

Full Length Research Paper

Assessing the Bioassay Performance of Spinosad Dust Against Storage Pests: Insights from Laboratory Trials in Kenya

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Laboratory bioassay trials were conducted at the National Agricultural Research Laboratories and Kiboko sub-centre in Kenya to determine the efficacy of spinosad dust 0.125% admixed with shelled grains, compared to a “cocktail” of pirimiphos-methyl 1.6% and permethrin 0.3% (Actellic super dust) against four important storage-insect pests: *Sitophilus zeamais* (Motsch), *Tribolium castaneum* (Herbst), *Rhyzopertha dominica* (F.) and the larger grain borer, *Prostephanus truncatus* (Horn). Spinosad dust was applied at 0.35, 0.7 and 1.44 parts per million (ppm), and Actellic super dust at 10.5 ppm. All treatments were significantly ($P=0.05$) better than the control except when applied against *T. castaneum*. Spinosad at 0.7 and 1.44 ppm was able to control *S. zeamais* over the 24-week trial period. Similarly, all treatments gave good control over *P. truncatus* and *R. dominica*, with not apparent significant differences ($P=0.05$) between treatments on the latter. On *P. truncatus*, Spinosad showed better performance than Actellic super dust ($P=0.05$). All levels of spinosad dust appeared to perform better on *P. truncatus* compared to Actellic super dust, but spinosad dust, unlike Actellic super dust, was unable to control *T. castaneum*. The evidence from this trial suggests that spinosad dust may have potential in controlling major storage-insect pests, with special applicability against the destructive larger grain borer, *P. truncatus*.

Key words: Spinosad, Actellic, dust formulations, storage pests, pest control.

INTRODUCTION

Spinosad is a metabolite of the soil actinomycete bacterium, *Saccharopolyspora spinosa* and is used for the control of insect pests in several crops (Thompson et al., 1997). Its activity is attributed to the metabolites spinosyns A and D the fermentation products of the bacterium (Metz and Yao, 1990). This is a product within the naturalyte class, and is grouped with other natural metabolites. Spinosad have a novel molecular structure and mode of action that provides good crop protection typically associated with synthetic products (Thompson et al., 1997). The product is toxic to insects by ingestion or contact thereby acting on the insect nervous system at the nicotinic acetylcholine and gamma-aminobutyric acid (GABA) receptor sites (Sparks et al., 2001). Extensive worldwide studies have demonstrated that liquid

spinosad provides effective control of key pests in many crops, including vegetables, cotton, turf and ornamentals (Bret et al., 1997). It meets requirements for low environmental and human risk (Saunders and Bret, 1997) and is ideal for insecticide resistance management (Salgado, 1997). While spinosad has considerable efficacy against pests of stored products (Toews and Subramanyan, 2003), no documented data exist on the efficacy of its dust formulation. Spinosad is the active ingredient in the dust that was formulated for a pilot trial. Although data on efficacy of the liquid formulation have been reported for the control of stored product insect pests (Toews and Subramanyan, 2003; Athanassiou et al., 2008; Vayias et al., 2010), this was the first trial worldwide to evaluate spinosad in form of a dust for its efficacy against major insect pests found in storage systems in Kenya. In the present study, we evaluated the efficacy and persistence of spinosad for the control of *Prostephanus truncatus* (Horn) (Coleoptera: Bostrichidae),

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Rhyzopertha dominica (F) (Coleoptera: Bostrichidae) *Sitophilus zeamais* (Motschulsky) (Coleoptera: Curculionidae) and *Tribolium castaneum* (Herbst) (Curculionidae:Tenebrionidae) by assessing adult mortality over a period of 24 weeks.

MATERIALS AND METHODS

Experimental location

The bioassay was laboratory based and carried out at two sites: the National Agricultural Research Laboratories (NARL) in a controlled temperature and humidity (CTH) room maintained at 25±2°C and 70±5% relative humidity, and Kiboko sub-centre, where the larger grain borer is endemic. At the latter site, the bioassay was conducted at ambient room conditions with mean temperature and relative humidity of 28±2°C and 65±5% respectively due to the unavailability of a CTH room.

Test insects

S. zeamais (Motsch.), *T. castaneum* (Herbst) and *R. dominica* (F.) were obtained from stock culture at NARL. *P. truncatus* (Horn) and additional *S. zeamais* were obtained from stock culture at Kiboko sub-centre. Kiboko is a semi-arid area located at latitude 2°1'S, longitude 37°7'E and altitude 975 m asl.

Insecticide dusts

Spinosad 0.12% dust was supplied by Dow agro-Sciences through Lachlan Kenya Ltd. A cocktail of pirimiphos-methyl 1.6% and permethrin 0.3% dust (Actellic super dust) was procured from the Kenya Farmers Association, stockists of agricultural products in Nairobi.

Grain treatment and bioassay

Twenty-eight kilograms of wheat grain from NARL was sieved and sterilized at 130°C for 1 h to kill any internal infestation. The grains were then weighed into 100-g samples in 1-L Kilner jars and thereafter treated with spinosad and Actellic super dusts. Each treatment was replicated four times, giving a total of 20 jars per batch for each insect species plus a control. Applications of dusts were calculated from the recommended rate of 50 g per 90-kg bag of maize, giving 0.06 g per 100 g of grains. For spinosad dust, a half rate (0.03 g) and double rate (0.12 g) were included to assess the optimal level of efficacy. This translated into dosage rates of 0.35, 0.7 and 1.44 parts per million (ppm) for spinosad dust and 10.5 ppm for Actellic super dust. After applying the dust in each jar, care was taken to ensure thorough admixing through constant shaking and swirling of each jar.

These treatments formed seven batches, for use at four week intervals over the 24-week trial period (weeks 0, 4, 8, 12, 16, 20 and 24), which was approximately equivalent to a six - month storage season. Laboratory bioassay for *P. truncatus* was based at Kiboko. Jars were prepared as described earlier using maize grain. For the NARL bioassay, the jars were transferred into the CT room and allowed to equilibrate for 24 h before the introduction of 30 adult, mixed-sexes *R. dominica* of unknown age into the first batch of jars. Assessment of knockdown mortality was done after seven days of exposure. The knocked-down insects were subsequently returned to 7.5 x 2.5 cm flat-bottomed test tubes with 10 g of maize

flour, and another mortality count was taken after seven days. By combining the two mortality counts, a final insect mortality was obtained. For the next batch of jars *T. castaneum* was used. Similar procedures to assess mortality were followed for *S. zeamais* only and *P. truncatus* and *S. zeamais* at Kiboko using maize grain, but there, the jars were placed on shelves in the laboratory at ambient room conditions.

Statistical analysis

Mortality records for both sites were taken for the entire 24 week period. The data were then subjected to analysis of variance using statgraphics software to determine any significant differences between the treatments. Mean numbers were compared using ANOVA Fisher's test.

RESULTS

S. zeamais

Actellic super dust at 10.5 ppm and spinosad 0.125% at 0.7 and 1.4 ppm produced total control of *S. zeamais* at week 24 (Table 1). There were no significant differences ($P=0.05$) in insect mortality between the two dosage rates and Actellic super (10.5 ppm) over the same period. However, spinosad at 0.35 ppm showed a strange efficacy profile: it was effective up to week 12, dipped to a level of 18.3 insects (61%) mortality response at week 20, and picked up again at 24 weeks at a level of 29.5 insects (98.3%). Overall, all the treatments were comparatively effective on *S. zeamais* over the trial period.

P. truncatus and *R. dominica*

Spinosad 0.125% gave good control over *P. truncatus* (Table 1) and *R. dominica* (Table 2) at all levels, ranging from 95 to 100% and comparable with that of Actellic super. *P. truncatus* and *R. dominica*, both bostrichids, were highly susceptible to all treatments. Spinosad at all levels managed to control these pests effectively, recording an overall kill of 29.8 (99.3%) and 30 (100%) for *P. truncatus* and *R. dominica* over the 24 week trial period. There were no significant differences ($P=0.05$) between the treatments except at weeks 20 and 24, when Actellic super showed a lower mortality response for *R. dominica* (86.7%) and *P. truncatus* (85%), respectively. As the post-treatment period progressed, the efficacy of Actellic super unlike that of spinosad decreased comparatively, with a significant difference ($P=0.05$) occurring at week 24. Spinosad at 0.35 ppm, the lowest level, performed better than Actellic super.

T. castaneum

With *T. castaneum*, the situation was more complex:

Table 1. Mean mortality (no. of dead adults, n =30) of *Sitophilus zeamais* and *Prostephanus truncatus* exposed to grains admixed with spinosad (n=4) compared to actellic super dust

Treatment (Dusts)	Concentration (%)	Application rate	Dosage (ppm)	Post-treatment period (weeks)						
				0	4	8	12	16	20	24
<i>Sitophilus zeamais</i>										
Spinosad	0.125	25 g/90 kg	0.35	13.3 ^b	28.8 ^b	29.3 ^a	27.8 ^a	21.5 ^b	18.3 ^b	29.5 ^a
Spinosad	0.125	50 g/90 kg	0.7	28.5 ^a	29.8 ^{ab}	30.0 ^a	27.8 ^a	28.5 ^a	26.5 ^a	29.8 ^a
Spinosad	0.125	100 g/90 kg	1.4	29.8 ^a	29.7 ^{ab}	30.0 ^a	29.3 ^a	29.5 ^a	28.0 ^a	30 ^a
Pirimiphos-methyl/Permethrin	1.6/0.3	50 g/90 kg	10.5	29.8 ^a	30.0 ^a	30.0 ^a	30.0 ^a	30.0 ^a	30.0 ^a	30.0 ^a
Control	0.0			0.0 ^c	1.5 ^c	0.3 ^c	0.0 ^b	0.5 ^c	0.5 ^c	0.0 ^b
LSD (p<0.05)				2.91	1.09	1.17	2.32	5.53	4.31	0.67
Standard deviation				1.89	0.70	0.76	1.51	3.59	2.80	0.44
CV (%)				9.34	2.94	3.17	6.57	16.3	13.54	1.83
<i>Prostephanus truncates</i>										
Spinosad	0.125	25 g/90 kg	0.35	30.0 ^a	30.0 ^a	30.0 ^a	29.8 ^a	28.5 ^a	29.3 ^a	29.8 ^a
Spinosad	0.125	50 g/90 kg	0.7	30.0 ^a	30.0 ^a	30.0 ^a	29.3 ^a	29.0 ^a	30.0 ^a	30.0 ^a
Spinosad	0.125	100 g/90 kg	1.4	30.0 ^a	30.0 ^a	30.0 ^a	30.0 ^a	29.8 ^a	30.0 ^a	29.8 ^a
Pirimiphos-methyl/Permethrin	1.6/0.3	50 g/90 kg	10.5	28.5 ^b	29.3 ^b	29.8 ^a	29.5 ^a	29.8 ^a	28.8 ^a	25.5 ^b
Control	0.0			0.0 ^c	0.0 ^c	0.0 ^b	0.0 ^b	0.0 ^b	0.0 ^b	0.0 ^c
LSD (p<0.05)				0.69	0.66	0.34	1.37	1.39	1.34	0.94
Standard deviation				0.45	0.43	0.22	0.89	0.90	0.87	0.61
CV (%)				1.89	1.80	0.93	3.75	3.84	3.69	2.66

Mean number followed by different letters are significant difference P<0.05.

Actellic super dust at 10.5 ppm killed 22.8 insects (76%) at 24 weeks, but spinosad 0.125% dust was not effective even at the highest rate of 1.4 ppm, which killed only 3.3 insects (11%) over the same period. As shown in Table 2. *T. castaneum* was less susceptible to spinosad than the other insects. Fewer than 18 dead insects were recorded at week 20. There were significant differences between spinosad and Actellic super throughout the trial period for this species. A progressive decrease was apparent by the 24th week for all the treatments, but in all cases Actellic super outperformed spinosad dust.

DISCUSSION

Spinosad at all dosage levels and Actellic super were able to control *S. zeamais*, *P. truncatus* and *R. dominica*. Maize grain admixed with 0.7 and 1.4 ppm Spinosad achieved similar control level of *S. zeamais*. The performance of spinosad dust at these doses was comparable to that of Actellic dust. The benefit of doubling the dose rate to 1.4 ppm was very low and we conclude that 0.7 ppm dose rate confers adequate control of *S. zeamais*. Athanassiou et al. (2008a) found that an increase in liquid spinosad dose from 0.5 to 1.0 ppm

resulted in similar mortality level for *Sitophilus oryzae*, a member of Curculionidae family as *S. zeamais*. The results of our study are in agreement with these findings as no significant differences were observed between 0.7 and 1.4 ppm dose rates. Other studies have found lower mortality caused by spinosad on maize grain for *S. oryzae* and that effectiveness of spinosad to be less on maize than wheat, rice and barley (Athanassiou et al., 2008b; Vayias et al., 2009). Our study found high mortality when *S. zeamais* was exposed to maize grain treated with spinosad and can only speculate that within the Curculionidae

Table 2. Mean mortality (no. Of dead adults, n =30) of three stored – product insect species exposed to grains admixed with spinosad (n=4) compared to actellic super dust.

Treatment (Dusts)	Concentration (%)	Application rate	Dosage (ppm)	Post-treatment period (weeks)						
				0	4	8	12	16	20	24
<i>Sitophilus zeamais</i>										
Spinosad	0.125	25 g/90 kg	0.35	20.5 ^b	19.5 ^b	28.0 ^b	15.5 ^c	22.5 ^a	24.5 ^b	25.5 ^b
Spinosad	0.125	50 g/90 kg	0.7	28.5 ^a	28.5 ^a	29.5 ^a	25.5 ^b	27.3 ^a	28.0 ^{ab}	29.5 ^a
Spinosad	0.125	100 g/90 kg	1.4	29.8 ^a	29.8 ^a	30.0 ^a	28.8 ^{ab}	22.8 ^a	29.5 ^{ab}	29.8 ^a
P-methyl/Permethrin	1.6/0.3	50 g/90 kg	10.5	30.0 ^a	30.0 ^a	30.0 ^a	30.0 ^a	29.8 ^a	30.0 ^a	30.0 ^a
Control	0.0			0.5 ^c	0.0 ^c	0.0 ^c	0.0 ^d	0.0 ^b	0.5 ^c	0.0 ^c
LSD (p<0.05)				4.35	2.65	1.09	3.73	9.67	5.16	1.96
Standard deviation				2.62	1.72	0.71	2.42	6.27	3.35	1.27
CV (%)				12.89	7.97	3.01	12.12	30.68	14.87	5.54
<i>Rhizopertha dominica</i>										
Spinosad	0.125	25 g/90 kg	0.35	30.0 ^a	30.0 ^a	30.0 ^a	30.0 ^a	30.0 ^a	30.0 ^a	29.8 ^a
Spinosad	0.125	50 g/90 kg	0.7	30.0 ^a	30.0 ^a	30.0 ^a	29.8 ^a	30.0 ^a	30.0 ^a	30.0 ^a
Spinosad	0.125	100 g/90 kg	1.4	30.0 ^a	30.0 ^a	30.0 ^a	30.0 ^a	30.0 ^a	30.0 ^a	30.0 ^a
P-methyl/Permethrin	1.6/0.3	50 g/90 kg	10.5	30.0 ^a	30.0 ^a	29.8	28.3 ^a	30.0 ^a	26.0 ^b	30.0 ^a
Control	0.0			0.5 ^b	1.5 ^b	0.0 ^b	0.0 ^b	0.0 ^b	0.3 ^c	0.0 ^b
LSD (pW0.05)				0.40	1.19	0.0	1.92	0.0	2.15	0.34
Standard deviation				0.26	0.77	0.0	1.25	0.0	1.40	0.22
CV (%)				1.07	3.19	0.0	5.29	0.0	6.01	0.93
<i>Tribolium castaneum</i>										
Spinosad	0.125	25 g/90 kg	0.35	6.0 ^c	3.5 ^b	8.0 ^c	1.5 ^{bc}	5.0 ^{bc}	11.5 ^b	1.3 ^b
Spinosad	0.125	50 g/90 kg	0.7	15.8 ^b	2.3 ^{bc}	12.0 ^{bc}	1.8 ^{bc}	6.8 ^b	11.8 ^b	1.3 ^b
Spinosad	0.125	100 g/90 kg	1.4	17.5 ^b	1.5 ^{cd}	16.0 ^b	2.5 ^b	11.3 ^b	18.0 ^b	3.3 ^b
Pirimiphos-methyl/Permethrin	1.6/0.3	50 g/90 kg	10.5	30.0 ^a	30.0 ^a	30.0 ^a	30.0 ^a	25.0 ^a	28.0 ^a	22.8 ^a
Control	0.0			1.5 ^c	0.0 ^d	1.3 ^d	0.0 ^c	0.0 ^c	0.0 ^c	0.0 ^b
LSD (pW0.05)				6.34	1.88	5.09	2.21	6.49	7.80	4.60
Standard deviation				4.12	1.22	3.28	1.43	4.21	5.06	2.98
CV (%)				29.1	16.35	24.49	20.02	43.9	36.56	52.34

Mean number followed by different letters are significant different P<0.05.

family differences in response to spinosad efficacy exist.

Further study is required to evaluate efficacy of spinosad dust on maize, wheat, rice and sorghum

for the control of both *S. oryzae* and *S. zeamais*. The Bostrychidae family is known to be more

highly susceptible to pyrethroids than organophosphates. Spinosad had a comparable effect on *P. truncatus* and *R. dominica*, suggesting a similarity in mode of action between spinosad and pyrethroids, and accords with spinosad's mode of action as described by Salgado (1997). Spinosad dust provided good protection of maize and wheat grains for 24 weeks against the two pests' infestation. This finding confirms results reported by Vayias et al. (2010). In our study, at the end of 24 – week period spinosad achieved 29.8 (99.3%) mortality of *P. truncatus* compared to 28.7 (95.7%) of Actellic dust while that of *R. dominica* was complete control. Again, data demonstrate that doubling the dose rate from 0.7 to 1.4 ppm achieved very little in the overall mortality response. This observation is in agreement with the findings of Athanassiou et al. (2008b). *T. castaneum* was more tolerant to spinosad dust at all dosage levels. The overall mortality response over the 24 –week period was 7.6 (25.2%) compared to 28 (93.2%) for Actellic dust. Similar observations have been reported by studies elsewhere (Nayak et al., 2005; Athanassiou et al., 2008b). The findings of this study are in accord that *T. castaneum* is highly tolerant to spinosad and that high survival rate was observed even at double dose rate of 1.4 ppm.

In conclusion, the observed ability of spinosad to control *S. zeamais*, *P. truncatus* and *R. dominica* suggests that if it is applied to grain early, before infestation sets in, it may be able to protect durable products against opportunistic secondary pests that attack damaged grains. The inability of spinosad to directly control *T. castaneum* – a problematic secondary pest with documented resistance to selected storage chemicals (Speirs et al., 1967; Dyte and Blackman, 1967; Warui, 1974; De Lima, 1977) is added reason to apply the dust before grains are damaged. The resultant protected sound grain will not be attacked by *T. castaneum* a pest capable of only feeding on grains that had prior damage. Based on our findings, the mortality response for the four insect species to spinosad can be classified in a descending order as *R. dominica* > *P. truncatus* > *S. zeamais* > *T. castaneum*. This trial demonstrates some potential, to be further investigated in field trials, for spinosad dust to control storage insect pests and serve as a protectant of stored products.

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