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Toxicity and efficacy of selected pesticides and new acaricides to stored product mites (Acari: Acaridida)

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Abstract Stored product mites can often infest stored products, but currently there is little information regarding the efficacy of pesticides that can be used for control. In this study we evaluated several common pesticides formulated from single active ingredients (a.i.) or commercially available mixtures (chlorpyrifos, deltamethrin, beta-cyfluthrin, and a combination of deltamethrin and *S*-bioallethrin), plus an acaricide composed of permethrin, pyriproxyfen and benzyl benzolate, for efficacy against *Acarus siro*, *Tyrophagus putrescentiae*, and *Aleuroglyphus ovatus*. The pesticides were incorporated into the mite diets in a dose range of 10–1000 μg a.i. g^{-1} diet. Concentrations for suppression of 50 and 90% population growth and eradication (rC_0) of mites were fit to linear regression models. None of the tested pesticides gave complete eradication of *A. siro*, which was the most tolerant of the three mite species tested. The most effective pesticide Allergoff 175 CS was a combination

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product (a nano-capsule suspension of permethrin, pyriproxyfen and benzyl benzolate) labeled for dust mites, with rC_0 range of 463–2453 μg a.i. (permethrin) g^{-1} diet depending on the species. Least effective were chlorpyrifos and deltamethrin.

Keywords Allergens · Pesticides · Food · Grain · Storage · Mites

Introduction

Storage sites containing raw grain commodities, animal feeds, and products for human consumption often contain residues and organic dust spillage that can be reservoirs for pest arthropods (Reed et al. 2003; Hubert et al. 2006). Included in this group of arthropods are stored-product mites (Krantz 1955; Athanassiou et al. 2003, 2005), which can be sources of allergens associated with occupational health hazards (Hage-Hamsten-van et al. 1991; Muesken et al. 2000). The control of stored-product arthropods is usually accomplished through the use of organophosphate and pyrethroid insecticides (Collins 2006; White and Leesch 1996; Zettler and Arthur 2000). In Europe, new regulations and policies have led to a re-evaluation and re-registration of all groups of insecticides and their active ingredients and some products may be eliminated from the market. Pyrethroid insecticides are recommended as an alternative to some of the traditional organophosphates due to their quick action, low odor and low toxicity to humans. However stored-product mites have been reported to be fairly tolerant to pyrethroids (Zdarkova and Horak 1974; Zdarkova 1994). Although there is evidence that diatomaceous earths (Cook and Armitage 1999; Palyvos et al. 2006), natural toxic compounds (e.g. essential oils; Sung et al. 2006), and bean flour (Hubert et al. 2007) can control stored mites, they have limited commercial application and use. In addition, currently there are few commercial pesticides that are specifically labeled for use against stored-product mites. The objective of our test was to evaluate several insecticides and an acaricide to control stored-product mites. These pesticides were three traditional insecticides: the organophosphate chlorpyrifos and the pyrethorids deltamethrin and beta-cyfluthrin, a combination of deltamethrin and bioallethrin, and an acaricide that is a combination of permethrin, pyriproxyfen and benzyl benzolate, labeled in Europe to control dust mites.

Material and methods

Pesticides

The sources of the pesticides tested in our experiments were chlorpyrifos (Empire 20, Dow AgroSciences, Indianapolis, IN, USA); deltamethrin (K-Othrine 25WP, Bayer AG, Leverkusen, Germany), beta-cyfluthrin (Responsar SC, Bayer AG); a commercial mixture of active ingredients (a.i.) deltamethrin and bioallethrin (Crackdown Rapide, Aventis Cropscience Inc., Paris, France), and Allergoff 175 CS (CB Pharma, Jaworzno, Poland) a nanocapsule suspension mixture of permethrin, pyriproxyfen, and benzyl benzolate.

Mites

Acarus siro (L.), *Tyrophagus putrescentiae* (Schrank) and *Aleuroglyphus ovatus* (Troupeau) used in our study were obtained from laboratory cultures at the Research

Institute of Crop Production, Prague, Czech Republic (CZ). The individual mite species were mass-reared in frit-chambers (a glass chamber containing filter glass of porosity S0, Kavalier Sazava, Czech Republic) plugged by rubber pierced by a steal tube (5 mm diameter). The muslin covered the both ends of the tube. The chambers were placed into Secador desiccator boxes (P-lab, Prague, CZ) at 85% relative humidity (RH) using saturated KCl solution and held in continual darkness at 25 ± 2°C.

Diets

The rearing diet (Hubert et al. 2007) was composed of oat flakes, wheat germ, lyophilized yeasts and dried fish food (Aqua Lounsky, Praha, CZ) (22:22:5:1 wt). The diet was powdered and sieved. The experimental diet was derived from the rearing diet and contained the individual pesticides in the following concentrations: 0 (control), 10, 100, 250, 500, 1000 μg a.i. g^{-1} diet. For the commercial mixtures Crackdown Rapide and Allergoff 175 CS, the concentrations mentioned are for deltamethrin and permethrin, respectively. The pesticides were homogeneously incorporated into the diet as a suspension in distilled water followed by lyophilization and remoistening of the mixture (Kluh et al. 2005).

Experimental design

The experimental chamber consisted of 20-ml glass tubes (Kavalier, Sazava, CZ) filled with 3 g zeolite and 0.5 g experimental diet. The zeolite was moisturized by 0.15 ml 1% Ajatin solution (Profarma, Prague, CZ) to prevent fungal contamination. Fifty mixed-sex adults of each species were placed in separate tubes covered with muslin. Ten replicates per species, pesticide and concentration were used. The tubes were placed into desiccators at 85% RH and 25°C and kept in darkness during the experiment. The population growth was determined after 21 days by extraction of live mites with Berlese-Tullgren funnels. The mites were collected in a saturated solution of picric acid and counted under a dissecting microscope.

Data analysis

All statistical analyses were done using S-Plus software (Insightful Corporation, Seattle, WA, USA) (see Crawley 2002). To analyze the effect of the different pesticides on mite growth, we tested the effect of concentration, mite species, and their interaction on the final number of mites. The data for the effect of pesticide on population growth were not linear and data were log-transformed. When a significant interaction between species and concentration was found, this could have been because the species either differed in their response to the biocide concentration or because populations of some species simply increased at a faster rate than others. To separate these two effects we repeated the above analyses after standardizing the density data by dividing all the densities within species by the mean density of that species in the treatment. To determine whether this effect altered the conclusions, we also performed an analysis testing the effect of species, pesticide and concentration and their interaction on mite density at the highest concentration only. The doses rC_{50}, rC_{10} and rC_0, causing 50, 90 and 100% mortality, respectively, and rC_{start} (concentration to obtain the same mite density at t = 0 and t = 21 days); were estimated from a linear regression model. To describe the reliability of these values we calculated the standard error of each estimate.

Table 1 Effect of species, pesticide and concentration on the final density of stored product mites

Density	Non-standardized			Standardized		
	df	*P*	R^2	df	*P*	R^2
Species	2	<0.001	0.211	2	0.001	0.004
Pesticide	4	<0.001	0.03	4	<0.001	0.071
Concentration	1	<0.001	0.448	1	<0.001	0.449
Species × pesticide	8	<0.001	0.021	8	<0.001	0.088
Species × concentration	2	<0.001	0.098	2	0.371	–
Pesticide × concentration	4	<0.001	0.006	4	<0.001	0.007
Species × concentration × pesticide	8	0.461	–	8	0.902	–

Concentrations were ln-transformed; the values are standardized by density in the control treatment (no active ingredient added), df error = 1185

Results

The results of the analyses using non-standardized data showed significant effects of species, pesticide, and concentration on mite density (Table 1). Combined permethrin/benzyl-benzoate/pyriproxifen had the strongest effect, followed by deltamethrin/*S*-bioallethrin, beta-cyfluthrin, deltamethrin and chlorpyrifos (Table 2). The separate effect of pesticide concentration on population growth of mite species is shown in Fig. 1. Linear regression was not significant for the final population density of *A. siro* exposed to chlorpyrifos (Table 2). Surprisingly, the final population of *A. siro* was greater in the treatments (10, 100, 250 μg a.i. g^{-1} diet) than in the untreated control (Fig. 1). However, the maximum concentration of chlorpyrifos decreased the final population significantly. In both cases there is also a significant interaction between species and pesticide, and between pesticide and concentration. The interaction between pesticide and concentration can be clearly seen from the rC_{50}, rC_{10} and rC_{start} values (Table 2). The growth of species on control diet differed; the final population density of *T. putrescentiae* was higher than of *A. siro* and *A. ovatus*, implying that *T. putrescentiae* is the most sensitive species, followed by *A. ovatus* and *A. siro*.

In all cases, permethrin/benzyl-benzoate/pyriproxifen had the lowest effective concentration for all species. The pesticide with the highest effective concentration was chlorpyrifos. In the non-standardized data, there is also a significant interaction between species and concentration. However, this interaction disappears after standardization, indicating that it was only a consequence of differential growth rate of the different mite species. When testing the effect of pesticide in the highest concentration only, all pesticides ($F_{4,185} = 61.85$, $P < 0.001$, $R^2 = 0.4$), species ($F_{2,185} = 59.68$, $P < 0.001$, $R^2 = 0.19$), as well as their interaction ($F_{8,185} = 7.8$, $P < 0.001$, $R^2 = 0.1$), were significant.

Discussion

Except for the combination of permethrin/benzyl-benzoate/pyriproxifen, all tested pesticides are specifically labeled for the control of stored-product insects. Although all of them showed some toxic effects on the three mite species, they did not eradicate any species. In particular, *A. siro* was very difficult to control, which is consistent with previous studies

Table 2 Effect of species, pesticides (active ingredients) and their concentrations on the final density of mites

Pesticide	Species	Fitted concentration				Linear regression			
		rC_{50}	rC_{10}	rC_{start}	rC_0	*m*	*b*	R^2	*P*
Permethrin/pyriproxyfen/ benzyl-benzolate	Aca-sir	2 ± 5	600 ± 211	305 ± 249	2453 ± 206	−24 ± 1.06	−34 ± 7.62	0.88	<0.001
	Ale-ova	1 ± 3	172 ± 247	134 ± 274	655 ± 242	−31 ± 1.05	−86 ± 11.48	0.91	<0.001
	Tyr-put	1 ± 3	123 ± 289	236 ± 320	463 ± 304	−74 ± 3.01	−228 ± 33.46	0.88	<0.001
Deltamethrin/*S*-bioallethrin	Aca-sir	13 ± 7	10813 ± 430	4812 ± 518	58468 ± 414	−22 ± 2.6	58 ± 18.69	0.50	<0.001
	Ale-ova	3 ± 3	1867 ± 387	1383 ± 435	9059 ± 378	−26 ± 1.31	10 ± 14.27	0.83	<0.001
	Tyr-put	4 ± 3	3051 ± 425	6910 ± 480	16170 ± 452	−59 ± 2.92	16 ± 32.41	0.83	<0.001
Deltamethrin	Aca-sir	58 ± 7	30080 ± 545	14217 ± 633	143464 ± 529	−20 ± 1.66	35 ± 11.94	0.68	<0.001
	Ale-ova	5 ± 3	2938 ± 428	2161 ± 481	14817 ± 418	−27 ± 1.23	−3 ± 13.26	0.85	<0.001
	Tyr-put	3 ± 3	2510 ± 421	5637 ± 474	13074 ± 447	−59 ± 2.87	28 ± 31.90	0.83	<0.001
beta-Cyfluthrin	Aca-sir	92 ± 7	89268 ± 613	39149 ± 707	497615 ± 596	−20 ± 3.04	77 ± 21.85	0.38	<0.001
	Ale-ova	2 ± 3	996 ± 321	745 ± 361	4596 ± 314	−27 ± 1.07	−21 ± 11.60	0.89	<0.001
	Tyr-put	2 ± 3	1175 ± 379	2532 ± 426	5626 ± 402	−63 ± 2.89	−36 ± 32.04	0.85	<0.001
Chlorpyrifos	Aca-sir	ns	ns	ns	ns	ns	ns	0.02	0.2485
	Ale-ova	18 ± 4	36804 ± 602	25657 ± 680	246207 ± 588	−22 ± 1.44	71 ± 15.66	0.74	<0.001
	Tyr-put	7 ± 4	9425 ± 495	22773 ± 559	56994 ± 526	−55 ± 2.95	95 ± 32.69	0.8	<0.001

Results (fit ± standard errors) were based on linear regression: $y = m * \ln(x + 0.0000001) + b$, with *x*: pesticide concentration (μg a.i. g^{-1} diet), and y: final mite density. rC_{50} and rC_{10}: concentration the causes 50% and 90% population decrease compared to control, respectively; rC_0: concentration necessary for mite eradication; rC_{start}: concentration necessary to bring population back to its initial size, i.e. 50 individuals; Aca-sir: *Acarus siro*, Ale-ova: *Aleuroglyphus ovatus*, Tyr-put: *Tyrophagus putrescentiae*; ns: not significant

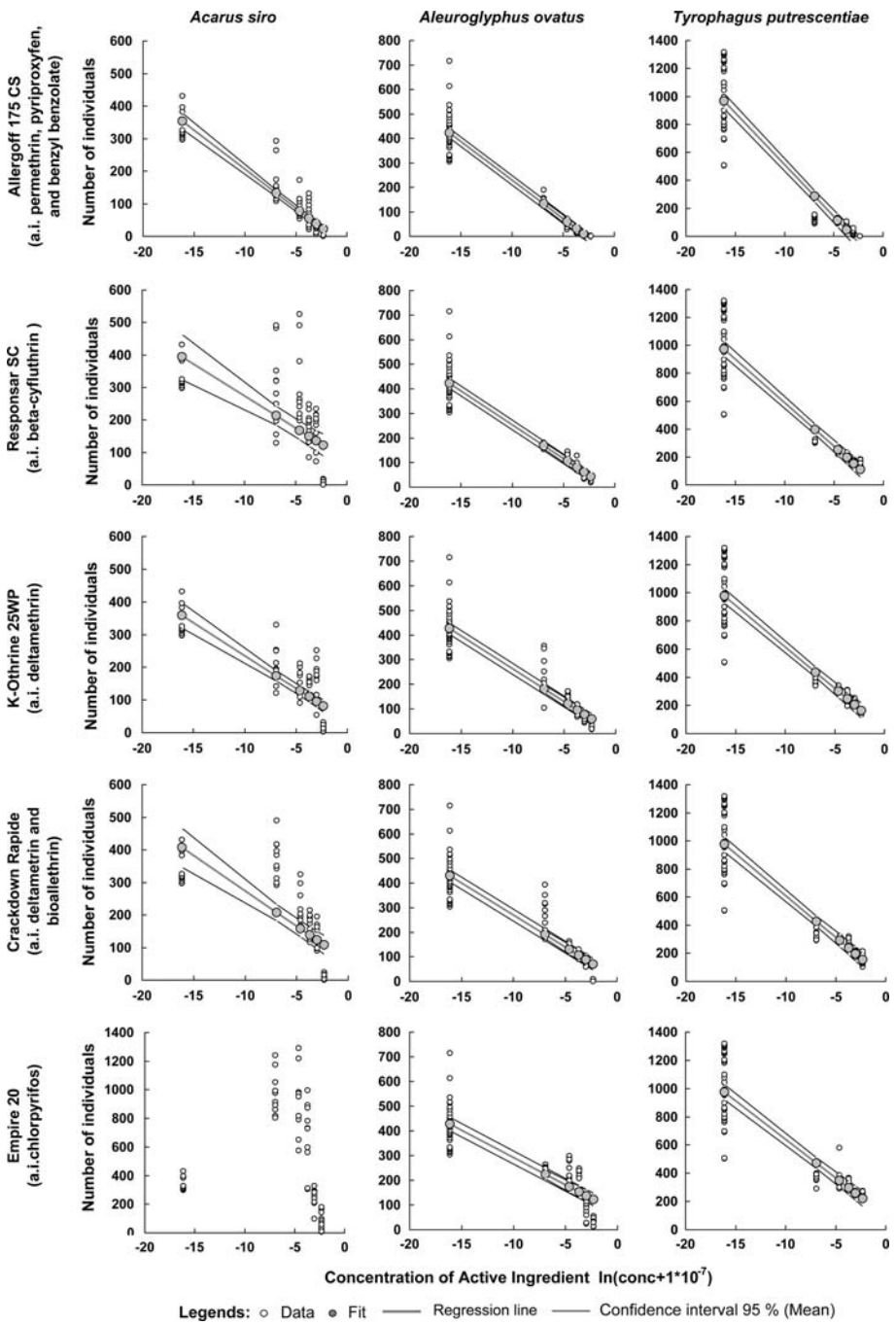

Fig. 1 Linear regression of final mite densities on pesticide concentrations. Description of linear regression parameters is given in Table 2

with this species (Thind and Muggleton 1998). Except for the combination permethrin/benzyl-benzoate/pyriproxifen, the fitted lethal doses (rC_0) are much higher than could be used for practical application (White 1984; White and Leesch 1996).

Chlorpyrifos was the least effective pesticide. In some early studies with stored-product mites, an application rate of 2 μg g^{-1} was reported to give complete control (Wilkin and Hope 1973; Bednarek and Lewandowsk 2006). In this study, the capsules in the specific formulation could have decreased the absorption of a lethal dose by the mites, or *A. siro* may have been resistant to chlorpyrifos, thereby accounting for the reduced control. In another test with chlorpyrifos-methyl, in which the methyl group is simply substituted by the ethyl group in chlorpyrifos, White and Sinha (1990) also reported a lack of effectiveness for mites. The pyrethroid deltamethrin is known to suppress population growth of mites in wheat grain (Collins 2006) and in our test we did achieve some control with deltamethrin. In other studies, bioallethrin was not toxic to stored product mites (Zdarkova and Horak 1974), but we obtained some additive effect compared to using deltamethrin alone. The pyrethroid beta-cyflutrin has been demonstrated to suppress population growth of the spider mite *Tetranychus urticae* after a spraying application (Trindade and Chiavegato 1999), however, in this study there were no differences between toxicity of deltamethrin and beta-cyfluthrin. *A. siro* was particularly tolerant, as indicated by the rC_{50} and rC_{90} values.

The combination of permethrin/benzyl-benzoate/pyriproxifen gave the highest level of population suppression in all species tested. Permethrin controlled stored product mites (Collins 2006) and the house dust mite *Dermatophagoides farinae* (see Hashimoto et al. 2001). Pyriproxyfen prevented immatures of *T. putrescentiae* from reaching the adult stage (Sánchez-Ramos and Castanera 2003). The third compound, benzyl benzoate, is repellant at low concentrations and toxic at higher concentrations to *T. putrescentiae* (Kwon and Ahn 2002; Kim et al. 2004). A possible combination or additive effect is indicated by the fact that in one study, application of 2 μg g^{-1} permethrin on wheat gave little or no mortality of *A. siro* (see Collins 2006), but in our study 2 μg g^{-1} permethrin in combination with other pesticides reduced the population growth of *A. siro* and *T. putrescentiae*. Mite eradication was possible only by using the combination of permethrin/benzyl-benzoate/pyriproxifen.

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