Using Airshowers to Decrease Laboratory Animal Allergy

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Summary

Airshowers may be an effective way of removing allergens from the clothing and uncovered skin and hair of personnel, but the optimal shower time and air speed for this use have not been elucidated. The aim of this study was to test the effect of differences in clothing type, shower time, and air speed on the efficiency of allergen removal by airshowering. In general we found that as air flow rate or shower time increased, so did allergen reduction. When tested in a real-life situation, the airshower removed more than 98% of the allergens from the shoulder area of personnel and more than 87% of the allergens from the thigh area. In addition, the allergens remaining after airshowering were not spread when protective clothing was removed. Factors such as clothing type, air speeds, and showering time need to be considered when establishing standard operational procedures for the use of airshowers.

Introduction

The use of wet showers when entering an animal facility, especially barrier units, has been common practice for many years. The shower reinforces required changes of attire and removes skin microorganisms and allergens, thereby preventing infections inside the animal facility and the spread of allergens outside. Laboratory animal allergy is indeed one of the most common occupational health hazards in laboratory animal work. In recent years, increased focus on laboratory animal allergy has led to the development of various types of allergen-reducing equipment, such as allergy cabinets, laminar-air–flow cabins and curtains. To ensure effectiveness, such equipment must undergo validation (*Gordon et al., 1997*; *Reeb-Whitaker et al., 1999*; *Gordon et al., 2001*; *Schweitzer et al., 2003*; *Krohn and Hansen, 2004*; *Krohn et al., 2006*). Airshowers are used commonly in the electronics industry to prevent spread of interfering particles from personnel to products and in the pharmaceutical industry to prevent spread of pharmaceutically active chemical dust (for example, from production of hormones) beyond the workplace. In these applications, airshowers are effective, although the type of clothing is an important factor in affecting the efficacy (*Whyte, 2001*). Airshowers might similarly be used to reduce the spread of allergens from animal facilities and as airlocks to control facility access. However, the optimal shower time and air speeds for removing allergens from the clothing and uncovered areas (for example, hair and face) are currently unknown. The aim of this study was to determine the effects of shower time, air speed, and fabric on the efficiency with which airshowering removes rodent allergens from clothing.

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Materials and Methods

Preparation of samples

Allergen powder was collected by sieving dirty aspen bedding (Tapvei, Finland), used for 4 d by male mice, by using a steel sieve with wire mesh (1.5 × 1.5 mm) to remove large material. The final

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powder was made up of small pieces of dirty bedding with a size of a few millimeters or less. The collected powder was mixed extensively to ensure that allergens were distributed equally throughout the mixture. The allergen content (μg/g powder) was estimated by enzyme-linked immunosorbent assay after diluting 0.5 g powdered bedding in 5.0 ml phosphate-buffered saline.

To prepare samples of clothing for testing, 0.5 g of the dry allergen powder was distributed over a 5 × 5 cm^2 square drawn on the material to be tested; excess powder was brushed away. Treated squares were excised by using forceps and scissors that had been sterilized in 70% alcohol. Inside the airshower, pieces of cloth were fixed in a 30 × 40 cm^2 array to a holder placed 160 cm above the floor, or complete sets of clothing were placed on a mannequin, which was fixed to a rotating platform on the floor. During airshowering, the holder or mannequin was turned 6 times per min to simulate a person turning during showering, as recommended by the manufacturer. As controls, contaminated areas were sampled prior to starting the airshower. For actual functional testing, clothing was worn by a person.

The airshower

A standard airshower (Scanbur A/S, Karlslunde, Denmark) was used. It delivers 3750 m^3/h air through 30 nozzles at a maximal rate of 35 m/s. The air speed through the nozzles was measured by placing the monitor (model 400, Testo, Lenzkirch, Germany) directly in front of a nozzle. The following equation was used to calculate the power or strength of the airstream coming through the nozzles:

$$P = {}^1/_2 \times \rho \times v^3 \times \pi \times r^2,$$

in which P is the power of the airsteam (in W, Watts), ρ is the density of dry air (1225 kg/m^3), v is the air speed (m/s), and r is the radius of the nozzle.

Testing various types of clothing and air-delivery parameters

To determine the effect of the clothing material, a total of 20 pieces each of a cotton labcoat and polyester suit (Countdown Clean Systems, Derby, England) were prepared, and after treating with test powder, 10 samples of each type were showered for 60 s at an air speed of 30 m/s with nozzle sizes of 38 mm. In addition, pieces of jumpsuit (Tyvek Pro-Tech Classic, DuPont™, US) were prepared and treated for 60 s by using 38-mm nozzles at air speeds of 25 m/s (12 W), 30 m/s (20 W), 35 m/s (32 W) and 20-mm nozzles at 30 m/s (5 W). For each air-delivery set-up, 10 samples were airshowered, and 5 were used as untreated controls.

Testing different airshowering times

For this test, the airshower was used at the standard set-up of 38-mm nozzles and 30 m/s. On each shoulder of 3 jumpsuits (Tyvek Pro-Tech Classic, DuPont™, US) 6 squares were drawn. Two of each were used as untreated controls, whereas the other 10 were allergen-spiked and the suit subjected to one of 3 showering times (15, 30, and 60 s).

Live functional test

The airshower was used in the standard configuration of 38-mm nozzles and 30 m/s. A volunteer donned a new jumpsuit (Tyvek Pro-Tech Classic) that had 3 areas (each 5 × 5 cm) marked on the shoulders and 3 areas (each 5 × 5 cm) marked on the thighs. Allergen powder was added to the marked spots. The wearer then jumped in place 10 times. The amount of allergen applied was estimated by sampling one of the marked spots before entrance into the airshower. Samples were collected after airshowering and again after the garments had been removed. To remove the suit, the wearer unzipped it, pulled it off his shoulders and then pulled out his arms. The wearer then gently pulled the suit down his body and removed his legs from the suit.

Analysis for allergens

To elute allergens from the sampled squares, a published method (*Renstrom, 1997*) was modified. Briefly each square was minced, placed in a 10-ml tube containing 5.0 ml phosphate-buffered saline with 0.5% Tween 20, and incubated at room tem-

perature for at least 2 h. A 1.0-ml aliquot of the eluate was spiked with 0.1 g of heat-fractioned bovine serum albumin (Sigma-Aldrich A7030) and stored at –20 °C until analysis. After thawing, the sample was analyzed (Mus m1 ELISA Kit, Indoor Biotechnologies, Manchester, UK) according to the manufacturer's instructions, except that visualization was done by orthophenylenediamine (S2045, DakoCytomation, Glostrup, Denmark) and samples were read at 492 nm and 630 nm (as reference). All samples were tested twice at both 1:10 and 1:100 dilutions, as previously described (*Krohn and Hansen, 2004*).

Data analysis

Data were tested for normal distribution (release 14.1, Minitab, State College, PA), after which correlations between allergen content (ng per cm^2) and power of air delivery (W) or showering time (s) were determined. All other comparisons were evaluated by the Mann–Whitney test (Minitab release 14.1) with significant level at $p<0.05$.

Results

The concentration of Mus m1 allergen in the powdered bedding was 23.9 µg/g. Compared with synthetic clothing, cotton bound more allergens both before and after showering (Figure 1). In addition, increasing the power with which the air was delivered (Figure 2) and increased showering time

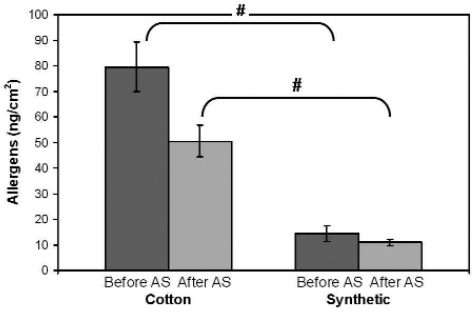

Figure 1. Amount of allergen (ng/cm^2; mean ± standard error, n = 10) on cotton and synthetic fabric before and after airshowering (AS). #, $P < 0.001$.

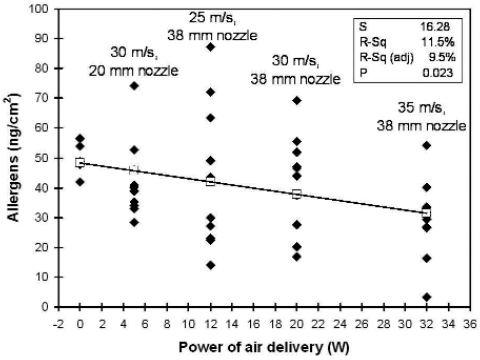

Figure 2. Correlation between power of air delivery and amount of allergen remaining on fabric (Tyvek Pro-Tech) after airshowering (AS) (n = 10). S (Sum of Squares), R^2 (The coefficient of determination), R^2 adj (Account for number of predictions)**.**

(Figure 3) also correlated significantly ($p=0.023$ for power, $p=0.004$ for showering time) to reducing the level of allergens on the clothing samples. In the functional test, in which a volunteer wore an allergen-spiked synthetic jumpsuit, airshowering decreased allergen levels (Figure 4). At the level of the thigh, removal of the jumpsuit further reduced the allergen level, regardless of whether airshower-

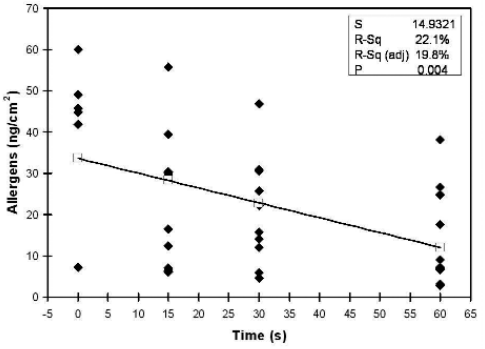

Figure 3. Correlation between shower time and amount of allergen remaining on fabric (Tyvek Pro-Tech) after airshowering (n = 10). S (Sum of Squares), R^2 (The coefficient of determination), R^2 adj (Account for number of predictions).

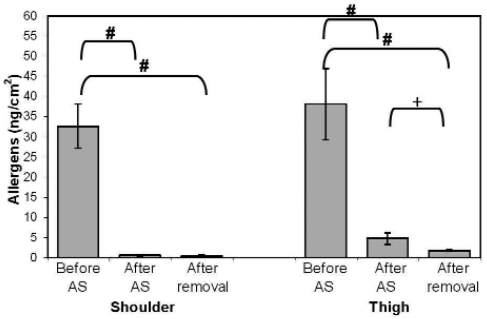

Figure 4. Amount of allergen (mean ± standard error, n = 10) on a synthetic jumpsuit (Tyvek Pro-Tech) before and after airshowering (before its removal) and after its removal. +, $P < 0.05$; #, $P < 0.001$.

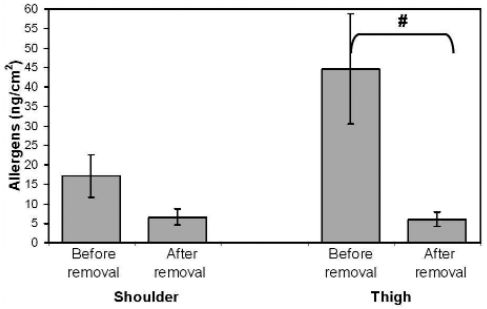

Figure 5. Amount of allergen (mean ± standard error, n = 10) on a synthetic jumpsuit (Tyvek Pro-Tech) before and after its removal in the absence of airshowering. #, $P < 0.001$.

ing had occurred (Figure 4, 5). The efficiency of allergen removal by the airshower at shoulder level was calculated as 98.4% after showering and 99.8% after both showering and jumpsuit removal. In comparison, the efficiency at thigh level was 87.4% after airshowering but 91.4% after both airshowering and jumpsuit removal.

Discussion

Careful consideration seems important when preparing standard operational procedures for the use of airshowers as allergy protectors in animal facilities. The differences between cotton and synthetic clothing reveal that, in regard to protection against exposure to laboratory animal allergens, a traditional cotton labcoat is a poor choice, as allergens stick to it very easily. In comparison, synthetic clothing binds approximately 80% less allergens, suggesting its suitability for minimizing the transfer of allergens from animal facilities. For optimal efficiency of the airshower, a complete suit either of tightly woven polyester or a nonwoven material, such as Tyvek, is preferable (*Whyte, 2001*). Studies on cat and mite allergens similarly show that the amount of allergens that binds to clothing varies greatly depending on the type of fabric (*Tovey et al., 1995*; *D'Amato et al., 1997*; *De Lucca et al., 2000*). Further, the higher the power with which the air is delivered, the greater the allergen reduction; the power of the air stream can be increased by increasing air speed or nozzle diameter. Increasing the power of the air delivered, however, has its limitations, because the associated expense and noise also increase. With the technology currently available, 20 to 25 W seems to be the optimal air-delivery level. Further, personnel may find airshowering at air speeds of approximately 35 m/s (equivalent to 78 miles/h—in the range of wind speed for a Category 1 hurricane) to be unpleasant. Our studies show that increasing the power of air delivery to greater than 25 W is unnecessary, considering that the additional allergen reduction gained by such an increase is slight. Finally, airshower efficiency increased with the duration of the shower: a shower time of 60 s was significantly more effective that one of 30 s.

Although initial tests showed that clothing fabric, air speed, and showering time affected airshower efficacy, results of functional testing involving a person wearing a contaminated jumpsuit revealed that, in practice, airshowering successfully removed more than 98% of the allergens at shoulder level and more than 87% of those at thigh level. These differences in efficacy likely reflect differences in the way the jumpsuit fit at these 2 sites, as there is more folding in the thigh area. In addition, the loss

of allergens from the thigh area during jumpsuit removal may be influenced by the way the suit is taken off – removal of the jumpsuit at thigh level requires more manipulation of the fabric than does that at the shoulders. However overall, the bulk of allergens remaining after showering remains on the jumpsuit after its removal. These findings indicate that airshowering is effective for reducing allergen levels, and those allergens that remain after airshowering are likely to be tightly bound and unlikely to be released and spread during the removal of the garment.

We conclude that airshowering can be an effective way of removing allergens from protective clothing, and those allergens remaining on protective clothing after airshowering are not spread when the clothing is removed. Factors such as clothing fabric (e.g. cotton versus synthetic, as above), air speed, and showering time should be considered when setting up standard operational procedures for using airshowers to minimize the spread of laboratory animal allergens.

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