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Effectiveness of spinosad against seven major stored-grain insects on corn

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Abstract In January 2005, the United States Environmental Protection Agency registered spinosad as a stored grain protectant. No referenced data on the efficacy of spinosad on corn in suppressing major stored-grain insects have been published. In this paper, we evaluated the efficacy of spinosad against seven major stored-grain insects on shelled corn in the laboratory. Insect species tested were the red flour beetle, *Tribolium castaneum* (Jacquelin duVal); rusty grain beetle, *Cryptolestes ferrugineus* (Stephens); lesser grain borer, *Rhyzopertha dominica* (F.); sawtoothed grain beetle, *Oryzaephilus surinamensis* (L.); rice weevil, *Sitophilus oryzae* (L.); maize weevil, *Sitophilus zeamais* (Motschulsky); and Indian meal moth, *Plodia interpunctella* (Hübner). Corn kernels were treated with spinosad at 0, 0.1, 0.5, 1, and 2 active ingredient (a.i.) mg/kg for controlling the seven species. Beetle adults or *P. interpunctella* eggs were introduced into each container holding 100 g of untreated or insecticide-treated corn. The seven insect species survived well on the control treatment, produced 28 to 336 progeny, and caused significant kernel damage after 49 days. On spinosad-treated corn, adult mortality of *C. ferrugineus*, *R. dominica*, *O. surinamensis*, *S. oryzae*, and *S. zeamais* was > 98% at 1 and 2 mg/kg after 12 days. Spinosad at $\geqslant$ 0.5 mg/kg completely suppressed egg-to-larval survival after 21 days and egg-to-adult emergence of *P. interpunctella* after 49 days, whereas 16% *T. castaneum* adults survived at 1 mg/kg after 12 days. Spinosad at 1 or 2 mg/kg provided complete or near complete suppression of progeny production and kernel damage of all species after 49 days. Our results indicate that spinosad at the current labeled rate of 1 mg/kg is effective against the seven stored-grain insect pests on corn.

Key words stored-grain insects, spinosad, efficacy assessment
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Introduction

In the US, several surveys showed that stored-grain insects are managed primarily by chemical methods, particularly with organophosphate grain protectants (NASS, 1999; Fang *et al*., 2002a). Pirimiphos-methyl (Actellic®) at the rate of 8 mg/kg is registered for use on stored corn and sorghum (U.S. EPA, 1999, 2003). This organophosphate was reviewed under the 1996 Food Quality Protection Act (FQPA) and received a favorable review (Anonymous, 1997; U.S. EPA, 2000). Malathion is also registered for use on coarse and small grains. However, this insecticide is not a suitable grain protectant because it breaks down rapidly and many stored-grain insects have developed high levels of resistance to the insecticide (Subramanyam & Hagstrum, 1995). There is also documented evidence of insect resistance to pirimiphos-methyl (Subramanyam & Hagstrum, 1995). Therefore, alternative pest management strategies are needed in post-harvest commodities to replace or complement the existing organophosphate grain protectants

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(Fang *et al*., 2002a).

Spinosad, a reduced-risk commercial insecticide based on the fermentation products of an actinomycete bacterium, has been labeled for use on over 250 crops in more than 50 countries (Mertz & Yao, 1990; Thompson *et al*., 2000). Spinosad has low mammalian toxicity and degrades quickly when exposed to sunlight (Thompson *et al*., 2000), but it was relatively stable in stored-grain (Fang *et al*., 2002b; Flinn *et al*., 2004). Spinosad acts on the nicotinic acetylcholine receptors and this mode of action is unique among other known insecticides (Thompson *et al*., 2000). These benign properties make it an ideal product for use in stored grain.

Laboratory and field tests on stored wheat showed that spinosad at 1 (a.i.) mg/kg of grain was effective against several insect pests including the lesser grain borer *Rhyzopertha dominica* (F.), rusty grain beetles *Cryptolestes ferrugineus* (Stephens), and Indian meal moth *Plodia interpunctella* (Hübner) (Fang *et al*., 2002a, 2002b; Flinn *et al*., 2004; Huang *et al*., 2004).

In 2005, the United States Environmental Protection Agency registered spinosad at 1 mg/kg as a grain protectant on commodities including wheat, corn, rice, millets, oats, sorghum, and barley (Bruggink, 2005). In this paper, we report the effectiveness of spinosad evaluated under laboratory conditions during 2002–2003 against seven major insect species associated with stored corn. The results of this study provided information to support the registration of spinosad as a stored-corn protectant.

Materials and methods

Insecticides

A liquid formulation of spinosad (SpinTor® 2SC) containing 240 (a.i.) mg/mL was obtained from Dow AgroSciences (Indianapolis, Indiana, USA). Insecticide was diluted in distilled water to make solutions of different concentrations for grain treatment.

Insects

Seven major stored-grain insect species were tested in the study. The insects were *T. castaneum*; *C. ferrugineus*; *R. dominica*; sawtoothed grain beetle, *Oryzaephilus surinamensis* (L.); rice weevil, *Sitophilus oryzae* (L.); maize weevil, *Sitophilus zeamais* (Motschulsky); and *P. interpunctella*. Colonies of *T. castaneum*, *C. ferrugineus*, and *R. dominica* were established from field collections in Kansas in 2002 and 2003. *O. surinamensis*, *S. oryzae*, and *S. zeamais* were laboratory-reared colonies, which have been maintained in the Department of Grain Science and Industry, Kansas State University since 1999. A colony of *P. interpunctella* was established from moths collected from two Kansas farms in 2002. *T. castaneum* and *C. ferrugineus* were reared on a mixture of 95% rolled oats and 5% (by weight) Brewer's yeast. *R. dominica*, *O. surinamensis*, and *S. oryzae* were reared on whole, hard red winter wheat, while *S. zeamais* was reared on yellow dent corn. *P. interpunctella* was reared on a chicken-mash diet (Subramanyam & Cutkomp, 1987). All insect colonies were reared at 28°C, 65% RH with a 14 : 10 h L : D cycle. Unsexed adults (1–3-week-old) of the six beetle species and eggs (⩽ 24-h-old) of *P. interpunctella* were used in the experiments.

Grain treatment and insect exposure

Shelled yellow dent corn, used in the experiments, was obtained from a local farmer and was frozen at −13°C for 1 week to kill any live insects. The corn was cleaned of broken kernels and debris by hand and by using a 4.76-mm round-holed sieve (US Standard Sieve No. A 12/64 inch sieve).

Corn (1 kg) of 13% moisture was treated with 1 mL of distilled water (control treatment) or 1 mL of aqueous suspension of spinosad to provide rates of 0.1, 0.5, 1, or 2 mg/kg. Fresh-prepared insecticide solutions (< 2 h) were applied to corn kernels using a micropipette. Corn treated with distilled water (control treatment) or insecticide solutions were placed in separate 1.9-L glass jars with lids and tumbled on a ball-mill roller for 10 min to obtain uniform coverage of applied materials on the kernels (Fang *et al*., 2002a). After tumbling, 100 g of corn were infested with 25 unsexed adults (1–3-week-old) of a beetle species in each 150-mL plastic container or with 50 eggs (⩽ 24-h-old) of *P. interpunctella* in each 0.45-L glass jar. After infestation, plastic containers and glass jars were fitted with wire mesh and filter paper lids and were placed in growth chambers maintained at 28°C, 65% RH with a 14:10 h L : D cycle. Plastic containers were examined after 12 days to assess adult mortality of the beetle species and after 49 days to determine progeny production and kernel damage for the beetles. Egg-to-larval survival of *P. interpunctella* was checked after 21 days, while egg-to-adult emergence, progeny production, and kernel damage for this species were recorded after 49 days. Beetle adults unable to move when prodded gently with a hair brush were considered dead. The number of progeny for *T. castaneum*, *C. ferrugineus*, and *O. surinamensis* was based on all visible live larvae, pupae, and adults found in corn, whereas that for *R. dominica*, *S. oryzae*, and *S. zeamais* included only adults as these three species complete larval and pupal development within kernels. Kernels with feeding or adult emergence holes were considered damaged. Kernel damage was assessed by counting all damaged kernels in each container or jar. A completely random design was used for all tests. Each combination of insect species, insecticide rate, and exposure duration was replicated five times, and each replicate was treated separately.

Data analysis

Data were analyzed by species. Mortality of adult beetles was expressed as percentages, and these percentages were transformed to angular values before analysis (Zar, 1984). The number of *P. interpunctella* larvae recovered after 21 days, egg-to-adult emergence of *P. interpunctella* after 49 days, and kernel damage and progeny production for each species were transformed to log ($x + 1$) scale to normalize treatment variances before analysis (Zar, 1984). Transformed data were subjected to one-way ANOVA to test differences among insecticide rates (SAS Institute, 1988). Treatment means were separated using the Fisher's Protected LSD test at the $\alpha = 0.05$ level (SAS Institute, 1988). For beetle species, the 25 adults originally used to infest corn were subtracted from the progeny data before statistical analysis. Untransformed means and standard errors are presented in the tables.

Results

Mortality of beetle adults

Adults of *R. dominica* and *C. ferrugineus* were most susceptible to spinosad, followed by *S. oryzae*, *S. zeamais*, and *O. surinamensis*, while *T. castaneum* was the least susceptible species. Corn treated with spinosad at 0.5, 1 and 2 mg/kg was very effective against adults of *C. ferrugineus*, *R. dominica*, *S. oryzae*, and *S. zeamais*. The effect of spinosad on adult mortality of the four species was significant (F, range among species = 92.88−298.05; df = 4,20; $P < 0.000\ 1$). After 12 days, mortality of these four species on control corn was < 4%, whereas at the rate of 0.5 mg/kg or higher, it was 100% for *C. ferrugineus*, *R. dominica*, and *S. oryzae* and greater than 99% for *S. zeamais* (Table 1). Mortality of *O. surinamensis* increased as the rate of spinosad increased ($F = 143.98$; df = 4,20; $P < 0.000\ 1$), reaching 98% at 1 mg/kg and 100% at 2 mg/kg (Table 1). Adult mortality of *T. castaneum* on spinosad-treated corn increased significantly as spinosad rate increased ($F = 135.67$; df = 4,20; $P < 0.000\ 1$) (Table 1). However, unlike the other beetle species, 16% and 1.6% *T. castaneum* adults survived the rates of 1 and 2 mg/kg, respectively (Table 1).

Progeny production of beetles

Spinosad was extremely effective in suppressing progeny production of *R. dominica* ($F = 1\ 013.06$; df = 4,20; $P < 0.000\ 1$). A mean ± SEM of 24.2 ± 2.6 progeny of *R. dominica* per container were produced on untreated (control) corn, while no progeny were produced at 0.1 mg/kg (Table 1). Spinosad also was effective in reducing the progeny production of *C. ferrugineus* ($F = 36.74$; df = 4,20; $P < 0.000\ 1$). On control corn, 23.4 ± 3.0 progeny of *C. ferrugineus* were found in each container, whereas 3.6 ± 2.2 progeny were observed at 0.1 mg/kg and no progeny was observed at 0.5 mg/kg or higher rates (Table 1). There was an inverse relationship between progeny production and spinosad rate for *T. castaneum*, *O. surinamensis*, *S. oryzae*, and *S. zeamais* (F, range among species = 64.63−170.38; df = 4,20; $P < 0.000\ 1$). A large number of progeny (178−336 per container) of *O. surinamensis*, *S. oryzae*, and *S. zeamais* were produced on control and corn treated with spinosad at 0.1 mg/kg, but these numbers were reduced significantly ($P < 0.05$) at the rate of 0.5 mg/kg and were zero at 1 or 2 mg/kg for the three species (Table 1). For *T. castaneum*, progeny production was 28.0 ± 4.2 per container on control treatment after 49 days. This number was reduced by 91% at 0.5 mg/kg and > 96% at 1 and 2 mg/kg.

Kernel damage by beetles

Kernel damage was significantly different among treatments after 49 days for the six beetle species (F, range among species = 46.79−424.47; df = 4,20; $P < 0.000\ 1$). The number of kernels damaged in control treatment ranged from 5.4 ± 0.7 per container for *C. ferrugineus* to 279.2 ± 4.3 for *S. zeamais* (Table 1). Kernel damage was significantly ($P < 0.05$) reduced at 0.1 mg/kg for *T. castaneum*, *C. ferrugineus*, and *R. dominica* and at 0.5 for all six species. Corn treated with spinosad at 1 or 2 mg/kg was essentially free from damage by any of the six species.

Egg-to-larval survival, egg-to-adult emergence, progeny production, and kernel damage of P. interpunctella

Spinosad provided excellent control of *P. interpunctella*. The effects of spinosad on egg-to-larval survival after 21 days, and egg-to-adult emergence, larval progeny production and kernel damage after 49 days were all significant (F, range among parameters = 525.4−1340.7; df = 4,20; $P < 0.000\ 1$) (Table 2). On control corn, approximately 57% of the 50 eggs developed to larvae after 21 days and 45% of these eggs emerged to adults after 49 days. An average 35.0 ± 4.8 larval progeny and 73.8 ± 2.1 damaged ker-

Table 1 Adult mortality, progeny production, and kernel damage (mean ± SEM) of six stored-grain beetle pests on untreated and spinosad-treated corn.

Spinosad rate (mg/kg)	Adult mortality after 12 days(%)[†]	No. progeny per container after 49 days[†]	No. damage kernels per container after 49 days[†]
		Tribolium castaneum	
0	1.6 ± 1.0 c	28.0 ± 4.2 a	41.8 ± 1.2 a
0.1	2.4 ± 1.6 c	35.0 ± 4.7 a	26.2 ± 1.5 b
0.5	36.8 ± 2.3 c	2.4 ± 0.7 b	7.0 ± 0.7 c
1.0	84.0 ± 5.5 b	1.0 ± 0.6 c	1.0 ± 0.8 d
2.0	98.4 ± 1.0 a	0.2 ± 0.2 c	0.0 ± 0.0 e
		Cryptolestes ferrugineus	
0	2.8 ± 1.3 c	23.4 ± 3.0 a	5.4 ± 0.7 a
0.1	83.2 ± 4.6 b	3.6 ± 2.2 b	1.6 ± 0.5 b
0.5	100.0 ± 0.0 a	0.0 ± 0.0 c	0.0 ± 0.0 c
1.0	100.0 ± 0.0 a	0.0 ± 0.0 c	0.0 ± 0.0 c
2.0	100.0 ± 0.0 a	0.0 ± 0.0 c	0.0 ± 0.0 c
		Rhyzopertha dominica	
0	2.4 ± 1.0 c	24.2 ± 2.6 a	52.4 ± 3.8 a
0.1	95.2 ± 2.3 b	0.0 ± 0.0 b	0.0 ± 0.0 b
0.5	100.0 ± 0.0 a	0.0 ± 0.0 b	0.0 ± 0.0 b
1.0	100.0 ± 0.0 a	0.0 ± 0.0 b	0.0 ± 0.0 b
2.0	100.0 ± 0.0 a	0.0 ± 0.0 b	0.0 ± 0.0 b
		Oryzaephilus surinamensis	
0	4.0 ± 2.2 d	336.0 ± 8.0 a	42.6 ± 2.6 a
0.1	4.8 ± 3.9 d	277.8 ±27.3 a	35.2 ± 1.4 a
0.5	62.4 ± 3.7 c	3.4 ± 0.9 b	5.2 ± 1.0 b
1.0	98.4 ± 1.0 b	0.0 ± 0.0 c	0.0 ± 0.0 c
2.0	100.0 ± 0.0 a	0.0 ± 0.0 c	0.0 ± 0.0
		Sitophilus oryzae	
0	2.4 ± 1.6 c	310.6 ± 19.4 a	250.6 ± 7.3 a
0.1	55.2 ± 12.6 b	256.4 ± 14.4 a	197.0 ± 4.7 a
0.5	100.0 ± 0.0 a	4.4 ± 2.2 b	4.6 ± 1.5 b
1.0	100.0 ± 0.0 a	0.0 ± 0.0 c	0.0 ± 0.0 c
2.0	100.0 ± 0.0 a	0.0 ± 0.0 c	0.0 ± 0.0 c
		Sitophilus zeamais	
0	3.2 ± 1.5 c	296.8 ± 21.6 a	279.2 ± 4.3 a
0.1	51.2 ± 7.8 b	178.8 ± 27.5 a	200.4 ± 25.2 a
0.5	99.2 ± 0.8 a	13.0 ± 4.7 b	11.6 ± 2.9 b
1.0	100.0 ± 0.0 a	0.0 ± 0.0 c	0.0 ± 0.0 c
2.0	100.0 ± 0.0 a	0.0 ± 0.0 c	0.0 ± 0.0 c

[†]Means within a column for each insect species followed by different letters are significantly different ($P < 0.05$; by Fisher's Protected LSD test).

nels per container were observed on the control corn after 49 days. On spinosad-treated corn, a total of only one live larva and a total of one damaged kernel were observed at 0.1 mg/kg. Spinosad at 0.5, 1, or 2 mg/kg completely suppressed egg-to-larval survival, egg-to-adult emergence, and totally suppressed both progeny production and kernel damage.

Table 2 Egg-to-larval survival, egg-to-adult emergence, larval progeny production, and kernel damage (mean ± SEM) of *Plodia interpunctella* on untreated and spinosad-treated corn.

Spinosad rate (mg/kg)	No. live larvae per jar after 21 days†	No. adults emerged per jar after 49 days†	No. larval progeny per jar after 49 days†	No. damaged kernels per jar after 49 days†
0	28.6 ± 1.4 a	22.6 ± 1.9 a	35.0 ± 4.8 a	73.8 ± 2.1 a
0.1	0.2 ± 0.2 b	0.0 ± 0.0 b	0.0 ± 0.0 b	0.2 ± 0.2 b
0.5	0.0 ± 0.0 b	0.0 ± 0.0 b	0.0 ± 0.0 b	0.0 ± 0.0 b
1.0	0.0 ± 0.0 b	0.0 ± 0.0 b	0.0 ± 0.0 b	0.0 ± 0.0 b
2.0	0.0 ± 0.0 b	0.0 ± 0.0 b	0.0 ± 0.0 b	0.0 ± 0.0 b

†Means within a column followed by different letters are significantly different ($P < 0.05$; by Fisher's Protected LSD test).

Discussion

The United States Environmental Protection Agency in January 2005 registered spinosad at 1 mg/kg as a grain protectant on commodities including wheat, corn, rice, millets, oats, sorghum, and barley (Bruggink, 2005). However, commercial formulations of spinosad for stored-grain will not be available until the international tolerance level is approved from countries that regularly purchase US grain. Our study showed that spinosad applied to corn at 1 mg/kg provided near complete suppression of the seven common stored-grain insect pests such as *C. ferrugineus*, *R. dominica*, *O. surinamensis*, *S. oryzae*, *S. zeamais*, *P. interpunctella*, and *T. castaneum*. Huang and Subramanyam (2004) reported that spinosad applied as a liquid to corn was also very effective against a pirimiphos-methyl tolerant strain of the rice moth, *Corcyra cephalonica* (Stainton). These results showed that spinosad at 1 mg/kg can be used as an effective grain-protectant against common stored-grain insects on corn.

Compared to the other six species, *T. castaneum* adults appeared less susceptible to spinosad. Reduced susceptibility of *T. castaneum* to spinosad relative to other stored-grain insects was also observed on stored wheat (Fang *et al.*, 2002a). However, our data showed that spinosad on corn at 1 mg/kg was more effective against *T. castaneum*, *O. surinamensis*, and *S. oryzae* than on wheat. For example, the 14-day mortality on four classes of wheat treated with spinosad at 1 mg/kg ranged only 1.6%−54.7% for *T. castaneum* and 4.1% to 76.3% for *O. surinamensis* (Fang *et al.*, 2002a). The 12-day mortality of the two species on corn treated with spinosad at the same rate and under similar test conditions was much higher as shown in the present study (84.0% for *T. castaneum* and 98.4% for *O. surinamensis*). Similarly, progeny of the two species and a significant number of damaged kernels were observed after 45 days on spinosad-treated wheat (Fang *et al.*, 2002a), whereas complete suppression of progeny production and kernel damage of the two beetle pests was observed on spinosad-treated corn after 49 days. A similar tend was also observed for *S. oryzae*. The reasons for the differences in performance of spinosad against these pests between corn and wheat could be related to spinosad residue coverage, distribution, and retention on the kernel surfaces (Amos *et al.*, 1986; Pomeranz *et al.*, 1988; McGaughey *et al.*, 1990). Relative to wheat, corn has a smaller total surface area for a certain amount of kernels that may result in a higher level of spinosad residue on corn than on wheat. A higher concentration of spinosad residues on corn could provide a higher level of efficacy. The variable performance of spinosad suggests that it is necessary to validate its efficacy on different commodities.

In recording progeny production, we could not count the number of eggs produced for all of the seven insect species, and the number of larvae and pupae of *R. dominica*, *S. oryzae*, and *S. zeamais*. However, the extremely effective suppression of spinosad against adult survival for five out of the six beetle species, egg-to-larval survival and egg-to-adult emergence of *P. interpunctella*, and kernel damage for all of the seven insects indicates that the progeny production, if any, would be negligible on the treatments containing spinosad at 1 mg/kg for all of these seven species.

Summary

Spinosad at the current registered rate, 1 mg/kg, on corn was very effective against six out of the seven major stored-grain insects including *C. ferrugineus*, *R. dominica*, *O. surinamensis*, *S. oryzae*, *S. zeamais*, and *P. interpunctella*. Although spinosad at the rate of 1 mg/kg provided an incomplete suppression of *T. castaneum* adults, it effectively suppressed progeny production and kernel damage of the insect at this rate. Our results suggest spinosad at 1 mg/kg can be used as an effective grain protectant in managing major stored-grain insects on corn.

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Note

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