

## Original article

# Disodium octaborate tetrahydrate (DOT) application and vacuum cleaning, a combined strategy to control house dust mites

**Background:** The effectiveness of acaricides in homes is controversial.

**Objective:** To determine whether disodium octaborate tetrahydrate (DOT) combined with vacuuming lowers dust mite numbers and their allergens in carpets and sofas.

**Methods:** A 6-month study was carried out with 93 homes, which were randomized into three groups: (i) active, received DOT; (ii) placebo, received water; and (iii) control, received no application. Active and placebo homes were vacuumed weekly. Dust was collected from carpets and sofas at the start of the study and every 2 months thereafter and quantified for live, total mites, and mite allergen levels.

**Results:** At 2 months, live mite numbers in active carpets were  $3 \pm 1$ , in placebo carpets  $129 \pm 48$ , and in control carpets  $177 \pm 39$  mites/g. The corresponding numbers in sofas were  $3 \pm 2$ ,  $81 \pm 31$ , and  $134 \pm 45$  mites/g, respectively ( $P < 0.001$  active vs placebo and vs. control). Live mites in carpets and sofas remained lower in the active group at 6 months ( $P < 0.001$ ). Total mites in active carpets decreased from  $555 \pm 69$  at baseline to  $223 \pm 32$  mites/g at 6 months ( $P < 0.001$ ) and mite allergen levels from  $1.36 \pm 0.13$  to  $0.85 \pm 0.16$   $\mu\text{g/g}$  ( $P < 0.001$ ). Total mites in active sofas remained unchanged, but mite allergen levels decreased from  $1.48 \pm 0.25$  at baseline to  $0.7 \pm 0.15$   $\mu\text{g/g}$  at month 6 ( $P < 0.05$ ).

**Conclusion:** DOT kills mites in carpets and sofas, and, combined with vacuuming, effectively reduces total mites in carpets and mite allergen levels in carpets and sofas.

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Exposure and sensitization to dust mites are risk factors for developing allergic asthma and/or allergic rhinitis; epidemiologic studies indicate that a level of  $2\mu\text{g/g}$  of Group 1 mite allergens (Der p1+Der f1) and/or 100 mites/g of dust are risk factors for sensitization of genetically predisposed individuals (1). Ten micrograms per gram of Group 1 allergens per gram of dust and/or 500 mites/g of dust provoke asthma symptoms in dust mite sensitive individuals. However, lower levels, found in homes of asthmatic children, may also sensitize and provoke symptoms, suggesting that the threshold value for sensitization to mite allergens is lower in children with asthma (2, 3). The first line of treatment for allergic asthma and allergic rhinitis is allergen avoidance (4), which targets carpets, beds, and upholstered furniture, the most important mite reservoirs in the home. Unless carpets are removed, most allergen avoidance trials have not been successful. However, removal of carpeting is expensive and carpets are desired by many homeowners. Thus, current research focuses on identifying strategies, which normally include bedding encasing combined with

a procedure to kill mites and vacuuming with a high-filtration vacuum cleaner (5). Physical methods, heat, cold, and reduced humidity, have been used to kill mites; none of them have gained wide acceptance to treat carpets because of their unproved effectiveness, lack of safety, and their high cost (6–12).

Several acaricides have killed mites in laboratory experiments (13), but their effectiveness in the home is controversial (14) or, even if effective, they have not been commercialized for home use (15, 16). Acaricides must: (i) lack toxicity and odor; (ii) not damage household items; (iii) be easy to apply and penetrate deeply into carpet and upholstery; and (iv) be relatively long-lasting and not requiring such frequent reapplication that its use becomes too expensive and time consuming.

Paragerm® (Promedica, Levallois-Perret, France), an acaricide that contains 10 components, including various antiseptics, has been used in Europe for over 45 years, acts quickly, is easy to apply, is effective for at least 2 months, but has an unpleasant odor (17). Benzyl benzoate has also been widely used as an acaricide, but may not be effective

even when applied according to the directions provided by the manufacturer (18). Disodium octaborate tetrahydrate (DOT) (The Ecology Works, San Rafael, CA) is nontoxic to humans (19), and in laboratory experiments effectively reduced live *Dermatophagoides pteronyssinus* and *D. farinae* mite populations at 4–6 weeks after application. However, the effectiveness of this product to kill mites in homes has not been previously investigated.

The purpose of this study is to ascertain whether DOT, combined with weekly vacuuming with a high-filtration vacuum cleaner, effectively lowers house dust mite populations and their allergens in carpet and upholstered furniture in homes.

### Methods

#### Recruitment, screening, and randomization

This is a 6-month, double-blind study conducted in Central Florida, USA, between July 2000 and September 2001. The study protocol and informed consent form were approved by the University of South Florida Institutional Review Board.

Subjects and their homes were obtained via newspaper advertising. The entry criteria for admission to the study were as follows: no children under 5 years of age, no pregnant or lactating women, or asthmatics living in the home; single family dwelling with a wall-to-wall carpet older than 2 years in the family room and master bedroom; couch covered with fabric; no treatment for fleas or other insecticides used indoors for 6 months prior to or during the study; no dehumidifiers in the home; homeowners capable of giving written consent to participate and comply with the study requirements; agreement not to replace or have the carpets cleaned during the study and intention to live in the house for 12 months following the beginning of the study.

More than 3000 homeowners were responsive to the advertisements. Applicants were interviewed by telephone, and if they met inclusion criteria, the home was visited to collect an initial dust sample from the master bedroom carpet next to the bed. Homes with 100 or more live mites per gram of dust in the initial sample were included in the study. This screening phase took place from July 2000 to March 2001 and the intervention phase from April to September 2001.

Four hundred and eighty homes met the first seven entry criteria and 107 of these (22.3%) had more than 100 live mites per gram of dust and qualified for the study. Fifty-three of these 107 homes were assigned to an active group (even numbers from the list of homeowners) and 54 (odd numbers from the list) were equally divided between placebo and control groups following the same procedure (alternated numbers from the list). Some dropouts occurred before the start of the study, and 93 of these 107 homes participated in the study, 43 in the active group, 24 in the placebo group, and 27 in the control group (Fig. 1).

#### Interventions

Homes of the active and placebo groups received pillow, mattress, and box spring encasings (SofTek®, National Allergy Supply, Duluth, GA) for the master bed at the start of the study to prevent re-infestation of the master bedroom carpets from mites contained in the mattresses. In these two groups, in addition to the homeowners' routine vacuum cleaning, a cleaning service vacuumed the

carpet in the entire house and living room sofa once a week with a 12 A vacuum cleaner containing a HEPA filter and a double-wall bag (Hoover Model U5435900, Hoover Company, North Canton, OH). Homes of the active group received DOT application (The Ecology Works Dust Mite Control, San Rafael, CA) in the master bedroom and living room carpets and living room sofa at the start of the study. Homes of the placebo group received water application in the same three locations. DOT was not utilized to treat mattresses since a more simple solution, such as bedding encasing, is available to eliminate dust mites in mattresses.

DOT or placebo solutions were applied with a steam vacuum machine (Hoover Model F5866900, Hoover Company, North Canton, OH) using the appropriate attachments to treat carpet and upholstered furniture. The application rate was performed according to the instructions provided by the manufacturer, 0.8 l/m<sup>2</sup> of a 5% w/v DOT solution in tap water heated to 50°C or water alone depending on the group. Homes of the control group received no intervention, but the homeowners were instructed to continue their routine domestic vacuum cleaning. Homeowners of the active and placebo groups did not know which treatment type they received.

#### Dust collection

Dust samples were collected from the master bedroom carpet immediately next to the bed, living room carpet immediately next to the sofa, and sofa at the beginning of the study prior to the intervention and every 2 months for 6 months from all homes. The numbers of live and dead mites and the concentration of Group 1 (Der p1 + Der f1) allergens were determined for each dust sample.

Dust samples were collected with a 7 A canister vacuum cleaner (Hoover Model C 2094, Hoover Company, North Canton, OH) with its inlet modified to accept a collecting chamber containing a 180 tread-count cotton filter. Dust samples were collected from the same areas of 1 m<sup>2</sup> from the master bedroom and living room carpets and sofa by vacuuming for 2 min. Samples were collected on the filter and stored at 4°C until analysis. All mite counts were performed by R.C. and Group 1 allergen level determinations by L.M., who had no knowledge of the source of the samples. One of the investigators (R.D.) was in charge of the sample coding and data entry, and the study file was kept in a separate laboratory computer to maintain the blinding of the study. The code was only broken at the end of the study after all dust samples were analyzed.

#### Mite counts

Live and dead mites were counted in a 50 mg sample of fine dust. The counts were performed within 48 h of collection by the suspension method according to the protocol of Arlian et al. (20). In addition to moving mites, those that showed no movement, but were full-bodied, were counted as live (20). Mite counts were expressed as number of live and dead mites per gram of dust.

#### Dust extraction and allergen assays

Fifty milligrams of fine dust were extracted 1 : 20 w/v overnight by shaking at 4°C with 0.1 M ammonium bicarbonate, pH 9.6. The extracts were centrifuged at 3000 g for 5 min, the supernatant isolated and stored at –20°C until analysis. Der p1 and Der f1 allergen levels were measured in duplicated samples by ELISA using monoclonal antibodies for each allergen (Indoor Biotechnologies, Charlottesville, VA) according to protocols described by Luczynska et al. (21). Results were expressed as micrograms of Group 1 (Der p1 plus Der f1) per gram of dust.

Statistical analysis

The Wilcoxon test was utilized to compare the values obtained each sampling time within each group and the Mann-Whitney test to compare values among pairs of groups. The chi-square test was utilized to check the association between DOT application, vacuum cleaning, and reduction of mite number and mite allergen levels. A *P*-value < 0.05 was considered significant.

Results

Fifty-eight homes completed the study (Fig. 1). The dropouts were based on participants' decision (14 homes) or because of protocol violations (19 homes) and loss to follow-up (two homes). Because Central Florida is a subtropical area, it was difficult for many homeowners to withhold application of pesticides or insecticides during the 6-month study. However, none of the homeowners mentioned any side effect or complaints because of the active or placebo treatment.

Live and dead mite counts and Group 1 allergen levels between master bedroom and living room carpets in each

group did not significantly differ at any sampling time throughout the study. The results were analyzed for carpet (bedroom and living room) and for sofa samples. The dust recovered from some homes at the end of the study was insufficient to perform the dust analysis.

Mite numbers

Live, dead, and total mite numbers in carpet and sofa samples in the three groups did not significantly differ at the start of the study. Live mite numbers in carpet samples in the active, placebo, and control groups are  $3 \pm 1$ ,  $129 \pm 48$ , and  $177 \pm 39$  mites/g, respectively, 2 months after the intervention; the corresponding values for sofa samples are  $3 \pm 2$ ,  $81 \pm 31$ , and  $134 \pm 45$  mites/g, respectively (*P* < 0.001 active vs. placebo and control groups). Live mite numbers in carpet but not in sofa samples were also reduced in the placebo and control groups (*P* < 0.05 at months 4 and 6 in the placebo group). However, numbers in the active group compared to baseline levels and to the placebo and control groups

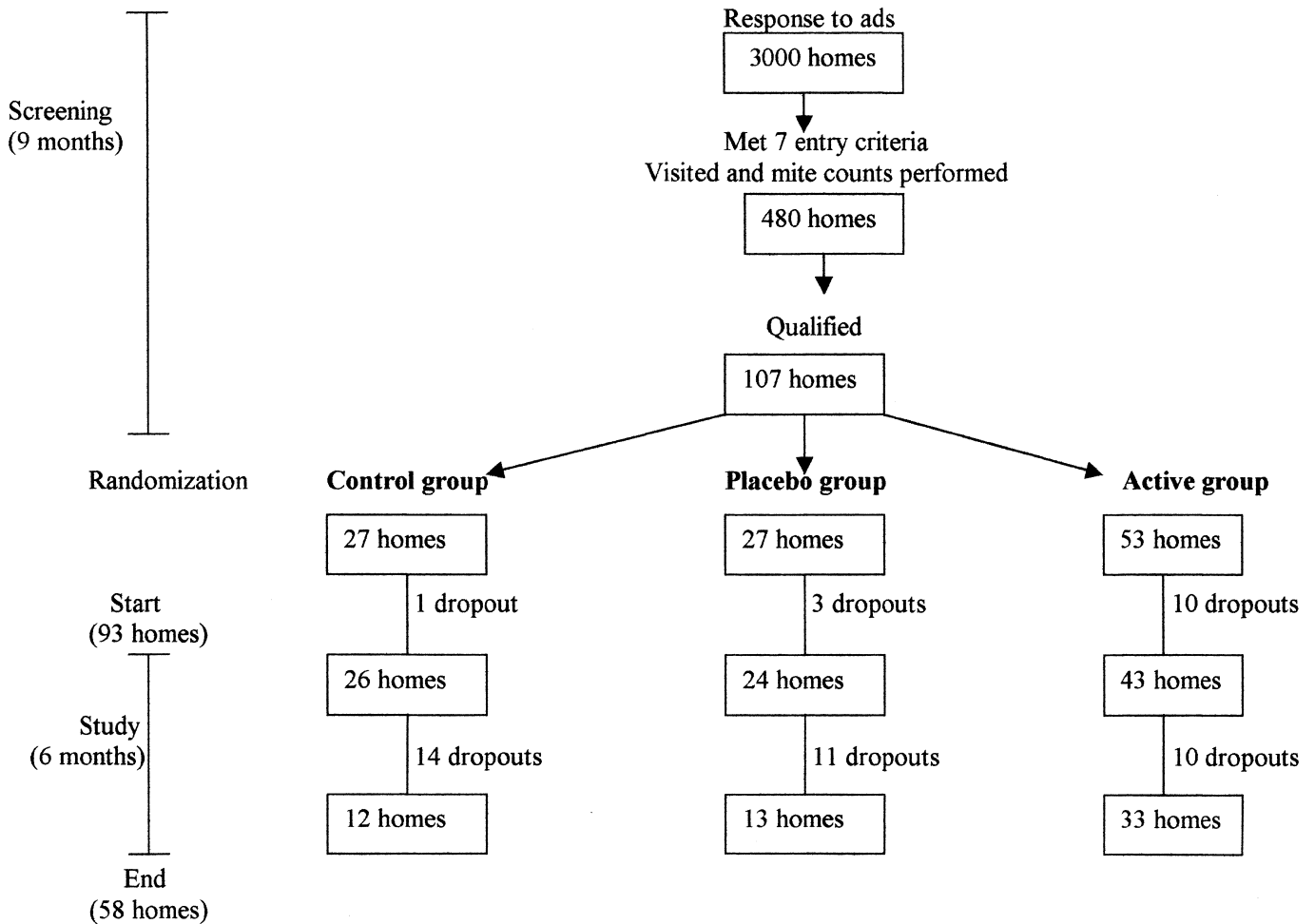


Figure 1. Protocol overview.

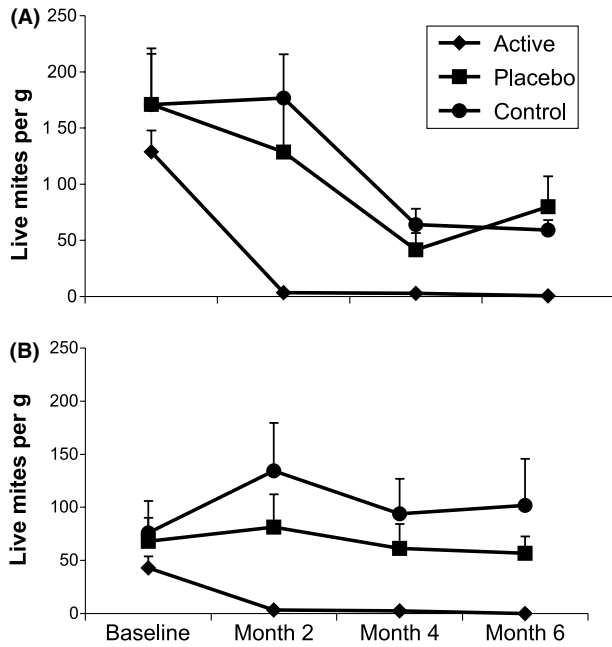


Figure 2. Live mite numbers in carpet (A) and in sofa (B) samples.

remain significantly lower at all sampling times ( $P < 0.001$ ) (Fig. 2A,B).

Sixty of 61 (98.3%) carpet and 30/30 (100%) sofa samples in the active group, 9/25 (36%) carpet and 3/13 (23%) sofa samples in the placebo group, and 5/24 (20.8%) carpet and 4/11 (36.3%) sofa samples in the control group contained no live mites at the end of the study ( $P < 0.001$ , active vs placebo and control groups).

There was a significant increase of total mite numbers at the second sampling time in carpet and fabric samples in the placebo and control groups ( $P < 0.05$ ) but not in the active group. There is a progressive significant reduction of total mite numbers in carpet samples in the active group throughout the study compared to baseline levels and placebo and control groups, decreasing from  $555 \pm 69$  mites/g before the intervention to  $223 \pm 32$  mites/g at the end of the study ( $P < 0.001$ ). This reduction is not observed in sofa samples. However, total mite numbers in sofa samples in the active group do not experience the increase observed in the placebo and control groups at month 2, and numbers are significantly lower at months 2 ( $P < 0.05$ ) and 4 ( $P < 0.001$ ) compared to the placebo and control groups (Fig. 3A,B).

Seventeen of 61 (27.8%) carpet and 16/30 (53.3%) sofa samples in the active group, 5/25 (20%) carpet and 2/13 (15.3%) sofa samples in the placebo group, and 0/24 (0%) carpet and 2/11 (18.1%) sofa samples in the control group contained fewer than 100 total mites (all dead in the active group with the exception of one carpet sample) at the end of the study ( $P < 0.05$  active vs control group for carpet and  $P < 0.05$  active vs placebo group for sofa samples).

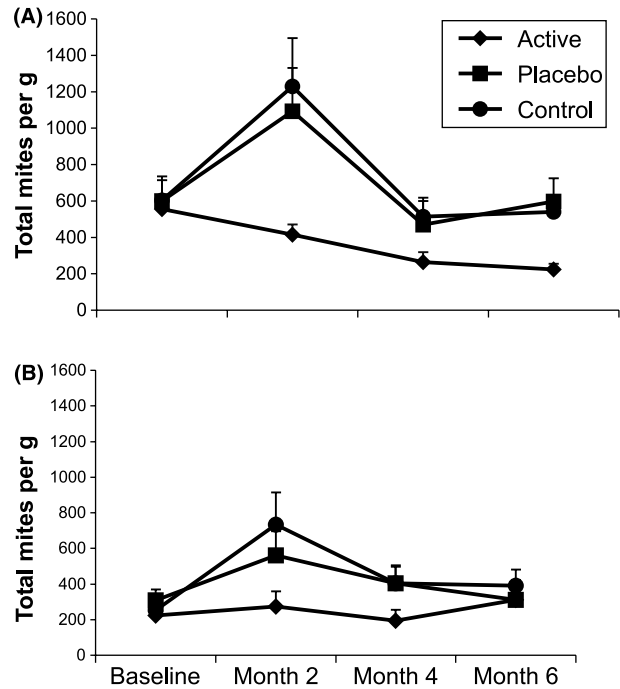


Figure 3. Total mite numbers in carpet (A) and in sofa (B) samples.

Fifty-six of 61 (91.8%) carpet and 27/30 (90%) sofa samples in the active group, 15/25 (60%) carpet and 9/13 (69.2%) sofa samples in the placebo group, and 13/24 (54.1%) carpet and 9/11 (81.8%) sofa samples in the control group contained fewer than 500 total mites (all dead in the active group with the exception of one carpet sample) at the end of the study ( $P < 0.05$  and  $P < 0.001$  active vs. placebo and control groups, respectively, for carpet, and  $P$  not significant for fabric samples).

Group 1 allergen levels

Group 1 allergen levels in carpet and sofa samples in the three groups did not significantly differ at the beginning of the study. Group 1 allergen levels in carpet and sofa samples in the placebo and control groups increased at month 4, probably as a result of the increase in total mite numbers observed at month 2 in these groups. Group 1 allergens are contained in mite fecal pellets, and an increase in mite numbers is normally accompanied by an increase in Group 1 allergen production, which peaks later in time. Group 1 allergen levels in carpet and sofa samples in the active group are below the values found in the placebo and control groups at most sampling times. There is a significant reduction of Group 1 allergen levels in the active group only at the end of the study, decreasing from  $1.36 \pm 0.13$  to  $0.85 \pm 0.16$   $\mu\text{g/g}$  ( $P < 0.001$ ) and from  $1.48 \pm 0.25$  to  $0.7 \pm 0.15$   $\mu\text{g/g}$  ( $P < 0.05$ ) in carpet and sofa samples, respectively (Fig. 4A,B).

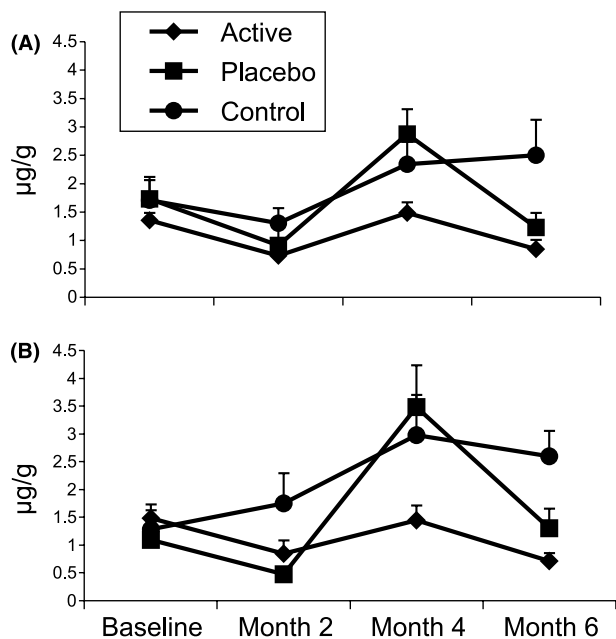


Figure 4. Group 1 allergen levels in carpet (A) and in sofa (B) samples.

## Discussion

Despite more than 30 years of research on control of house dust mites, no single method has been successful, resulting in a research focus on evaluating combined environmental strategies to control dust mite populations. These combined strategies include a procedure to kill dust mites. Several acaricides kill mites in laboratory experiments (13), but their effectiveness and usefulness in homes has not been adequately established.

This 6-month study applied DOT with a steam vacuum machine, which killed dust mites in carpets and sofas, and with a residual effect which remained active at the end of a 6-month period of observation. The combination of DOT application followed by weekly vacuuming with a high-filtration vacuum cleaner significantly reduced total mite numbers in carpets throughout the study, reducing mite counts in most samples to levels below the threshold values which induce sensitization and symptoms. No live mites were present in sofas at the end of the study, although this strategy was less effective in reducing total mites. Control of dust mites in upholstery remains a problem, since acaricides have failed to reduce mite allergen levels significantly in sofas (22–25), probably because of the difficulty of vacuuming upholstered furniture containing pillows.

Group 1 allergen levels in carpets and sofas were significantly lower than baseline levels at the end of the study in the active group only, and were below levels of the placebo and control groups; this indicates that the environmental control strategy tested also reduces mite allergen levels. Some man-made substances such as tannic acid, pesticides, and boric acid, interfere in the assays to

measure indoor allergens (26–28), but DOT does not interfere with the assays of Group 1 allergen levels (29), and DOT interference is not responsible for the lower levels of Group 1 allergens found in the active group.

Nearly complete control of live mites and a reduction of total mites and Group 1 allergen levels at the end of the 6-month period suggests that an additional 6 months of monitoring would be necessary to determine full term control. This study was limited to only one application of DOT using a steam vacuum machine to accurately determine the duration of effectiveness of DOT in mite control. Mite allergens are water soluble (30), and more frequent additional cleanings with hot water could be helpful in facilitating further reduction of allergens. However, additional cleaning could leave residual moisture and potentiate mite growth (7). A strategy of pre-treatment of new carpet and upholstery with DOT could also be useful, and deserves further investigation.

Although most studies on the efficacy of acaricides in homes do not perform live and dead mite counts, one uncontrolled study conducted in 1992 investigated the effectiveness of benzyl benzoate in killing mites in four chairs, four carpets, and four mattresses (31). Benzyl benzoate was applied at the start of the study, 4, and 26 months later. Results suggested a reduction of live mites even 3 years after the application.

Studies that included active and control groups failed to demonstrate a significant reduction of Group 1 allergen levels in intervention groups treated with benzyl benzoate compared to control groups (32, 33). Other studies performed with benzyl benzoate, alone or combined with tannic acid, revealed a transient effect with 1 or 2 months reduction of Group 1 allergen levels when applied according to the manufacture's instructions, brushing it into the carpet and allowing it to dry for 4 h before vacuuming (16, 23–25, 28, 34, 35). An application time of 12–18 h had no additional effect (22). In addition, although tannic acid denatures mite allergens, it interferes with several immunoassays to measure indoor allergens and causes an apparent reduction of Group 1 allergen levels (28, 29). Therefore, the temporary reduction of Group 1 allergen levels observed in the studies that utilize benzyl benzoate with tannic acid (28, 29) should be treated with caution.

However, another dust mite allergen avoidance study attempted to reduce dust mite allergen levels over a 12-month period by using bedding encasing combined with benzyl benzoate application in carpet and upholstered furniture and vacuuming with a high-filtration vacuum cleaner. When the results were expressed as total allergen recovered (nanograms per unit area sampled), a progressive and significant reduction of Der p1 allergen levels was observed in bedroom carpets but not in living room carpets and sofas at 6 months in the active group compared with the control group. No significant differences were observed when expressed as allergen concentration (micrograms per gram) (36).

Although the mechanism of action of DOT is unknown, it does not work on contact, but kills mites progressively over the 2 months following its application. In addition, DOT was not applied combined with a denaturant in this study. This is perhaps the reason why a significant decrease of Group 1 allergen levels in the active group was not observed until the end of the study.

Total mite numbers and Group 1 allergen levels in the placebo group, which received the same bedding encasing and weekly vacuuming as the homes in the active group, did not significantly differ from the values obtained in the control group at most sampling times. This observation indicates that vacuum cleaning alone does not significantly reduce mite numbers and their allergens, in agreement with other studies (24, 37, 38).

In conclusion, DOT applied with a steam vacuum machine kills dust mites in carpets and sofas in homes.

DOT application followed by weekly vacuuming with a high-filtration vacuum cleaner progressively reduces dust mites numbers and their allergens in carpets over a 6-month period, and it shows promising results in upholstered furniture.

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