

Reduction of house dust mites and mite allergens: effects of spraying carpets and blankets with Allersearch DMS, an acaricide combined with an allergen reducing agent

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Summary

Two studies were made to determine the acaricidal and allergen reducing properties of Allersearch DMS solution. In Study 1, samples of dust and carpet containing living mites were treated, *in vitro*, with DMS solution. No living mites were found in dust after 3–4 min or in carpets 50 min after treatment. In Study 2, mite counts and allergen estimation made on bed blankets before and after spraying with DMS solution showed a marked reduction in mite numbers (95%) and allergenicity (100-fold). Mite numbers and allergenicity stayed at this low level for 6 weeks. At 16 weeks both mites and allergens showed a slight increase, but were still significantly ($P < 0.001$) below pre-treatment levels.

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Introduction

It is now well established that the house dust mite *Dermatophagoides* spp. is an important source of allergens which may trigger attacks of asthma, hay fever or eczema. In the 21 years since the house dust mite's implication in these diseases [1] many attempts have been made to control mites in houses [2–5]. Methods including washing fabrics, vacuum cleaning and dust suppression, encasing mattresses, pillows and cushions in dust impermeable covers, regulation of humidity and, latterly, killing of mites by chemical or physical methods have been employed.

Studies of subjects living in natural or contrived mite free places have shown improvement of asthma and bronchial hyperresponsiveness [6–8]. However, studies of the efficacy of hygienic measures, vacuum cleaning and mattress encasement, have had equivocal results [2–5]. Likewise, cold water washing or shampooing has little or no effect on mites [6]. Control of humidity, while theoretically a sound approach, is extremely difficult to achieve and is only possible using expensive air-conditioning equipment.

Recent studies have shown that liquid nitrogen can kill mites in bedding and textiles [9] and there are several chemical acaricides which effectively reduce mite numbers in houses [10,11]. However, when using acaricides the possibility of toxic or other effects must be considered. Acaricides only kill the mites, leaving the allergenic mite bodies and faecal pellets to be removed by the previously discussed methods of control: vacuuming, washing, etc. It is obvious that an agent that could both kill mites and render them non-allergenic would have advantages over agents which simply kill mites.

A chemically non-toxic solution called Allersearch DMS (Allersearch, Revesby, NSW, Australia) containing such an agent has been formulated. The solution comprises a mixture of polyphenols and a benzyl derivative in an aqueous alcoholic base. The following study shows firstly its effectiveness as an acaricide when applied to mite containing dust and carpets, and, secondly, its acaricidal and anti-allergenic properties in bed blankets.

Materials and methods

Mites in dust

Ten 20 mg samples of liver powder culture medium containing living *D. pteronyssinus* mites were placed on a

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welled glass slide and carefully sprayed at a rate of 0.25 g/100 cm² with DMS solution. Viewed microscopically, one light spray was sufficient to wet the surface of the mites without causing excessive accumulation of solution on the slide. After spraying, the mites were observed continuously under a binocular microscope, objective $\times 40$, and the time recorded when all movement had ceased.

Mites in carpet samples

Living active mites in dessicated liver powder medium were placed on the basement layer within the pile of 10-cm squares of two carpet types: Berber, pile length 1.0 cm, and synthetic, pile length 1.2 cm. DMS solution was sprayed onto the surfaces of the carpet squares (140 ml/m²) after which they were examined continuously for 1 min, then at 1-min intervals for 2 hr. Cessation of movement was considered to indicate mite death and the time at this point was noted. Non-living mites were further observed for 16 hr after treatment to detect possible recovery.

Mites in blankets

Ten blankets from the homes of laboratory staff and patients living in Sydney, NSW, were taken from the beds, analysed for mite and allergen content then returned to the beds. After 14 days the mite and allergen content of the blankets was assessed again and the mean of the values found was used as the baseline for the study. At this stage the blankets were sprayed on both sides with DMS solution at 23 ml/m², delivered from a hand pumped pressure sprayer, allowed to dry and returned to the beds. Further mite counts and allergen assessments were made on dust samples from the blankets at 2, 4, 6 and 16 weeks after treatment. At 16 weeks two blankets were not available for study and a further two blankets yielded insufficient dust for preparation of extracts. The blanket study was made over the period from spring (September) to mid-summer (January).

Dust sampling

Dust samples were collected from a measured area of blanket, either 1 or 2 m², at the rate of 0.5 m²/min with a hand held vacuum cleaner (Hoover Dustette). To minimize possible fluctuation of mite numbers of allergens due to the disruptive effects of the study, dust was collected from a different area of the blanket at each sampling time. A new paper bag was placed in the vacuum cleaner for each sample collection and the weight of dust obtained was recorded. Mite counts per gram of dust were made as described previously [12] and the number of mites per square metre of blanket calculated. *Dermatophagoides* species were not differentiated.

Assessment of allergen in blanket dust

Allergen content of the blanket dusts was assessed by a skin-prick test bioassay [13], using aqueous extracts of the dusts. Dust samples of 6 mg were thoroughly mixed with 100 μ l distilled water in a conical microfuge tube. The samples were allowed to stand for 5 min, then spun at high speed setting for 5 min in a HERMLE microcentrifuge (B. Hermle, GmbH & Co., Gosheim, FRG). A 50- μ l aliquot of the supernatant was carefully taken off and added to 50 μ l glycerine, mixed and filtered through a millipore 0.3 μ m filter. In order to increase the sensitivity of the assay dilutions of 10⁻¹ and 10⁻² were prepared with 50% glycerine/water. Glycerine was used to prevent evaporation during the skin tests. The assay, using these dilutions and the undiluted sample, was made by skin-prick testing on the scapular aspect of the back of a consenting subject known to be skin test positive only to *Dermatophagoides*. Skin tests were performed at the same time of day on each occasion. Skin weal size was estimated 15 min after testing as the mean of the longest axis and its perpendicular. The median skin weal size produced by a blanket extract was regarded as an index of its mite allergen content.

Safety of DMS solution

DMS solution is considered non-toxic [LD₅₀ (rats) = 31 g/kg], however the following tests for adverse respiratory reactions in asthmatic and normal subjects were made. Informed consent was obtained from each subject.

Inhalation of aerosolized DMS. Five asthmatic and three normal subjects were tested by inhalation of aerosolized DMS solution from a DeVilbiss No 40 nebulizer (De Vilbiss Somerset, U.S.A.). Subjects took 1, 3, 5 and 10 inhalations, each inhalation delivering approximately 0.003 ml to the mouth of the subject. The 1-sec forced expiratory volume (FEV₁) was measured before and 1 min after each dose.

Environmental exposure. Nine asthmatic subjects recorded their expiratory flow rates using an Airflometer (Glaxo Australia) in the morning and evening before and after their bronchodilator medication, for 1 week before, during, and 1 week after their houses were treated with DMS. The subjects were present in the rooms during spraying.

Data analysis

Ninety-five percent confidence intervals are given for mite survival times and Student's *t*-tests were used to determine the significance of differences in the mite counts.

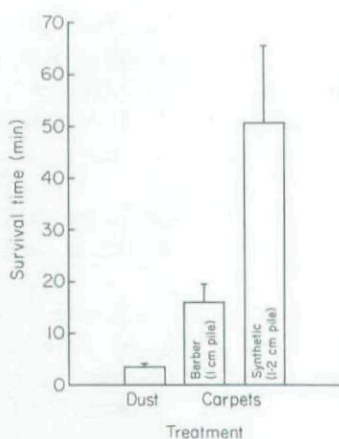


Fig. 1. Mean survival times of mites in dust and carpet samples after treatment with DMS.

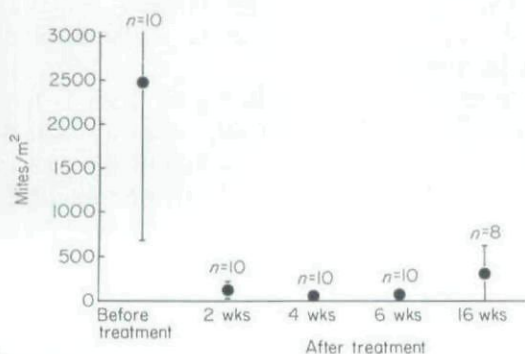


Fig. 2. Mean mite counts in blankets before and after treatment with DMS.

As the skin test weal sizes are not normally distributed median values are reported and the Mann-Whitney two-sample *U*-test was used to determine the significances of differences between weal sizes.

Results

Mites in dust

The mean survival time of *D. pteronyssinus* mites in culture medium dust after treatment with DMS was 3.56 min (95% confidence interval (CI): ± 0.38).

Mites in carpet

Mean survival time of mites in carpet samples after treatment with DMS solution was 16 min (95% CI: ± 3.4) for the Berber carpet pile length 1 cm, and 50.5 min (95% CI: ± 14.5) for the synthetic carpet, pile length 1.2 cm (Fig. 1). No mite recovery was observed.

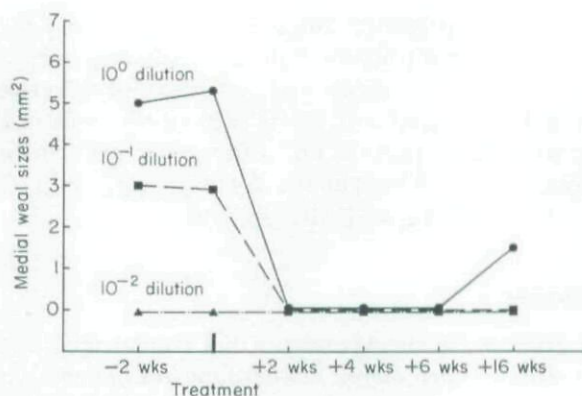


Fig. 3. Median skin weal sizes produced by skin testing with undiluted, 10^{-1} and 10^{-2} dilution of blanket dust extract before and after treating blankets with DMS. $n=10$ for all readings except at 16 weeks where $n=6$.

Mites in blankets

The mean mite counts in the blankets was significantly reduced from 2473/m² pre-treatment to 125/m² at 2 weeks, 58 m² at 4 weeks, 77/m² at 6 weeks and 309/m² at 16 weeks ($P < 0.001$ in all cases) (Fig. 2).

Allergens in blankets

Median skin weal sizes produced by serial 10-fold dilutions of extracts of dust from the blankets before and after spraying with DMS are shown in Fig. 5. At the highest concentration of blanket dust extract (i.e. undiluted) the median weal size was 5.0 mm and 5.3 mm 2 weeks before and immediately before treatment of the blankets, and decreased to zero at 2, 4 and 6 weeks after treatment. At 16 weeks after treatment the median weal size had increased to 1.5 mm. Similarly, at 10^{-1} dilution median weal size decreased from 3.0 mm 2 weeks before and 2.9 mm immediately before treatment to zero afterwards and stayed at this level for 16 weeks. The 10^{-2} dilution gave no median values greater than zero before or after treatment.

Safety of the solution

Inhalation of aerosolized Allersearch DMS. Neither asthmatic nor normal subjects showed any change in FEV₁ after inhaling aerosolized DMS. Five asthmatic subjects had a mean FEV₁ value of 85% of predicted (± 17.0 s.d.) before inhalation of aerosolized DMS, and a mean FEV₁ of 84% (± 17.4) after inhalation. Three normal subjects had a mean FEV₁ of 113.7% (± 21.4) before and 116.0% (± 24.2) after inhalation.

Environmental exposure. No changes in expiratory flow rates or symptoms occurred in nine asthmatic subjects after the bedding, carpets and soft furnishings in their houses were sprayed with DMS. Mean airflometer readings were $117.4 (\pm 59.5 \text{ s.d.})$ during the week before spraying, $121.7 (\pm 59.7)$ on the day of spraying and $119.3 (\pm 61.9)$ during the week after spraying.

Discussion

There is now convincing evidence that avoidance of house dust mite or other allergens is desirable for patients with asthma or other diseases where allergenic mechanisms are involved. The present study has demonstrated that *Dermatophagoides* mites and their allergens can be reduced to extremely low levels by spraying with the acaricide and anti-allergen agent Allersearch DMS. This may be an important adjunct to other methods of mite control in houses or other places.

The results show that both the numbers of mites and the allergenicity of the blanket dust are still at low levels 16 weeks after treatment. This is surprising because between testing times the blankets were returned to untreated mite infested beds. Thus it appears that the mites were reluctant to recolonize the treated blankets. Figure 2 suggests that the best time for respraying would be between 4 and 6 weeks after the initial treatment, i.e. before mite numbers begin to increase.

Although DMS proved lethal to mites, its effectiveness against mite eggs was not determined in this study. The life cycle of mites from egg to egg is about 30 days [14], so if the eggs are still viable after the initial treatment then a repeat spraying after 1 month would be necessary to eliminate mites. However, after 30 days no increase in mite numbers was seen. Although it is possible that the eggs were killed, a more definitive study of the effect of the solution on the viability of mite eggs is now in progress.

In carpets, the length of the pile and type of fibre influences the rate of penetration of the solution and therefore the time taken to kill mites (Fig. 1). It is likely that some carpets would rarely be wetted to the basement layer by spraying the surface. However, as the alcoholic component of the solution evaporates, droplets of pure liquid acaricide are deposited in the carpet fibres. These droplets in turn evaporate and their fumes penetrate sufficiently to kill mites (unpublished observation). Thus, it is not always essential for the spray to make direct contact with the mites, and *in vitro* results here show that mites in deep pile carpet die in less than 1 hr after treatment. This is supported by four preliminary studies in houses which showed a mean reduction in mite numbers of 83% after spraying.

Mite densities can be expressed either as numbers per

gram or per area of blanket. Here they have been expressed as mites per square metre of blanket surface because the dust samples were collected by vacuuming and blankets which contain much inert dust, e.g. talcum powder, will show a lower 'mites per gram' content than blankets that have the same number of mites but little dust. However, when we looked at the data expressed as mites per gram, densities showed a similar fall after treatment.

Several methods of assaying *Dermatophagoides* allergen are now available [15,16]. At the beginning of this study these were not available in our laboratory and the RAST inhibition method was discarded because of its variability in antibody specificity and low sensitivity [17]. On the other hand, the skin test bioassay has the advantage of demonstrating direct biological activity of the multiple allergens present and was chosen for this reason. Some non-specific variation in skin weal sizes is likely in any skin tests. However our study was suitably controlled and the results from ten different blankets show good agreement.

The mode of action of Allersearch DMS is not yet known but it is likely that the mites are killed by interference with neural activity. Its anti-allergenic properties are probably due either to its precipitating and denaturing of the allergenic proteins and glycoproteins or to alterations in the surface antigenic determinants so that they fail to elicit an IgE mediated response. More studies are needed to clarify this. Further work must also be done to determine if the solution is effective against other allergens such as pollens as suggested by a previous study [18].

It is suggested that DMS be used in conjunction with other measures for mite reduction. It can be sprayed on mattresses, pillows, blankets, carpets, upholstered furniture, soft toys or wherever mites accumulate, particularly in bedrooms. However, ideally the whole house should be treated, at least initially, if a mite avoidance program is to be successful [19,20].

This study has shown that DMS solution kills *Dermatophagoides* mites in dust and carpets, reduces mite numbers in bed blankets and reduces mite allergens in dust from bed blankets. These results suggest that spraying with DMS is an effective way to control mites and, more importantly, to reduce mite allergens in houses. Furthermore, the study has shown that the solution causes no adverse respiratory effects in asthmatic or normal people. In view of recent reports of improvement in patients bronchial hyperresponsiveness and allergic symptoms when mite allergens are avoided, spraying with DMS should be an important consideration in the management of patients with asthma, allergic rhinitis and eczema.

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