Insecticidal action of the combined use of spinosad and deltamethrin against three stored-product pests in two stored hard-wheat varieties.

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Abstract

The combined use of spinosad with deltamethrin against adults of *Sitophilus oryzae*, *Sitophilus granarius* and *Tribolium confusum* was evaluated in a series of laboratory bioassays in two hard wheat varieties (Athos and Sifnos). Two groups of bioassays were carried out. In the first group of bioassays, spinosad or deltamethrin were applied alone at the tested wheat varieties at the doses of 0.01, 0.1 and 0.5 ppm for spinosad and 0.125 ppm for deltamethrin. In the second group of bioassays, the tested wheat varieties were treated with the combination of the above spinosad rates with 0.125 of deltamethrin. In both series of bioassays, mortality of the tested species was evaluated after 7 d of exposure on the treated wheat varieties at 25°C and 65% r.h. Mortality for all species was always significantly higher in Athos than Sifnos. The highest mortality of *S. oryzae* (73 and 40% for Athos and Sifnos respectively) or *S. granarius* (88% and 58% for Athos and Sifnos respectively) was recorded in the cases that spinosad was applied alone at 0.5 ppm. On the contrary, in the case of *T. confusum*, 0.125 ppm of deltamethrin was significantly more effective than any of the application rates of spinosad either when applied alone or in combination with deltamethrin. Despite the fact that the highest mortality of *S. granarius* adults was recorded after exposure on the wheat varieties treated with 0.1 ppm of spinosad x 0.125 ppm of deltamethrin, in light of the results of the present study, the combination of spinosad with deltmethrin requires further investigation since in most of the tested cases of the present study, single application of spinosad or deltamethrin was more effective or of equal effectiveness than the respective combination of spinosad with deltamethrin.

Keywords: Spinosad, Deltamethrin, *Tribolium, Sitophilus*, Wheat, Variety

1. Introduction

The consumers' growing demand for residue-free goods as well as the fact that many species have now developed resistance to the most commonly used grain protectants (Arthur, 1996) have made essential the evaluation of new insecticides for the control of stored-product pests. Spinosad can be considered as one promising alternative to the currently used grain protectants, as it has low mammalian toxicity (Sparks et al., 2001; Subramanyam et al., 2003) and also it is effective against many of the most important stored-product pests (Fang et al., 2002; Athanassiou et al., 2008; Vayias et al., 2009). Deltamethrin is a pyrethroid insecticide, registered in many parts of the world for stored grain protection. This insecticide is also effective against stored-product pests and can provide a long-term protection that lasts four months or more (Athanassiou et al., 2004).

Insecticides vary regarding efficacy against different target species. For instance, spinosad is moderately effective against *Tribolium* spp. (Fang et al., 2002, Vayias et al., 2009), while deltamethrin is generally more effective against these species (Athanassiou et al., 2004). Hence, the combined use of more than one pesticide is likely to moderate these differences, and provide a grain protectant with satisfactory protection against a wider range of species. In a recent study, Athanassiou et al. (2009) found that the combined use of deltamethrin with chlorpyriphos-methyl successfully controlled stored-grain psocids, which could not be controlled with spinosad or natural pyrethrum. In the present study, the combined use of low spinosad combined with low deltamethrin application rates was evaluated on two hard wheat varieties originating from Greece. This combination was assessed against three stored-product pests which are very common to in bulk grains stored in Greece; two primary colonizers, the rice weevil *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae), and the granary weevil *Sitophilus granarius* (L.) (Coleoptera: Curculionidae), and one secondary colonizer, the confused flour beetle *Tribolium confusum* Jacquelin du Val (Coleoptera: Tenebrionidae).

2. Materials and methods

Unsexed, <2 week-old adults of *S. oryzae, S. granarius* and *T. confusum,* obtained from laboratory cultures reared on hard wheat at 27 ± 1°C and 60 ± 5% relative humidity (r.h.) were used in the study. The tested wheat varieties were Athos and Sifnos obtained from Greek crops. The moisture content of the tested varieties at the beginning of the tests ranged between 10.9 and 11.5%. The grain characteristics of the tested varieties are given in Table 1. In the first group of tests four 1-kg lots of each variety were separately treated with three spinosad application rates (0.01, 0.1 and 0.5 ppm) and one deltamethrin rate (0.125 ppm). In the second group of experiments three 1-kg lots of each variety were separately treated with the combination of each spinosad rate x 0.125 ppm deltamethrin (e.g. 0.01 ppm spinosad x 0.125 ppm deltamethrin; 0.1 ppm spinosad x 0.125 ppm deltamethrin; 0.5 ppm spinosad x 0.125 ppm deltamethrin). In addition, a 1-kg lot of each grain was sprayed only with distilled water and served as the untreated control. From each treated (or untreated) 1-kg lot of grain, of a specific variety, three 30 g samples were obtained and used as a bioassay substrate with the above insect species at 25°C and 65% r.h. Mortality was assessed after 7 d of exposure of the tested species on the treated or untreated grains. The total procedure was repeated three times (3 x 3 vials per treatment) by preparing new 1-kg lots from each wheat variety each time. Data were analyzed separately for each species by a two way ANOVA with mortality as the response variable with grain variety and treatment as the main effects. For the separation of means the Tukey and Kramer HSD test was used at $P<0.05$ (Sokal and Rholf, 1995).

Table 1 Grain characteristics of the tested wheat varieties.

	Wheat variety	
Specifications	**Athos**	**Sifnos**
Brush length	Short	short
Kernel shape	Ovoid	semi elongate
Mean (± SE) weight of 100 kernels (g)	4.3 ± 0.1	5.2 ± 0.1
Mean (± SE) kernel size of 100 kernels (ml)	5.7 ± 0.1	6.9 ± 0.2
Protein content (%N x 5.7)	16.5	14.5
Gluten index	22.14	64.4
Mean (± SE) bulk density (g/lt)	742.5 ± 0.2	744.5 ± 0.6

3. Results

Mortality of the tested species on the untreated wheat varieties was negligible and did not exceed 2.5% in any of the tested cases. Irrespective of the treatment, susceptibility of all of the tested species was overall higher in treated Athos in comparison with treated Sifnos with the sole exception of the combination of 0.1 ppm spinosad x 0.125 ppm deltamethrin against *S. oryzae*, while the reverse was noted (Table 2). Among the tested species, *T. confusum* was the most tolerant to spinosad, deltamethrin or their combination. *Sitophilus granarius* was more susceptible than *S. oryzae* in all of the tested cases. Generally, the combination of spinosad with deltamethrin did not appear to be compatible for *S. oryzae* or *T. confusum* since the highest mortality ratio for those species was with 0.5 ppm of spinosad for the former and 0.125 ppm for the latter when these substances were applied alone rather than in combination. For instance, although 73% of the exposed *S. oryzae* adults were dead after exposure on Athos variety treated with 0.5 ppm of spinosad, mortality was only 39% on the same variety treated with 0.5 ppm spinosad x 0.125 ppm (Table 2). With *T. confusum*, mortality from exposure to spinosad was generally low and did not exceed 15% on both varieties, while it was slightly increased to 22% when 0.125 ppm of deltamethrin was applied to Athos prior to application of 0.5 ppm spinosad. The combination of deltamethrin with spinosad also gave low mortality levels with *T. confusum*. Mortality was only 28% on wheat treated with 0.125 ppm of deltamethrin alone, and was significantly higher or of equal effectiveness with the tested spinosad combinations (Table 2). An additive effect of deltamethrin with spinosad was observed only when *S. granarius* was exposed to 0.1 ppm spinosad x 0.125 ppm deltamethrin. In the latter case, efficacy of 0.1 ppm spinosad x 0.125 ppm deltamethrin increased to 90% in Athos and 62% in Sifnos while efficacy of the respective spinosad or deltamethrin doses when applied alone did not exceed 54% in Athos or 26% in Sifnos. The combination of 0.5 ppm of spinosad x 0.125 ppm deltamethrin, slightly improved the performance of the respective spinosad dose in Athos against *S. granarius*, but did not demonstrate an additive effect in Sifnos against the same species (Table 2).

Table 2 Mean (±SE) mortality of adults of *S. oryzae*, *S. granarius* and *T. confusum* after 7 d exposure on two hard wheat varieties treated with spinosad alone, deltamethrin alone as well as with the combination of spinosad and deltamethrin. Within a given species, means followed by the same letter are not significantly different (lowercase letters for treatments; uppercase letters for varieties). For treatments *df*=6, 62; For varieties *df*=1, 17; Tukey and Kramer HSD test at $P<0.05$.

			Mortality (%)	
	Dose (ppm)		Wheat variety	
Species	**Spinosad**	**Deltamethrin**	**Athos**	**Sifnos**
S. oryzae	0	125	47.4 ± 3.1 Abc	25.9 ± 3.0 Bbc
	0.01	0	42.6 ± 3.9 Abc	20.0 ± 1.0 Bc
	0.1	0	53.3 ± 4.4 Ab	29.3 ± 3.1 Bb
	0.5	0	73.0 ± 3.7 Aa	39.6 ± 3.7 Ba
	0.01	0.125	15.2 ± 3.6 Ad	19.3 ± 2.9 Ac
	0.1	0.125	17.8 ± 3.1 Ad	24.4 ± 3.1 Abc
	0.5	0.125	35.9 ± 2.3 Ac	32.6 ± 2.7 Aab
S. granarius	0	125	66.3 ± 5.1 Ab	25.2 ± 1.4 Bc
	0.01	0	53.3 ± 2.4 Ab	17.8 ± 1.8 Bc
	0.1	0	57.4 ± 2.3 Ab	25.2 ± 2.2 Bc
	0.5	0	88.1 ± 2.1 Ab	58.1 ± 3.0 Bab
	0.01	0.125	61.1 ± 3.6 Aa	28.1 ± 1.9 Bc
	0.1	0.125	90.0 ±2.5 Aa	62.2 ± 3.3 Ba
	0.5	0.125	91.1 ± 3.0 Aa	49.6 ± 3.9 Bb
T. confusum	0	125	28.1 ± 2.2 Aa	23.3 ± 3.0 Aa
	0.01	0	1.9 ± 0.8 Ac	0.7 ± 0.5 Ac
	0.1	0	4.8 ± 1.0 Ac	3.0 ± 1.5 Ac
	0.5	0	14.4 ± 3.7 Ab	8.9 ± 1.6 Ab
	0.01	0.125	2.6 ± 1.3 Ac	1.9 ± 0.6 Ac
	0.1	0.125	3.7 ± 0.9 Ac	4.4 ± 1.4 Ac
	0.5	0.125	21.5 ± 5.4 Aa	18.6 ± 3.3 Aa

4. Discussion

In our study, the combined use of spinosad with deltamethrin at low application rates was not successful against *S. oryzae* or *T. confusum*, but was effective against *S. granarius*. With *S. granarius* only the specific combination of 0.1 ppm spinosad x 0.125 deltamethrin was highly effective since effectiveness of the remainding combinations against the same species was lower than or at least equal to the effectiveness of a single spinosad or deltamethrin application. It is possible that spinosad could synergize deltamethrin under specific conditions and for specific insect species; therefore more extensive research on this combination is needed. The fact that effectiveness of deltamethrin or spinosad varied between the tested varieties could be attributed to differences in physical or chemical characteristics of the grain, variations in insect behavior after contact with the treated kernels of a specific variety, or a combination of the above factors. Sifnos had more elongated, heavier and larger kernels compared to Athos. Also, gluten index was higher in Sifnos than in Athos. The above characteristics may have affected the efficacy of the tested formulations, as the treated species were overall less susceptible on Sifnos than in Athos, although in the case of *T. confusum* the differences were not significant. Kernel size is likely to play an important role in efficacy of insecticides, since better distribution of the toxicant is achieved on smaller kernels compared to larger ones (Huang and Subramanyam, 2007). As a result, insects may be able to more easily avoid the treated areas of larger kernels and consequently, avoid contact with the toxic substance (Athanassiou et al., 2003).Since distribution of deltamethrin or spinosad was better in the smaller kernels of Athos compared to Sifnos, this could partially explain the higher mortality that occurred in Athos compared to Sifnos. Nevertheless, correlation of grain characteristics with efficacy of grain-insecticides is not always feasible. Fang et al. (2002), found that a significant variation in the performance of spinosad against several stored-product pests occurred among different wheat classes. The authors however, could not correlate this differential spinosad performance to grain kernel diameter, kernel weight, kernel hardness, protein content, dockage, or fiber. Hence, differential performance of insecticides among commodities or even varieties of a given commodity is a very complicated issue, and requires further experimental work.

Generally, pyrethroids provide quick mortality against insects, and this fact may influence their combination with other slow-acting insecticides. Le Patourel and Singh (1984) found that the combination of silica, which is a slow acting insecticide, with high doses of permethrin, cypermethrin and deltamethrin, did not exhibit any additive effect against the red flour beetle, *Tribolium castaneum* (Herbst) due to rapid knock down caused by the pyrethroids. In our study, additive effect was evident only in the case of the low doses of the insecticides tested, and this could be attributed to the delayed mortality, allowing both insecticides to act. Additional measurements at shorter intervals (e.g. 24 or 48 h) are necessary to clarify the basis of this hypothesis.

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