

A simple washing procedure with eucalyptus oil for controlling house dust mites and their allergens in clothing and bedding

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In many countries, allergy to mites is a major risk factor for asthma.¹ Reducing exposure to house dust mite allergen from bedding and clothing is likely to be an important environmental control measure for asthma management and prevention. Although more than 95% of allergen can be removed from items by washing in cold or warm water,² this does not significantly reduce the live mite populations because killing mites requires washing at 55° C.² We previously demonstrated in laboratory studies that several essential oils can be emulsified in low concentrations of the laboratory detergent Tween to form effective acaricides.³ However, this detergent is not available to the general public. In this study we demonstrate that eucalyptus oil, which is a widely available essential oil, can be formulated with a specific kitchen detergent concentrate to form an inexpensive acaricidal wash. We also demonstrate its ability to reduce the recovery of wild populations of live mites found in blankets by more than 95% during normal machine washing.

METHODS

Laboratory testing

Live mites of mainly adult and tritonymph stages (*Dermatophagoides pteronyssinus*) were harvested from laboratory culture and triplicate groups ($n \sim 50$ each) enclosed in small permeable capsules.^{2,3} For emulsification, four parts eucalyptus oil were emulsified with one part Kit (L&K, Rexona; Sydney, Australia) dishwashing liquid detergent concentrate. The groups of three capsules were immersed in 0.05%, 0.1%, 0.2%, or 0.4% final concentration of eucalyptus oil (The Sheldon Drug Company, Sydney, Australia) for either 7.5, 15, 30, or 60 minutes, all at $30 \pm 3^\circ$ C. Aqueous 0.1% or 0.012% Kit was used as a control. After immersion, mites were rinsed to remove eucalyptus oil, drained onto nylon mesh, allowed to recover in enclosed dishes at 75% relative humidity for 12 hours, and classified as dead or alive on the basis of mobility.² All experiments were repeated for each treatment group, providing six observations in each block. The proportion of mites killed in each treatment group

for each treatment duration was calculated with asymptotic 95% confidence limits⁴ (EpiInfo; Centers for Disease Control, Atlanta, Ga.). The effect of treatment type, treatment duration, and the interaction between these variables on the proportion of mites killed was tested by two-way analysis of variance. Group means were compared by using Duncan's multiple range test with SAS software (SAS Institute, Cary, N.C.). The distribution of residuals from this analysis of variance was not significantly different from normal (Shapiro Wilk statistic = 0.98, $p = 0.64$).

Laundry procedures

The heat escape² method was used to determine the number of live mites present in four areas, each 50 cm² (total 200 cm²), for each of 10 woolen blankets that were in normal use. Mean numbers and range of live mites per blanket in the control and the treated groups were 96 (49 to 154) and 74 (32 to 189), respectively. For washing, 100 ml of eucalyptus oil was dispersed in 25 ml of Kit and added to 50 L of warm ($\sim 30^\circ$ C) water in a top-loading washing machine (Hoover; Elite 920, Sydney, Australia). After brief mixing, blankets were added and soaked for 30 or 60 minutes before the normal laundry cycle was completed. Controls were presoaked in a Kit-only solution. Blankets were hung outside until dry (mean drying time, 3.7 hours). One day after washing, live mites in each blanket were sampled from four areas adjacent to the previously sampled areas. The delay was to allow any remaining live mites time to recover. Wilcoxon's rank sum test was used for analysis because of the nonparametric nature of the data.

RESULTS

In the laboratory experiments more than 80% of mites were killed after immersion in 0.2% and 0.4% solutions of eucalyptus oil for 30 and 60 minutes (Fig. 1). There was a significant interaction between treatment type and treatment duration ($p < 0.0001$), indicating that the effect of treatment time varied with the type of treatment. Across all treatment durations, the proportion of mites killed was significantly greater with the 0.05% solution than with the control solution. The proportion of mites killed was also significantly greater with each successive increase in concentration ($p < 0.05$ by Duncan's multiple range test). Among groups of capsules treated with all concentrations of the active solution, the proportion of mites killed did not differ significantly between the 30- and 60-minute treatment times. However, the proportion killed at these treatment durations was greater than the proportion killed after 15 minutes, which was in turn greater than the proportion killed after 7.5 minutes ($p < 0.05$ by Duncan's multiple range test). Although many of the mites were immobile imme-

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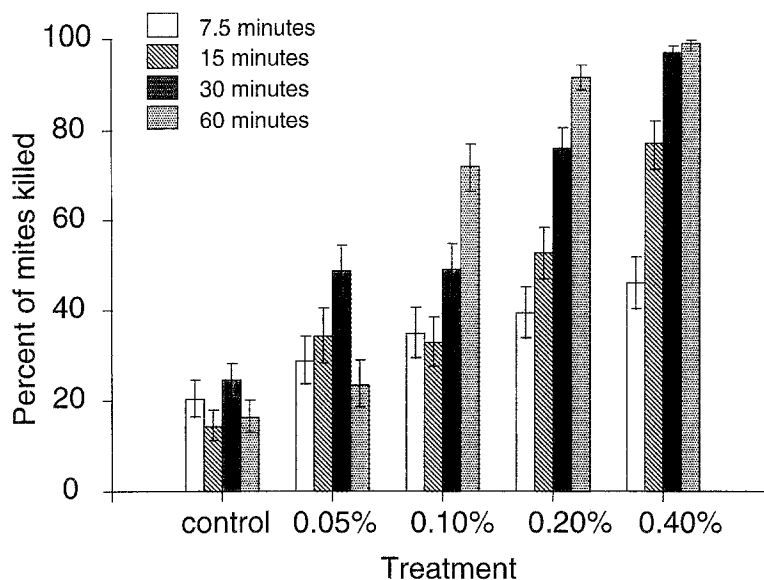


FIG. 1. Interaction of eucalyptus oil concentration and time on the percentage of mites killed at water temperatures of 30° C ($\pm$ 95% confidence interval).

diately after exposure for 7.5 and 15 minutes, most recovered in 12 hours.

The percentage of mites that survived on the six eucalyptus oil-treated blankets was significantly different from the percentage that survived on the four control blankets ($Z = 2.48$, $p = 0.001$). The six eucalyptus oil-treated blankets had a median percentage of mite survival of 0.6% after washing, whereas the four control-treated blankets had a mean percentage of mite survival of 97.6% after washing. Machine washing of woolen blankets in a 0.2% eucalyptus oil solution for 30 and 60 minutes reduced the recovery of live wild-type mites by 97% and 99%, respectively (Fig. 2); this difference in exposure time was not significant ($Z = 0.42$, $p = 0.67$).

DISCUSSION

These laboratory studies established a practical domestic formulation of a eucalyptus oil-based acaricide, the activity of which was time and concentration dependent. We have demonstrated that a 30-minute prewash soak of woolen blankets in this formula reduced the number of live mites that could be recovered by 97%. The low concentration (0.2%) of eucalyptus oil required to achieve this acaricidal effect is practical for normal household purposes and eliminates the necessity of washing in hot water ($>55^{\circ}\text{C}$),² which may be difficult to achieve in practice and may damage some fabrics.

Eucalyptus oil is widely available in supermarkets and health food stores in Australia and elsewhere. It is likely that numerous other locally available plant³ and aromatic tree oils would be suitable, although their acaricidal concentrations would need to be determined. Eucalyptus oil is inexpensive and has relatively low

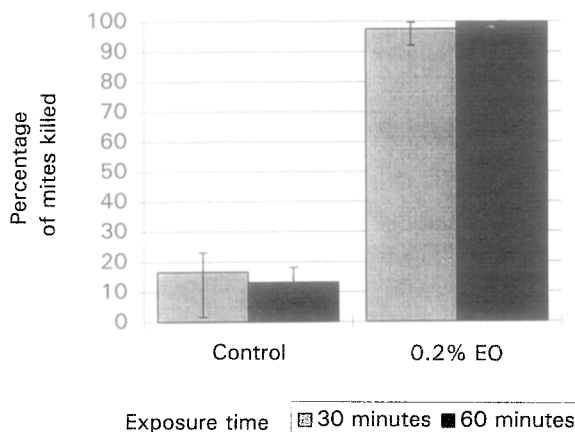


FIG. 2. Effect on mite survival of warm machine washing (30° C) of blankets in a 0.2% eucalyptus oil solution ($\pm$ 95% confidence interval). Percentage killed is based on comparison of numbers of live mites recovered from four sites before washing and then 24 hours after washing from four adjacent sites on each blanket. EO, Eucalyptus oil.

human toxicity,⁵ being classified as a Schedule 6 poison requiring specific packaging (e.g., childproof caps) and labeling. Neat essential oils are not directly soluble in water and need to be blended with a detergent to form an emulsion in water. The detergent used here was the only one suitable of the six available domestic concentrates tested. For testing locally available detergents, the two selection criteria are: (1) the oil should dissolve and form a clear solution at a ratio of one part detergent to three to five parts oil (the less detergent the better, because biocide activity is reduced by the emulsifier); and (2) when a teaspoon of the emulsion is mixed into a glass of water (i.e., ~ 5 ml added to 200 ml), a

milky, opaque solution that is stable (does not "break oil") for at least 10 minutes should form.

We are not aware of significant problems with the use of this formula. The undiluted emulsified oil can function as a solvent and should not come into direct contact with the plastic drum of a washing machine, so it is suggested that the drum be filled with water before the emulsified oil is added. Eucalyptus oil at lower concentrations is already used as a washing additive for delicate fabrics in Australia.² The smell is generally regarded as pleasant and it does not stain. Washing in diluted eucalyptus oil does leave a faint characteristic odor for 2 to 3 days despite rinsing and drying, and some persons may find this irritating.

The precise mechanism by which the mites are killed is unknown. Studies on the Red cotton bug (*Dysdercus koenigii*) suggest that eucalyptus oil may break down lipids and proteins⁶ and that the volatiles of eucalyptus oil can reduce the hatchability of eggs and fecundity of adults.⁷ We have also found that eucalyptus oil at laundry concentrations is ovicidal to *Dermatophagoides pteronyssinus* (P. Bell, unpublished data).

Even bedding that has been encased in an occlusive cover shows an increase in surface allergen with time, and regular washing of all bedding is advised. Such periodic washing is likely to be the most simple and effective component of strategies to maintain control of mite allergens. Bedding is also likely to be the site of the highest allergen concentrations in the home, an important source of aeroallergen exposure, and the most likely domestic site for mite colonization under adverse microclimate conditions. We have previously demonstrated² that normal laundry detergent removes more than 95% of mite allergen (the remainder only elutes slowly from mite bodies, unpublished observation). To this treatment, we now add the capacity to kill most mites.

Mite allergen in bedding arises from both local production by mites and from settling of allergen from airborne dust. It could be expected (though it is not proven) that the rate of allergen accumulation would be

lower in bedding if all local mites were killed during washing. The rate of recolonization of mites would depend on the rate of transfer from adjacent populations determined by opportunity, population size, and mobility, as well as by the buildup of food resources (human skin scales) in bedding, which would be depleted by washing. Because mites are adversely affected by low concentrations of eucalyptus oil vapor, there may be a residual negative effect on mite repopulation. However, whether there is a real clinical advantage in killing mites in addition to just removing allergen can only be established by a separate experiment.

In conclusion, this study shows that it is possible to make a simple, effective, inexpensive laundry acaricidal wash that eliminates the need for very hot water and that may maintain low allergen levels in bedding for longer than normal laundering alone.

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