would be similar to their size. However, fibers and flakes would behave as particles smaller than their largest dimension would suggest. For example, long fibers behave as particles of a size equivalent to approximately 3× their width. Many of the fibers and flakes observed here would have an aerodynamic diameter less than $10 \mu\text{m}$, which would therefore increase their like-

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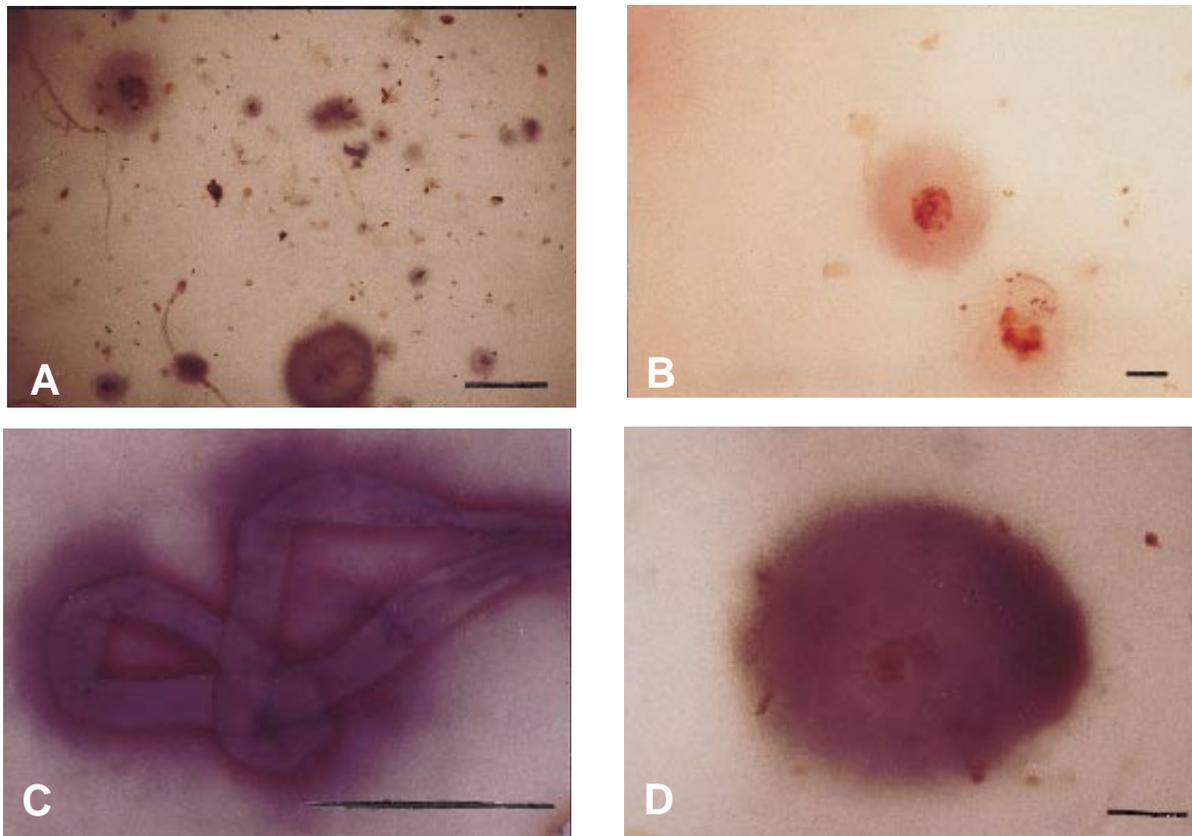


FIG 1. **A**, Representative sample of inhaled particles collected by the nasal impaction sampler after immunostaining with Der p 1-specific mAb. Only particles eluting Der p 1 are visualized, with purple staining around them. (*Bar* = 150 μ m at 100 \times magnification). **B**, Inhaled mite fecal particle (*Bar* = 40 μ m). **C**, Inhaled fiber eluting Der p 1 (*Bar* = 45 μ m). **D**, Inhaled flake particle eluting Der p 1 (*Bar* = 50 μ m)

likelihood of lung deposition. However, it would appear, in agreement with previous reports,^{2,3} that only a small quantity of allergen is carried on these smaller particles.

There are a number of possible mechanisms by which mite allergen becomes associated with these particles. There may be active secretion of gut contents during feeding, a hypothesis supported by the immunostaining of Der p 1 in the esophagus of mites.⁵ Another mechanism may be that in humid conditions Der p 1 allergen elutes from feces onto nearby particles.

In summary, we have identified feces, fibers, and flakes as sources of inhaled mite aeroallergen. Although the majority of allergen is carried on larger particles, some is carried on smaller particles and on heterologous particles, which behave aerodynamically as small particles. These particles could be inhaled more readily into the lungs and cause airway inflammation.

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