

# EFFECTIVENESS OF VACUUM-CLEANING IN REMOVING FEL D 1 ALLERGEN FROM COTTON FABRICS EXPOSED TO CATS

G. Liccardi (1), D. Barber (2), M. Russo (1), G.-W. Canonica (3),  
G. D'Amato (1), G. Senna (4), G. Passalacqua (3)

**Background:** The major cat allergen, Fel d 1, is ubiquitous, and clothes of cat owners are important vehicles for its distribution and dispersal in cat-free environments.

**Objective:** We assessed the efficiency of a standard vacuum-cleaning in removing the cat allergen from cotton fabrics, experimentally contaminated by a prolonged contact with cats.

**Methods:** Fifteen rectangles (80x100 cm) of a widely used cotton material were kept in baskets of male cats for 1 week. Afterwards, the squares were cut in half. One half was immediately sampled for cat allergen, and the other half was vacuum-cleaned for 15 minutes by a commercially available device before sampling. Five identical cotton rectangles not exposed to cats served as controls. Samples were collected with a high-volume vacuum air-sampler. Particulate material was harvested onto filters with 25- mm diameter and 2 µm pore size, extracted in phosphate buffer with BSA and then assayed for the Fel d 1 allergen with an ELISA method. The results were expressed as micrograms of allergen per filter.

**Results:** After vacuum-cleaning the amounts of Fel d 1 extracted from cat-exposed fabrics were not significantly reduced ( $t = 3.118$ ;  $P > 0.005$ ), with respect to the non vacuum-cleaned tissues. No Fel d 1 was found in control fabrics.

**Conclusions:** Our study suggest that vacuum-cleaning by using a commercially available device is not able to remove significant amounts of cat allergen from cotton fabrics exposed to cats.

Allergen avoidance - Cat allergen - Fel d 1 - Respiratory allergy - Vacuum-cleaning.

## INTRODUCTION

Exposure to allergens derived from domestic animals, particularly cats, is a risk factor for airway sensitization and may trigger respiratory symptoms in already sensitized patients (1, 2). Cats are an important indoor allergenic source, because cats are commonly owned as domestic animals. The major cat allergen, Fel d 1, is ubiquitous, being present in dust samples of the environments where the cat lives, but also in homes and other indoor environments, including public places, where a cat has never been kept (3, 4). The indirect exposure to cat allergen is important from a clinical standpoint since also low levels of exposure may be able to induce allergic sensitization or to trigger nasal/bronchial symptoms in sensitized individuals (5).

It is now widely recognized that clothes of cat owners represent an important vehicle for the transport and dispersal of Fel d 1 in cat-free environments (6). Consequently, removal of cat allergen from the clothes of cat owners would be considered a logical

approach to reduce the dispersal of Fel d 1 and, consequently, the degree of indirect exposure to these allergenic materials. The aforementioned data suggest that it is crucial to clearly assess the efficiency of various methods to clean fabrics which have been directly or indirectly exposed to cats for optimal removal of cat allergens.

Since standard vacuum-cleaning is a simple and widely used procedure in allergen avoidance, we aimed to investigate its efficacy in removing cat allergen from cotton fabrics contaminated by a prolonged contact with cats.

## METHODS

### Materials

We used 15 identical rectangles of a widely used cotton material (80x100 cm, approximately 2 mm thick), prepared in a cat-free environment and kept in single plastic envelopes to avoid contamination by cat allergens before the experimental exposure. These fabrics were given to cat owners, and put in baskets of male cats for 1 week. Male cats were selected because the production of Fel d 1 is higher in male than in female cats.

(1) Department of Chest Diseases, Division of Pneumology and Allergology - A. Cardarelli " Hospital, Naples (Italy).

(2) Department of R&D, Abello S.A, Madrid (Spain).

(3) Allergy and Respiratory Diseases, DIMI, University of Genoa (Italy)

(4) Allergy Service, Verona General Hospital (Italy).

The squares were then transferred from the cat owners' homes to our laboratory in new plastic envelopes to avoid further contamination or loss of allergens. The fabric squares were cut in half (40x50 cm). One half was immediately sampled for assessment of level of Fel d 1 allergen (see below). The other half was vacuum-cleaned on both sides for an arbitrary time of 15 minutes with a commercially available vacuum-cleaner (VL 1840, Black & Decker Corporation, Towson, MD), before sampling for allergen level. Five rectangles (exactly the same material and size but not exposed to cats ) served as controls to exclude previous Fel d 1 contamination.

## Sampling

Collection of dust samples from the cotton pieces was performed by means of a high-volume vacuum- air sampler applied orthogonally to both sides of the cotton for 5 minutes in a cat-free room as previously described (6-8). The high-volume vacuum air sampler (CF/20 Gelaire Flow Labs, Milan Italy), consists of a double-head membrane pump connected with two control flow-meters and a volumetric counter. Airflow was kept constant at 20 L/minute. The particulate material was harvested onto glass-fiber filters with 25 mm diameter and 2  $\mu$ m pore size (borosilicate microfiber glass with acrylic resin binder; AP 20 Millipore, Milan Italy). This device entraps particles with a diameter that exceeds 2 to 3  $\mu$ 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.

## Measurement of Fel d 1

Each collected 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  $\mu$ m pore diameter membrane, the supernatants obtained were stored at - 20°C until required for testing. The membrane used for filtration after the extraction (Millex Millipore filters) had a low protein-binding capacity; in addition, the buffer contained 0.2% bovine serum albumin. Thus, the chances of an specific Fel d 1 adsorption were very low.

Fel d 1 content was determined with a previously described two-site enzyme-linked immunosorbent assay method, with a slight modification (9). The specificity of the anti-Fel d 1 monoclonal antibody (Mab) utilized, and of the Fel d 1 immunoassay is well documented. Affinity-purified Mab (C5/24) was used at a concentration of 10  $\mu$ g/mL to coat a polystyrene plate. A standard curve (1 to

75 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 1:1,000 dilution. After incubation and washing, streptavidin peroxidase (Amersham, UK) was added, at a dilution of 1:5,000, and the enzymatic activity was evaluated by using O-phenylenediamine dihydrochloride as chromogen. The allergen concentration was estimated from duplicate absorbances of three different dilutions interpolated into the standard curve range.

## 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

The non-vacuum-cleaned and nonexposed five control cotton squares did not contain appreciable amounts of Fel d 1 (< 0.05  $\mu$ g of Fel d 1 per filter, lower detection limit).

Before vacuum-cleaning, Fel d 1 (in micrograms per filter) was detected on all cat exposed cotton squares. The amount of Fel d 1 was 0.53  $\pm$  1.23 micrograms per filter in the vacuumed tissue samples, whereas it was 0.26  $\pm$  1.35 in the samples not vacuum-cleaned. As shown in figure 1, no significant difference could be therefore seen after vacuum-cleaning in the amounts of Fel d 1 extracted from cat-exposed fabrics ( t = 3.118 ; P > 0.005 ).

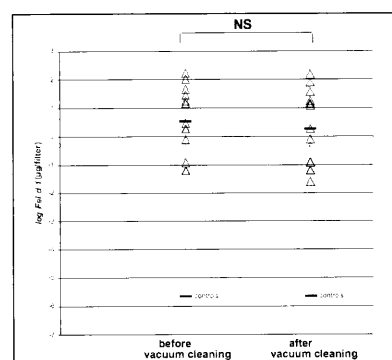


**Figure 1:** Concentration of Fel d 1 allergen expressed as log Fel d 1 ( $\mu$ g/ filter) on 15 cat exposed cotton squares and 5 control cotton squares before and after vacuum- cleaning.

## DISCUSSION

Several studies have demonstrated that exposure to allergens derived from cats or dogs may be relevant in many indoor settings that should be animal-free, (3, 4) and that no indoor environments can be considered completely free from pet allergens. In addition, in susceptible individuals, even low levels of these allergens are able to maintain or worsen airway sensitization. In fact, Bollinger *et al.* (5) have shown 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.

Clothes of pet owners constitute the main factor for the distribution of the most common pet allergens, such as Fel d 1 and Can f 1, but also less common ones (e.g. rabbits) in animal-free environments (6, 10). Human hair constitute a new unexpected source of cat allergen contamination (11).

The only feasible approach to reduce the transport and the dispersal of cat allergens is adequate cleaning of contaminated clothes worn by cat owners outdoors. This study was performed to assess under rigorous experimental conditions the efficiency of vacuum cleaning in removing the allergen. Because it is not possible to ensure identical allergen exposure to clothes (difficulty in obtaining identical clothing and providing reliable identical exposure in situ), we approached the problem by using samples of naïve cotton tissue. Among various methods available to capture allergens from a piece of material (e.g. use of extraction fluids), we choose high-volume sampling because of its ease of use. In addition, this procedure has been used in earlier studies (6-8).

The results of our study suggest that vacuum-cleaning by using a commercially available vacuum-cleaner for 15 minutes is not able to remove significant amounts of cat allergen from cotton fabrics exposed to cats. This procedure is less effective than other cleaning procedures that we have previously carried out by using identical methods such as same exposure, transport, division and sampling procedures (only with different fabrics), as in the current investigation. Washing in water without the use of detergents completely abolished Fel d 1 allergen from cotton fabrics (7). On the contrary, routine commercial dry-cleaning is effective for removal of large (and statistically significant amounts) of cat allergen from wool squares exposed to cats. Nevertheless, residual amounts of allergen have been found on the tested material.

Finally, Tovey *et al.* (12) demonstrated the effectiveness of laundry washing using detergent solutions at 25° for at least 5 minutes in extracting most mite and cat allergen from dust of bedding.

In conclusion, an efficient cleaning of clothes of cat owners should be included in the preventive strategies for controlling the indirect exposure to cat allergens.

In comparison to other cleaning procedures, the common vacuum cleaning is not effective in removing cat allergens from passively contaminated cotton fabrics and, consequently, should not be used as a single preventive measure.

**Acknowledgments:** We wish to thank all participating cat owners for their kind co-operation. Funding and competing interests: None to declare.

## References

1. Wahn U., Lau S., Bergmann R. *et al.* - Indoor allergen exposure is a risk factor for sensitization during the first 3 years of life. *J Allergy Clin Immunol* 1997;99:763-69.
2. Linneberg A., Nielsen N.-H., Madsen F. *et al.* - Pets in the home and development of pet allergy in adulthood. The Copenhagen Allergy Study. *Allergy* 2003;58:21-26.
3. Enberg R.-N., Shamie S.-M., Mc Cullough J., Ownby D.-R. - Ubiquitous presence of cat allergen in cat-free buildings : probable dispersal from human clothing. *Ann Allergy* 1993;70:471-74.
4. Custovic A., Taggart S., Woodcock A. - House dust mite and cat allergen in different indoor environments. *Clin Exp Allergy* 1994;24:1164-68.
5. Bollinger M.-E., Eggleston P.-A., Flanagan E., Wood R.-A. - Cat antigen in homes with and without cats may induce allergic symptoms. *J Allergy Clin Immunol* 1996;97:907-14.
6. D'Amato G., Liccardi G., Russo M., Barber D., D'Amato M., Carriera J. - Clothing is a carrier of cat allergens. *J Allergy Clin Immunol* 1997;99:577-78.
7. Liccardi G., Russo M., Barber D., Carreira J., D'Amato M., D'Amato G. - Washing the clothes of cat owners is a simple method to prevent cat allergen dispersal. *J Allergy Clin Immunol* 1998;102:143-44.
8. Liccardi G., Russo M., Barber D. *et al.* - Efficacy of dry-cleaning in removing Fel d 1 allergen from wool fabric exposed to cats. *Ann Allergy Asthma Immunol* 2002;88:301-05.
9. Luczynska C.-M., Li Y., Chapman M.-D., Platts-Mills T.-A. - Airborne concentrations and particle size distribution of allergen derived from domestic cats (*Felis domesticus*). Measurements using cascade impactor, liquid impinger, and a two-site monoclonal antibody assay for Fel d 1. *Am rev respir Dis* 1990;141:361-67.
10. Liccardi G., D'Amato G., Caninica G.-W., Dente B., Passalacqua G. - Severe respiratory allergy induced by indirect exposure to rabbit dander. A case report. *Allergy* 2004;59:1237-38.
11. Liccardi G., Barber D., Russo M., D'Amato M., D'Amato G. Human hair: an unexpected source of cat allergen exposure. *Int Arch Allergy Immunol* 2005;137:141-44.
12. Tovey E.-R., Taylor D.-J., Mitakakis T.-Z., De Lucca S. - Effectiveness of laundry washing agents and conditions in the removal of cat and dust mite allergen from bedding dust. *J Allergy Clin Immunol* 2001;108:369-74.