
Efficacy of dry-cleaning in removing Fel d 1 allergen from wool fabric exposed to cats

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Background: The main cat allergen (Fel d 1) is ubiquitous, having been found even in indoor environments and public places where a cat has never been kept. Clothes of cat owners constitute a carrier for the distribution of Fel d 1 allergen in these environments. Schools, for example, may be a site of indirect exposure to cat allergens.

Objective: Our goal was to investigate the efficacy of commercial dry-cleaning in removing cat allergens from wool fabrics that had been exposed to cats to evaluate a possible preventive procedure.

Methods: Twenty-six identical wool "squares" (80 × 100 cm) were put in cat baskets for 1 week. In our laboratory, the squares were cut in half (40 × 50 cm), and one half was subjected to high-volume sampling for 5 minutes in a cat-free room. The other half was subjected to commercial dry-cleaning and then the high-volume sampling. Five wool squares not exposed to cats served as controls. Dust was collected from the wool squares with a high-volume air sampler. Particulate material was harvested onto glass fiber filters (AP 20 Millipore, Milan, Italy) with 25-mm diameter and 2-μm pore size. Each dust sample was assayed by affinity-purified monoclonal antibody against purified Fel d 1. The results were expressed as micrograms per filter. Statistical analysis was done by using the paired *t* test.

Results: Before dry-cleaning, Fel d 1 allergen was detected on all cat-exposed wool squares. No appreciable cat allergen was detected on control materials. After commercial dry-cleaning, the amounts of Fel d 1 extracted from cat-exposed squares were significantly reduced (*t* = 14.63; *P* < 0.001) but not abolished. Three of the five control squares were contaminated by Fel d 1.

Conclusions: Commercial dry-cleaning effectively removes large amounts of cat allergen from wool materials exposed to cats but does not completely abolish this protein. Further, low Fel d 1 contamination may occur during this procedure.

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INTRODUCTION

Exposure to allergens derived from domestic animals, particularly cats, is a risk factor for airway sensitization and may trigger exacerbations of asthma in sensitized patients.^{1–4} Recent studies demonstrated that the major cat aller-

gen, Fel d 1, is ubiquitous, being found in dust samples from private homes and many different indoor environments, including public places where a cat has never been kept.⁵ These places that should be cat-free include day nurseries, hospitals, day care centers, means of transport, and, especially, classrooms.^{6–13} This indirect exposure to pet allergens is important from a clinical standpoint because it might be of sufficient magnitude to induce or maintain respiratory symptoms in sensitized persons.¹⁴ Further, Bollinger et al¹⁵ demonstrated that the low level of exposure to cat allergens that occurs in many indoor environments without

cats may be able to induce or maintain allergic symptoms in some cat-sensitized patients.

We¹⁶ and Patchett et al¹⁷ previously showed that wool clothes constitute an important means for the distribution of Fel d 1 in cat-free environments. This finding has been confirmed by other authors as well.^{10,18} Investigators have also demonstrated that cat allergens may be transferred from the clothes of cat owners to the clothes of those who do not own a cat.¹⁰

On the basis of this background, removal of cat allergens from the clothes of cat owners should be considered a relevant approach to attempt to reduce the dispersal of cat allergens in cat-free environments and, consequently, the degree of indirect exposure to these allergens.

Collectively, the foregoing data indicate that it is crucial to determine the effectiveness of various methods to clean fabrics which have been directly or indirectly exposed to cats for optimal removal of cat allergens. Recently, we demonstrated that the washing of allergen-containing cotton fabrics in water, without the use of detergents, completely eradicated Fel d 1 from these materials.¹⁹ In many industrialized countries, however, commercial dry-cleaning is widely used for some types of garments and is recommended for some fabrics. The aim of the current study was to evaluate the efficacy of commercial dry-cleaning in removing cat allergens from wool fabrics that had been exposed to cats.

MATERIALS AND METHODS

Materials

Because it was not possible to ensure identical allergen exposure to wool clothes (in light of the difficulty with

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such factors as obtaining identical clothing and providing reliable identical exposure in situ), we approached the problem by using 26 identical rectangles of a widely used wool material (80×100 cm; approximately 2.5 mm thick). These wool “squares” were obtained from a single bale (2×15 m), which was stored within a plastic cover to avoid any contact with domestic animals.

After each square was cut, it was put in a single plastic bag to avoid contamination by cat allergens before the experimental exposure, and then it was given to a cat owner. We used wool material because Patchett et al¹⁷ demonstrated that wool clothing may contain higher Fel d 1 levels than cotton garments. The wool squares were put in the baskets of 26 male cats for 1 week; male cats were selected because the production of Fel d 1 is higher in male cats than in female cats.²⁰

Cat owners were asked to rearrange the squares daily to ensure total contact of the entire square with the animals. The squares were transferred from the cat owners’ homes to our laboratory in new plastic envelopes to avoid contamination by cat allergens, loss of allergens, or both.

The fabric squares were cut in half (40×50 cm); dust was sampled from one half by high-volume sampling by

vacuuming for a standardized time (5 minutes) in a cat-free room of our laboratory. This procedure was performed by direct sampling (with the device at an angle of approximately 90°) of both surfaces of each wool web.

For all experiments, the high-volume sampling was performed by the same laboratory personnel. Although these personnel had no cats in their homes, they were asked to change their clothing and to wear sterilized laboratory coats to avoid contamination of the cat-free room by cat allergens transferred by clothes. The high-volume sampling was done after each square was placed on a table covered by a sterilized sheet. After each sampling, this sheet was changed to avoid contamination of the next wool web.

The other half of each wool square was subjected to routine commercial dry-cleaning (steeping in perchloroethylene, spinning, and drying at 60° to 80° C). To our knowledge, the procedure and perchloroethylene used for commercial dry-cleaning are identical throughout the world. Each web was transferred to and from the dry-cleaners in a new plastic bag to avoid contamination by cat allergens. The dry-cleaned squares were sampled for 5 minutes by the same modalities and personnel in the cat-free room of our laboratory. Five squares (exactly the

same material and size but not exposed to cats) served as controls to exclude previous Fel d 1 contamination.

One-half of each of the 26 wool squares exposed to cats and one-half of each of the five control squares were sent for commercial dry-cleaning on different days to avoid contact between them. The study design is shown in Figure 1.

High-Volume Sampling

Dust was collected from the wool squares with a high-volume vacuum air sampler (CF/20 Gelaire Flow Labs, Milan, Italy), consisting of a double-head membrane pump connected with two flow-control flowmeters and a volumetric counter. Airflow was constant (20 L/minute), and particulate material was harvested onto glass fiber filters with 25-mm diameter and $2\text{-}\mu\text{m}$ pore size (borosilicate microfiber glass with acrylic resin binder; AP 20 Millipore, Milan, Italy).

The device traps particles with a diameter that exceeds 2 to $3\text{ }\mu\text{m}$ with an 84% efficiency. To reduce the risk of contamination, each filter was placed in a separate sealed tube. The device for the dust collection was also kept in a sealed box.

Among various methods available to capture allergens from a piece of material (eg, use of extraction fluids), we chose high-volume sampling because of its ease of use. In addition, this procedure has been used in earlier studies.^{16,19}

Assay of Allergens in Vacuumed Dust Samples

Each vacuumed dust sample was supplemented with 2 mL of phosphate-buffered saline in conjunction with 0.2% bovine serum albumin. After centrifugation for 10 minutes at 10,000 rpm and filtration through a $0.45\text{-}\mu\text{m}$ pore diameter membrane, the supernatants obtained were stored at -20° C until required for assay.

The membrane used for filtration after extraction (Millex Millipore filters) had a low protein-binding capacity; in addition, the buffer had 0.2% bovine serum albumin. Thus, the changes with

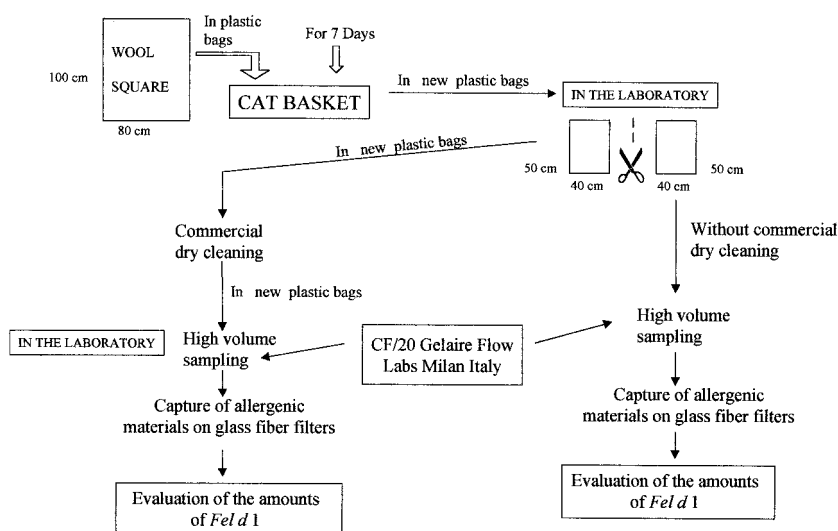


Figure 1. Overview of study design.

specific Fel d 1 adsorption were very low. Fel d 1 content was determined with a previously described two-site enzyme-linked immunoadsorbent assay method, with a slight modification.²¹

The specificity of the anti-Fel d 1 monoclonal antibody (MAb) and of the Fel d 1 immunoassay itself is well documented, and the presence of nonspecific antibody binding has been eliminated.^{22,23} Nevertheless, in all the assays, a negative control (sample dilution) was included. Affinity-purified MAb (5/24) was used at a concentration of 10 µg/mL to coat a polystyrene plate. A standard curve (0.2 to 150 ng/mL) was determined from a freeze-dried cat dander extract titrated against purified Fel d 1. Standard and samples diluted to the appropriate concentration were incubated for 2 hours. After washing, biotin-labeled secondary MAb (C5/8) was added at a dilution 1:1,000. After incubation and washing, streptavidin peroxidase (Amersham, Uppsala, Sweden) was added, at a dilution of 1:5,000, and the enzymatic activity was evaluated by using *o*-phenylenediamine dihydrochloride as substrate.

The allergen concentration was estimated from duplicated absorbances of three different dilutions interpolated into the standard curve. The mean intraassay coefficient of variation was <2%, and interassay variation versus a control was <7%.

Statistical Analysis

Statistical significance was evaluated by using the paired *t* test, mean values, and 95% confidence interval of the differences on data normalized by log conversion.

RESULTS

Before dry-cleaning, Fel d 1 (expressed as log Fel d 1 in micrograms per filter) was detected on all cat-exposed wool squares (Table 1). In contrast, non-dry-cleaned nonexposed control wool squares did not contain appreciable amounts of Fel d 1 (<0.005 µg of Fel d 1 per filter, which is considered the lower detection lim-

it). As shown in Figure 2, after commercial dry-cleaning the amounts of Fel d 1 extracted from cat-exposed squares were significantly reduced (*t* = 14.63; *P* < 0.001; 95% confidence interval = 1.04 to 138) but not abolished. Interestingly, after dry-cleaning, 3 of the 5 nonexposed control wool squares were contaminated by Fel d 1 (0.020, 0.020, and 0.009 µg/filter, respectively).

DISCUSSION

In many industrialized countries, the increasing frequency of keeping furred pets (especially cats and dogs) indoors has raised the degree of exposure to allergens derived from these animals.²⁴⁻²⁶ A significant correlation has been reported between the community prevalence of cat ownership and the

community prevalence of specific sensitization to cats.^{25,26}

In northern Europe, an estimated 23 to 34% of schoolchildren own a dog or a cat.²⁷ In addition, sensitization to cats or dogs (or both) has been reported in 15 to 20% of schoolchildren 11 to 13 years of age,²⁸ and 50 to 70% of children with asthma are sensitized to domestic animals.²⁹ Further, pets (not mites) are also the most important sensitizing allergens in Swedish adults.²⁴

In the past few years, several studies have demonstrated that exposure to allergens of furred pets may be relevant in many indoor settings that should be cat-free environments. Classrooms have been demonstrated to be a major site of exposure to pet allergens because the levels of these allergens strongly depend on pet ownership.

Table 1. Fel d 1 Allergen* in 26 Cat-Exposed and 5 Control Wool Squares Before and After Dry Cleaning†

Exposed wool squares‡			Control wool squares		
Sample	Before DC	After DC	Sample	Before DC	After DC
1	0.92	0.27	1	<0.005	<0.005
2	1.46	0.27	2	<0.005	<0.005
3	8.27	0.26	3	<0.005	0.020
4	13.03	0.19	4	<0.005	0.020
5	5.94	0.61	5	<0.005	0.009
6	4.92	0.32			
7	22.19	0.22			
8	2.97	0.20			
9	4.76	0.28			
10	1.62	0.54			
11	7.79	0.17			
12	8.52	0.35			
13	10.80	0.20			
14	9.30	0.55			
15	3.29	0.93			
16	12.69	0.54			
17	21.39	1.05			
18	7.09	0.54			
19	6.09	0.26			
20	5.92	0.52			
21	3.95	0.47			
22	17.60	0.23			
23	3.58	0.30			
24	4.95	0.68			
25	28.14	0.70			
26	1.80	0.32			
Mean value	8.42	0.42		0.005	0.0118
DS	7.01	0.23		0	0.007

* Expressed as log Fel d 1 (micrograms/filter).

† DC = dry cleaning; DS = degree of scatter (standard deviation).

‡ Significantly reduced (*P* < 0.001) after dry cleaning.

Various studies have suggested that the clothes of pet owners constitute the main factor for the distribution of pet allergens in animal-free environments. As a consequence, no indoor environment may be considered completely free from pet allergens. Recently, Egmar et al³⁰ demonstrated that even during a 3-week period, pet allergens accumulate in the environment if the personal allergen load is sufficient, despite the absence of pets. In many susceptible persons, even low levels of allergens are able to maintain or worsen airway sensitization.

In light of the high level of cat ownership in industrialized countries, the only practicable way to reduce the transport and the dispersal of cat allergens is adequate cleaning of contaminated clothes worn by cat owners outdoors.

The results of our study suggest that routine commercial dry-cleaning is effective for removal of large amounts of cat allergen from wool squares exposed to cats. Nevertheless, residual amounts of allergen have been found on the material tested. Further, Fel d 1 contamination of three control (previously nonexposed) wool fabrics after dry-cleaning suggests the possibility that cat allergens can be transferred from other clothes during the dry-cleaning process. Because wool fabrics were transferred to and from commercial dry-cleaning by using different plastic bags, we excluded the possibility of contamination of control materials both before and after the dry-cleaning process.

In contradistinction, in a previous investigation conducted with cat allergen-contaminated cotton squares,¹⁹ we demonstrated that washing in water completely abolished Fel d 1 allergen from cotton fabrics. In our earlier study, we used the same exposure, transport, division, and sampling procedures as in the current investigation. The same high-volume vacuum air sampler (CF/20 Gelairé Flow Labs) was also used. Further, the amounts of the Fel d 1 allergen found on the cotton squares before washing in water were comparable with those found in the

current study: between 1.35 and 23.40 $\mu\text{g}/\text{filter}$.

Although we can not directly compare these two studies because one was conducted with cotton material that was washed in water and the other with wool material that was dry-cleaned, washing in water seems more effective in removing cat allergens from fabrics.

Whether the residual cat allergens could cause airway sensitization or could trigger symptoms of asthma in cat-allergic patients is difficult to evaluate. To our knowledge, no published studies have correlated the levels of cat allergen contamination of clothes and the clinical activity of asthma. Most likely, however, the amounts of Fel d 1 allergen remaining on wool fabrics after commercial dry-cleaning are of sufficient magnitude to be transferred to cat-free environments and, consequently, to contribute to airway sensitization.

The persistence of Fel d 1 after dry-cleaning indicates that perchloroethylene does not modify the characteristics of cat allergen. Similarly, Vandenhove et al³¹ demonstrated that mite allergens were not denatured by perchloroethylene and, therefore, that the effect of dry-cleaning on mite allergen levels in blankets resulted from the physical washing out of dust and allergens.

CONCLUSION

An efficient cleaning of clothes of cat owners should be included in the pre-

ventive strategies for controlling the indirect exposure to cat allergens. We conclude that commercial dry-cleaning is effective in eliminating, for all practical purposes, these allergens from contaminated fabrics but does not completely abolish this protein. Further studies should be conducted to evaluate alternative methods for cleaning pet-exposed materials or to improve the actual procedures of dry-cleaning.

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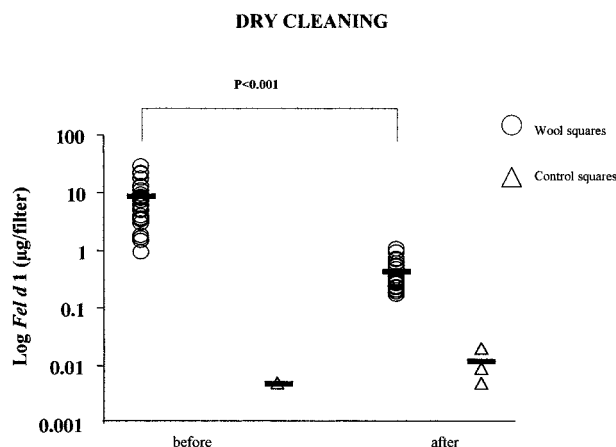


Figure 2. Concentration of Fel d 1 allergen expressed as log Fel d 1 ($\mu\text{g}/\text{filter}$) on 26 cat-exposed wool squares and 5 control wool squares before and after dry-cleaning.

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