Acaricidal Effects of Three Plant Oil Extracts Against the Two Dust Mites, *Dermatophagoides farinae* Hughes and *D. pteronyssinus* Trouessart (Acari: Pyroglyphidae)

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(*Received: May 19, 2010*)

ABSTRACT

The acaricidal effects of three plant oil extracts; thyme, clove and camphor oils; against mobile stages of the two house dust mites, *Dermatophagoides farinae* Hughes, and *D. pteronyssinus* Trouessart, were examined using bioassay test. Results were compared with the lethal activity of pirimiphos-methyl as a standard acaricidal compound. After 24 h period and based on the LC_{50} values the most toxic compound against *D. farinae* adult was thyme oil, followed by pirimiphos-methyl, clove and camphor oils. In case of *D. pteronyssinus*, thyme oil was also the most toxic, followed by clove oil, pirimiphos-methyl and camphor oil. Interestingly, the larvae and males of both mite species presented a mortality rate significantly higher than females (about 2 folds) when exposed to the same concentration of oil extracts. The three plant oil extracts, particular thyme and clove oil, have potential for development as new agents for the control of the two tested dust mites.

Key Words: Control; House dust mites; *D. farinae*; *D. pteronyssinus*; Plant oil extracts.

INTRODUCTION

House dust mites are a cosmopolitan guest in human habitation live in dwellings and feed on organic detritus and microbes that grow on cast skin flakes and flourish in the stable environment of dwellings and other detritus. They are a common cause of asthma and allergic symptoms worldwide, affecting more than 50% of all allergic patients and up to 80% of asthma sufferers (Baly *et al.*, 1994 and Pittner *et al.*, 2004). Strong evidence supports a direct relationship between allergens from house dust mites and allergic diseases. Some of the gut enzymes produced by the house mite persist in their fecal matter, and can be strongly allergenic (Platts-Mills *et al.*, 1992 and Kemp, 1996).

Pyroglyphid mites are the most important and worldwide source of house dust allergens in homes (Larson *et al.*, 1969 and Wharton, 1976). They usually account for over 90% of the mite populations in houses (Arlian *et al.*, 1992). The pyroglyphid, American dust mite *Dermatophagoides farinae* Hughes, and European dust mite, *D. pteronyssinus* Trouessart, are cosmopolitan inhabitants of human dwellings in many parts of the world (Panizzi *et al.*, 1993; Kwon and Ahn 2002 and Lee, 2006 & 2008). Both species are found in homes in various areas of Egypt (Rezk, 2004).

Chemical control of dust mites depends on chemical methods such as fumigation and spraying with acaricidal compounds (Arnau and Guerrero, 1994 and Lee, 2007). There is an urgent need to develop safer, environmentally and more selective alternatives against house dust mites in indoor environment. Therefore, this study was conducted to evaluate the acaricidal activity of three plant oil extracts (thyme, clove and camphor) against the mobile stages of *D. farinae* and *D. pteronyssinus*. The toxicity of the three oil extracts was compared with that of the commercial acaricide, pirimiphos-methyl.

MATERIALS AND METHODS

1. Target mites:

Colonies of *D. farina* and *D. pteronyssinus* were isolated from mattress dust and grown in clean dried jars on finely-ground mixture of dust, dried yeast and dried milk (1: 1: 0.5). The stock jars were closed by double layers of muslin and kept in an incubator at 25±2 ^{0}C and 80±5% RH. After five months, large numbers of different stages were available for experimentation (Saint Georges & Gridelete, 1987; Andersen, 1988 and Rezk & Gadelhak, 2003).

2. The tested compound:

Three commercial plant oil extracts namely (thyme, clove bud and camphor oils) and one synthetic acaricide (Pirimiphos-Methyl) were used in the present study. Five concentrations from each compound were prepared (Table 1).

Table (1): Tested essential oils and standard acaricide against two dust mites, *D. farinae* and *D. pteronyssinus*.

Tested compounds	Concentration (ppm)	
	D. farinae	*D. pteronyssinus*
Thyme oil (*Thymus vulgaris* L.)	0.1 – 250	0.1 – 250
Clove bud oil (*Eugenia caryophyllata*)	10 – 500	1 - 200
Camphor oil (*Cinnamomum camphora* L.)	10 – 500	50 – 500
Pirimiphos-methyl	1 - 250	1 – 250

3. Assays:

Filter paper discs, 1.5 cm diameter, were impregnated with 0.1 ml of the tested concentration of chemical solution. Control discs were trated with 0.1 ml of acetone. The filter paper discs were dried for 2 minutes at room temperature. Each disc was placed in the bottom of a glass micro cell (1.5 cm diameter X 2 cm height). Ten females of each mite species were used in the screening assays, since preliminary observations revealed these as the most resistant mobile stage to the selected materials. Females were solitary placed in each micro cell and covered with a lid. Experiments were held at 25 ± 2 ^{0}C. Mortalities were determined 24 h after treatment and mites were considered dead if appendages did not move. All treatments were replicated three times. The LC_{50} and their confidence limits were calculated by probit analysis.

The LC_{50} previously estimated for each product was applied to elucidate the differences in mortality among larvae, males and females for both mite species. Ten individuals per product, replicated 5 times, were used.

Differences between larvae, males and females mortalities for the different compounds were compared by Duncan's Multiple Range Test (DMRT).

RESULTS AND DISCUSSION

The acaricidal activity of thyme, clove, and camphor oils extracts was tested against *D. farinae* and *D. pteronyssinus* adults (Table 2). The synthetic acaricide pirimiphos-methyl served as a reference material. Responses varied according to compound and dose. No mortality was recorded in the control. As judged by the 24 h LC_{50} values, thyme oil (LC_{50}=3.51 ppm) was the most toxic compound against *D. farinae* and was 12 times more toxic than pirimiphos-methyl (LC_{50} = 42.28 ppm), followed by clove oil (LC_{50} = 64.5 ppm) and camphor oil (LC_{50} = 68.13 ppm) (Table, 2). Regarding *D. pteronyssinus*, thyme oil (LC_{50}=7.24 ppm) was 6.7 times more toxic than pirimiphos-methyl (LC_{50} = 48.36 ppm), followed by clove bud oil (LC_{50} = 31.21 ppm) and camphor oil (LC_{50} = 186.25 ppm).

Direct contact bioassay, proved that thyme oil had a great acaricidal effect on adult *D. farinae* and *D. pteronyssinus*. This may be mostly attributed to some compounds like p-cymene and thymole which both of them form more than 50% of thyme oil (El Zemity *et al.*, 2006). In addition, Lee (2004) reported an acaricidal activity constituent of thymol and the structurally related compounds like *p*-cymene isolated from *Foeniculum vulgere* against *Dermatophagoids* spp. Regarding acaricidal activities against house dust mites, these results suggest that *D. pteronyssinus* is more susceptible than *D. farinae* to the clove oil. Interestingly, acaricidal activity of clove oil was about 1.5 times more toxic than pirimiphos-methyl, a synthetic acaricide against *D. pteronyssinus*, the toxic effect of this oil may be due to eugenol which forms 60-80% of clove oil (Kim *et al.*, 2004). Similar results ware obtained by Kim *et al.*, (2003), who found that the clove bud oil revealed potent acaricidal activity against adult *Tyrophagus putrescentiae*. On the other hand, camphor oil is less toxic against *D. farinae* and *D. pteronyssinus* than pirimiphos-methyl, a reference acaricide.

The activity of each compound on larvae and males of both mites *D. farinae* and *D. pteronyssinus* was determined at a dose corresponding with the LC_{50} previously obtained for females (Table 3). For each compound, the percentage mortality of larvae and males was similar except *D. farinae* larvae which were significantly sensitive than males when thyme oil was applied. All mortalities of both larvae and males of *D. farinae* and *D. pteronyssinus* were significantly higher ($P<0.05$) than that for females (Table 3).

Much more interesting, larvae and males of *D. farinae* and *D. pteronyssinus* had a mortality rate significantly higher (about 2 folds) than females when exposed to the same dose (near female LC_{50}) of the selected oils and acaricide. The acaricidal constituents of many plant extracts and essential oils are mainly monoterpenoids. In 2001, Ramos and Castanero found that, death associated with desiccations by monoterpenes in mobile stages of *Tyrophagus putresectiae*. Astigmata have thin and weakly sclerotized cuticles (Evans, 1992) and are commonly associated with moist environments. As such females will tend to lose relatively less water than larvae and males, since they are larger, and their surface/volume ratios are lower, permitting less water loss by diffusion through the body surface (Wharton *et al.*, 1979; Evans, 1992). Consistent with this assumption, mortality of larvae or males of *D. farinae* and *D. pteronyssinus* was higher than females when exposed at the same dose of the selected oils and acaricide.

Recently, many acaricides have been replaced with newer, safer agents owing to the toxicity, resistance and environmental damage caused by earlier agents (Pollart *et al.*, 1987 and Hayes & laws,

Table (2). Acaricidal activity of three plant oil extracts and Pirimiphos-methyl against adult *farinae* and *D. pteronyssinus* by direct contact bioassay during a 24 h exposure.

Compound	Mite species	Slope (± SE)	LC_{50} (ppm)	Confidence Limits 95%	RT*
Thyme oil	*D. farinae*	0.6 ± 8.83	3.51	1.42 – 8.08	12
	D. pteronyssinus	0.61 ± 8.46	7.24	3.17 – 16.19	6.7
Clove oil	*D. farina*	1.2 ± 4.16	64.5	41.45 – 99.85	0.6
	D. pteronyssinus	0.93 ± 2.42	31.21	18.1 – 53.59	1.5
Camphor oil	*D. farinae*	1.17 ± 4.0	68.13	43.68 – 105.73	0.6
	D. pteronyssinus	2.13 ± 0.1	186.25	147.91 – 234.47	0.2
Pirimiphos-methyl	*D. farinae*	1.04 ± 2.82	42.28	25.93 – 68.92	1.0
	D. pteronyssinus	0.8 ± 2.15	48.36	26.25 - 90.26	1.0

*Relative toxicity = LC_{50} value of pirimiphos-methyl / LC_{50} value of each compound.

Table (3): Percentage mortality of larvae, males and females of *Dermatophagoides farinae* and *D. pteronyssinus* by direct contact to the selected oils and acaricide.

Compound	*D. farinae*				*D. pteronyssinus*			
	LC_{50} ppm	Larvae	Males	Females	LC_{50} Ppm	Larvae	Males	Females
Thyme oil	3.51	98± 2.09^a	82 ± 4.8^b	52 ± 3.7^c	7.24	92 ± 3.7^a	94± 2.4^a	48 ± 3.7^b
Clove oil	64.5	88 ± 3.7^a	80 ± 4.4^a	54 ± 4.0^b	31.21	96 ± 2.4^a	92± 4.8^a	54 ± 4.0^b
Camphor oil	68.13	92 ± 3.7^a	86 ± 6.0^a	46 ± 4.0^b	186.25	90 ± 4.5^a	96± 4.0^a	52 ± 3.7^b
Pirimiphos-methyl	42.28	96 ± 2.4^a	90 ± 3.2^a	48 ± 2.0^b	48.36	98 ± 2.0^a	100± 0.0^a	54 ± 2.4^b

Concentration correspond with the LC_{50} previously obtained from females values are mean percentage mortality ± SE (n=50). Values followed by different letters within each compound are significantly different ($P<0.05$).

1991).Thus, of the three plant oil extracts tested, thyme oil is the most promising for possible use against *D. farinae* and *D. pteronyssinus* and clove oil against *D. pteronyssinus* due to the low doses required to produce a high mortality in the immature and adult stages.

In conclusion, thyme and clove oil appear to be effective in the control of house dust mites and may protect humans from their asthma effects. Nevertheless, for practical use of the thyme and clove oils as acaricidal agents, further research should be done on safety issues of these compounds for human health, acaricidal mode of action and formulations to improve the acaricidal potency and stability.

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