

Spinosad: an effective replacement for organophosphate grain protectants

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Abstract

Spinosad is a bacterial fermentation product that has low mammalian toxicity. It exhibits both stomach and contact activities against insects. Spinosad is commercially registered on 250 crops in over 24 countries. It is not registered for use on stored grains. Laboratory and field tests on wheat and corn have shown that this product is effective against the lesser grain borer, rice weevil, flat grain beetle, rusty grain beetle, confused flour beetle and larvae of the Indian meal moth at 1 mg/kg. On stored grain, adults of the red flour beetle and saw-toothed grain beetle are less susceptible to spinosad than other species. In farm bins, spinosad on wheat at 0.1–6 mg/kg was stable for one year. This was confirmed by residue data and bioassays against the lesser grain borer and red flour beetle. The trend in species susceptibility was consistent among grain types (corn and wheat), but varied among wheat classes. On concrete, steel, floor tile and waxed floor tile surfaces, spinosad provided >98% mortality of adults of eight stored-product beetles exposed for 24 h to deposits of 0.05 and 0.1 mg cm⁻². Activity against a variety of stored-product insects, persistence in farm-stored grain, and low mammalian toxicity make spinosad a viable alternative to currently registered organophosphate grain protectants, such as malathion, chlorpyrifos-methyl and pirimiphos-methyl.

Keywords: Spinosad; Efficacy assessment; Persistence; Contact toxicity

Introduction

The organophosphates, malathion, chlorpyrifos-methyl (Reidan[®]) and pirimiphos-methyl (Actellic[®]), are currently registered by the United States Environmental Protection Agency (USEPA) for treating stored wheat and corn, respectively, to manage insects. However, under the 1996 Food Quality Protection Act (Anonymous, 1997), which set tougher standards for reviewing registered pesticides, the future of these organophosphate grain protectants remains uncertain. In addition, resistance in key stored-product insects has limited the effectiveness of these three protectants (Zettler and Cuperus, 1990; Subramanyam and Hagstrum, 1995). Therefore, alternative pesticides or pest management strategies are urgently needed to replace currently registered organophosphates.

Spinosad, the fermentation product of the bacterium *Saccharopolyspora spinosa* Mertz and Yao (Mertz and Yao, 1990), is a commercial insecticide reported to be effective against insect pests in the orders Lepidoptera, Diptera and Thysanoptera, and some species of Coleoptera and Orthoptera (Sparks et al., 1995; Cloyd and Sadof, 2000; Peck and McQuate, 2000; Thompson et al., 2000). Spinosad affects the insect nervous system at the nicotinic acetylcholine receptor and GABA receptor sites (Salgado, 1997, 1998). It has low mammalian toxicity (Thompson et al., 2000) and degrades quickly on exposure to sunlight (UV light) (Brunner and Doerr, 1996; Saunders and Bret, 1997; Liu et al., 1999). It is labelled for use on over 250 crops in 24 countries (Gary Thompson, Dow AgroSciences; personal communication). At the present time, spinosad is not registered for use on stored grains.

Our research team has been evaluating the performance of spinosad against stored-product insects since 1997. This paper summarises our research results from 1997 through July 2002.

Effectiveness against stored-product insects on different wheat classes and grain types

Effectiveness of spinosad against the lesser grain borer, *Rhyzopertha dominica* (F.); rice weevil, *Sitophilus oryzae* (L.); saw-toothed grain beetle, *Oryzaephilus surinamensis* L.; red flour beetle, *Tribolium castaneum* (Herbst); confused flour beetle, *Tribolium confusum* (Jacquelin du Val); rusty grain beetle, *Cryptolestes ferrugineus* (Stephens); flat grain beetle, *Cryptolestes pusillus* (Schöenherr); maize weevil, *Sitophilus zeamais* (Motschulsky), and Indian meal moth, *Plodia interpunctella* (Hübner) was tested on different classes of wheat (durum, hard red winter, hard red spring, soft red winter wheat) and on yellow dent corn.

Subramanyam et al. (in press) reported that the mortality of *R. dominica*, *S. oryzae*, *O. surinamensis* and *T. castaneum* was significantly higher ($P < 0.05$) on spinosad-treated hard red spring wheat of 15% moisture than on untreated wheat. *Rhyzopertha dominica* was the most susceptible species, followed by *S. oryzae*, *O. surinamensis* and *T. castaneum*. Spinosad at 1 mg/kg killed all *R. dominica* adults after 8 days. Complete mortality of *S. oryzae* adults occurred at 3 mg/kg after 8 days and at 1 mg/kg after 14 days. *Oryzae-*

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philus surinamensis mortality at both exposure durations at 1–20 mg/kg ranged from 60–99% and that of *T. castaneum* ranged from 22–72%. At all spinosad rates, *R. dominica* progeny production was suppressed by 99.9–100%. Suppression of *S. oryzae* progeny production was 72% at 1 mg/kg and ≥91% at rates ≥3 mg/kg. Progeny production of *O. surinamensis* was significantly suppressed (99% reduction) only at 20 mg/kg. Complete suppression of *T. castaneum* progeny occurred at 6 mg/kg. Across all four species, some damage to spinosad-treated wheat was evident, which suggested that insects poisoned with spinosad did not die quickly and continued to cause grain damage. Generally, kernel damage was inversely related to spinosad rate. Subramanyam et al. (in press) also tested *P. interpunctella* emergence on spinosad-treated wheat. Compared with egg-to-adult emergence on untreated wheat, *P. interpunctella* emergence was suppressed by 99–100% at 1–20 mg/kg. About 444 untreated wheat kernels were damaged by *P. interpunctella* larvae, whereas only 6–20 spinosad-treated kernels were damaged.

In another test, Subramanyam et al. (in press) found that mortality of *R. dominica*, *S. oryzae*, *O. surinamensis* and *T. castaneum* adults on spinosad-treated soft red winter wheat was significantly greater ($P < 0.05$) than on untreated wheat after 7 and 14 days of exposure. However, mortality on spinosad-treated wheat was similar at both exposure periods. Mortality of *R. dominica* adults was 99–100% even at 0.1 mg/kg. All *S. oryzae* adults were killed at 1 mg/kg. The mortality of *O. surinamensis* and *T. castaneum* adults was 46–56% and 28–39%, respectively. The 21-day egg-to-larval mortality of *P. interpunctella* was 95.2% at 0.1 mg/kg and 99.6% at 1 mg/kg.

All adults of *C. ferrugineus*, *C. pusillus* and *T. confusum* exposed to hard red winter wheat treated with 1 mg/kg of spinosad died within 14 days (Subramanyam et al., in press). Progeny production at this rate was also completely suppressed.

Fang et al. (2002) reported a similar trend in species susceptibility in their tests with different wheat classes. All *R. dominica* adults were killed at 0.1 mg/kg after 7 days. At 1 mg/kg, all *S. oryzae* adults were killed on durum wheat, and mortality on other wheat classes ranged from 69–93%. Mortality of *O. surinamensis* and *T. castaneum* after 7 and 14 days of exposure at 1 mg/kg ranged from 46–76% on durum wheat, while mortality of these species on other wheat classes was <15%. Spinosad at 1 mg/kg suppressed progeny production of *R. dominica* (91.2–100% reduction) and *T. castaneum* (72.4–100% reduction) on all wheat classes. At 1 mg/kg, suppression in progeny production of *S. oryzae* was 85.2–95% and that of *O. surinamensis* was 75–93.8% only on durum wheat. There was significant ($P < 0.05$) positive correlation between progeny production and kernel damage caused by *R. dominica*, *S. oryzae* and *T. castaneum*. In a study by Fang et al. (2002), spinosad activity against insects varied among wheat classes, but not among exposure durations (7, 14 and 45 days). Performance of spinosad against *P. interpunctella* was consistent among all four wheat classes. Spinosad at 1 mg/kg killed nearly all *P. interpunctella* larvae (97.6–99.6%), suppressed egg-to-adult emergence completely (0–1.2 of adults emerged/50 eggs) and decreased kernel damage significantly (0.2–3.4% of kernels were damaged).

The mortality of *R. dominica* adults on spinosad-treated yellow dent corn was similar at 0.1, 1 and 10 mg/kg and

ranged from 85.3–98.7% (Bh. Subramanyam, unpublished data). However, the mortality of *S. zeamais* and *T. castaneum* increased with an increase in spinosad rate. The mortality of these two species was significantly greater ($P < 0.05$) at 10 mg/kg (ranges for *S. zeamais* and *T. castaneum*: 81.4–93.3% and 26.2–62.7%) than at 0.1 (5.3–28.4% and 1.3–2.6%) or 1 mg/kg (28.5–42.2% and 3.9–24.3%). Spinosad suppressed *R. dominica* progeny production (100% reduction) and kernel damage (97.4% reduction) at 1 mg/kg. The progeny production of, or kernel damage caused by, *T. castaneum* was significantly lower on spinosad-treated corn than on untreated corn, and the magnitude of reduction proportionally increased with spinosad rate. Spinosad failed to suppress progeny production of, and kernel damage caused by, *S. zeamais*. The 21-day mortality of *P. interpunctella* larvae on spinosad-treated corn increased with spinosad rate. Mortality was 91.3% at 1 mg/kg and 98% at 10 mg/kg.

On both wheat and corn, *R. dominica* and *P. interpunctella* were highly susceptible to spinosad. Effective control of *S. oryzae* with spinosad may be achieved at ≥3 mg/kg. Adults of *O. surinamensis*, *T. castaneum* and *S. zeamais* are less susceptible to spinosad; however, some reduction in progeny production of the first two species can be expected, as they are external kernel feeders.

Influence of grain temperature and moisture on activity against *R. dominica* adults

L. Fang and Bh. Subramanyam (unpublished data) evaluated spinosad at 0, 0.1 and 1 mg/kg against *R. dominica* on 12.5% and 14.5% hard red winter wheat at temperatures of 22, 28 and 34°C over a period of four months. Each month, *R. dominica* adults were exposed for 14 days to grain at different temperature–moisture combinations to determine mortality. All *R. dominica* adults were killed on spinosad-treated wheat at the three temperatures and two moistures during the four-month test period. On untreated wheat, mortality was below 10%, except on one occasion where the mortality was 15–39%. The mortality on spinosad-treated wheat was consistently and significantly higher than on untreated wheat. These results suggest that spinosad's activity against *R. dominica*, a key pest of stored wheat, is not affected under the conditions tested.

Persistence and efficacy of spinosad residues on farm-stored wheat

Fang et al. (2002) conducted field and laboratory tests to determine persistence of spinosad residues and effectiveness of residues against *R. dominica* and *T. castaneum* adults. Samples (250 g) of untreated hard red winter wheat and wheat treated with spinosad at 0.1, 0.5, 1, 3 and 6 mg/kg were sealed in plastic mesh pouches and buried below the surface of grain stored in three farm bins located in Maniattan, Kansas. Wheat in pouches was sampled bimonthly from November 2000 to November 2001. Temperatures at all sampling locations in bins ranged from –10°C (December 2000 and January 2001) to 32°C (July 2001). Relative humidity at these same locations ranged

from 55% to 70%, and the sample moisture ranged from 12.4% to 14%.

The slope of the linear regression line of spinosad residues against storage time at each rate (between 0.1 and 6 mg/kg) was not significantly different from zero ($P > 0.05$), indicating that the residues were stable throughout the year. *R. dominica* mortality assessed in laboratory bioassays at bimonthly intervals was consistently 99–100% on spinosad-treated wheat and significantly higher ($P < 0.05$) than mortality on untreated wheat. Mortality of *T. castaneum* adults at 1 to 6 mg/kg was significantly higher ($P < 0.05$) than at 0 and 0.1 mg/kg, and was 75–100% at ≥ 3 mg/kg. The LD_{50} values for *T. castaneum* ranged from 0.3 to 1.3 mg/kg during most months. Despite a slight increase in LD_{50} with storage time, spinosad toxicity to *T. castaneum* adults was not affected during the one year of storage, as indicated by the non-significant slope of the linear regression of LD_{50} s against storage time (Fang et al., 2002).

Spinosad degrades quickly under sunlight (Brunner and Doerr, 1996; Saunders and Bret, 1997; Liu et al., 1999). However, spinosad is persistent in farm-stored grain, because the grain is protected from sunlight.

Contribution of contact toxicity and wheat condition to mortality of stored-product insects exposed to spinosad

Toews and Subramanyam (in press) designed experiments to evaluate contact toxicity of spinosad and determine the effect of wheat condition on susceptibility of stored-product insect adults and larvae to spinosad. In their tests, *R. dominica* adults were the most susceptible species when exposed to spinosad-treated glass surfaces (Petri dishes), followed by *S. oryzae* and *T. castaneum* adults. All *R. dominica* adults were killed within 48 h. Mortality of *S. oryzae* and *T. castaneum* increased with spinosad rate.

The LD_{50} values (95% CL) for *R. dominica*, *S. oryzae* and *T. castaneum* were 0.0004 (0.0003–0.0006), 0.0768 (0.0670–0.0879) and 0.1848 (0.1075–0.3212) mg/cm², respectively.

Toxicity of spinosad to *R. dominica*, *O. surinamensis* and *T. castaneum* adults and *O. surinamensis* and *T. castaneum* first instars was influenced by spinosad rate and wheat condition. Generally, insect mortality was greater on whole wheat than on cracked wheat or wheat flour. The mortality of *R. dominica* adults was $\geq 97\%$ on whole wheat at 0.1 mg/kg and at all three wheat conditions, but mortality was 42% on cracked wheat and 19% on wheat flour. Spinosad failed to kill *T. castaneum* adults at 0.1 mg/kg, but more adults were killed in whole wheat than in cracked wheat or wheat flour at 1 mg/kg. Mortality of *O. surinamensis* was greater in whole wheat than in cracked wheat or wheat flour. However, mean mortality of this species was $< 20\%$ in all treatments. The mortality of *O. surinamensis* and *T. castaneum* first instars was significantly higher ($P < 0.05$) on whole wheat than on cracked wheat and wheat flour at 1 mg/kg.

Excellent contact toxicity to some key stored-product insects indicated that spinosad could be used to treat surfaces (empty bins, warehouse floors, mill floors, cracks and crevices) for managing stored-product insects. Better performance of spinosad on whole grain suggested that using spinosad on clean grain may improve its efficacy.

Contact toxicity of spinosad applied to various surfaces

M. Toews and Bh. Subramanyam (unpublished data) exposed adults of *T. confusum*, *R. dominica*, *O. mercator*, *T. castaneum*, *C. ferrugineus*, *S. oryzae*, *O. surinamensis*, and the warehouse beetle, *Trogoderma variabile* Ballion to different surfaces, such as concrete, floor tile, steel and waxed floor tile, treated with spinosad at 0.05 and 0.1 mg/cm². Insects were exposed to untreated panels (0 mg/cm²; surface area, 30 cm²) or panels treated with spinosad for 24 h. Mortality was assessed after insect recovery on diets for an additional 24 h. Mortality of each species on spinosad-treated panels was higher than that on untreated panels (Fig. 1). Nearly all *R. dominica*, *O. mercator*, *C. ferrugineus*, *S. oryzae*, *O. surinamensis* and *T. variabile* adults were killed on spinosad-treated panels. All *T. castaneum* and *T. confusum* adults were killed when exposed to concrete panels treated with spinosad. The mortality for these two species on steel, floor tile and waxed floor tile treated with spinosad was $> 70\%$. The excellent contact toxicity against a broad range of insect species suggested that spinosad could be used as a surface or crack or crevice spray in empty grain stores or in food-processing facilities.

Conclusions

Spinosad is effective against *R. dominica* and *P. interpunctella* at 1 mg/kg on both corn and wheat, although 0.1 mg/kg was effective against the former species. The reasons for the reduced susceptibility of *S. oryzae*, *T. castaneum* and *O. surinamensis* require further study. Both the field and the laboratory tests have confirmed that spinosad's activity against *R. dominica* is unaffected by grain temperature $\leq 34^{\circ}\text{C}$ and grain moisture $\leq 15\%$. Spinosad residues in farm-stored grain are stable, and provided effective control of *R. dominica* for one year. Spinosad has excellent contact toxicity at rates < 0.1 mg/cm² against a wide variety of stored-product insects on different surfaces. Our results showed that spinosad's activity is greater on whole grain than on cracked grain or flour. Grain sanitation is therefore important to enhance spinosad's activity. The low mammalian toxicity of spinosad, its activity in killing and suppressing progeny production of various stored-product insect pests, and its persistence in farm-stored grain make it a viable replacement for existing grain protectants.

Current and future research directions

The United States Environmental Protection Agency in May 2002 granted an experimental use permit for evaluating spinosad as a potential grain protectant at the 1 mg/kg level. Field trials are currently being conducted on three Kansas farms to compare activity of spinosad at 1 mg/kg with untreated grain (0 mg/kg) and grain treated with chlorpyrifos-methyl (3 mg/kg) and spinosad + chlorpyrifos-methyl (1 + 3 mg/kg). The combination of spinosad + chlorpyrifos-methyl is aimed at controlling *T. castaneum* and *O. surinamensis*, species that are less susceptible to spinosad. Similar evaluation in shelled corn will begin on

farms in Indiana during November 2002, and this effort is being coordinated by Dr Dirk Maier, Purdue University, Indiana. On corn, pirimiphos-methyl (4 or 8 mg/kg) will be used instead of chlorpyrifos-methyl.

In a related project, the benefits of using spinosad at 1 mg/kg are being compared with using aeration alone. Our objective is to determine if spinosad is a suitable treatment for unaerated grain.

We are also evaluating the effects of spinosad on natural enemies of stored-product insects, in an attempt to identify specific parasitoids or predators that can be used in conjunction with spinosad for insect management in stored-grain ecosystems.

Information, in the form of articles, refereed papers, PowerPoint presentations and pictures, on our past and current spinosad projects is available online at www.oznet.ksu.edu/grsc_subi, under the "What's New" link.

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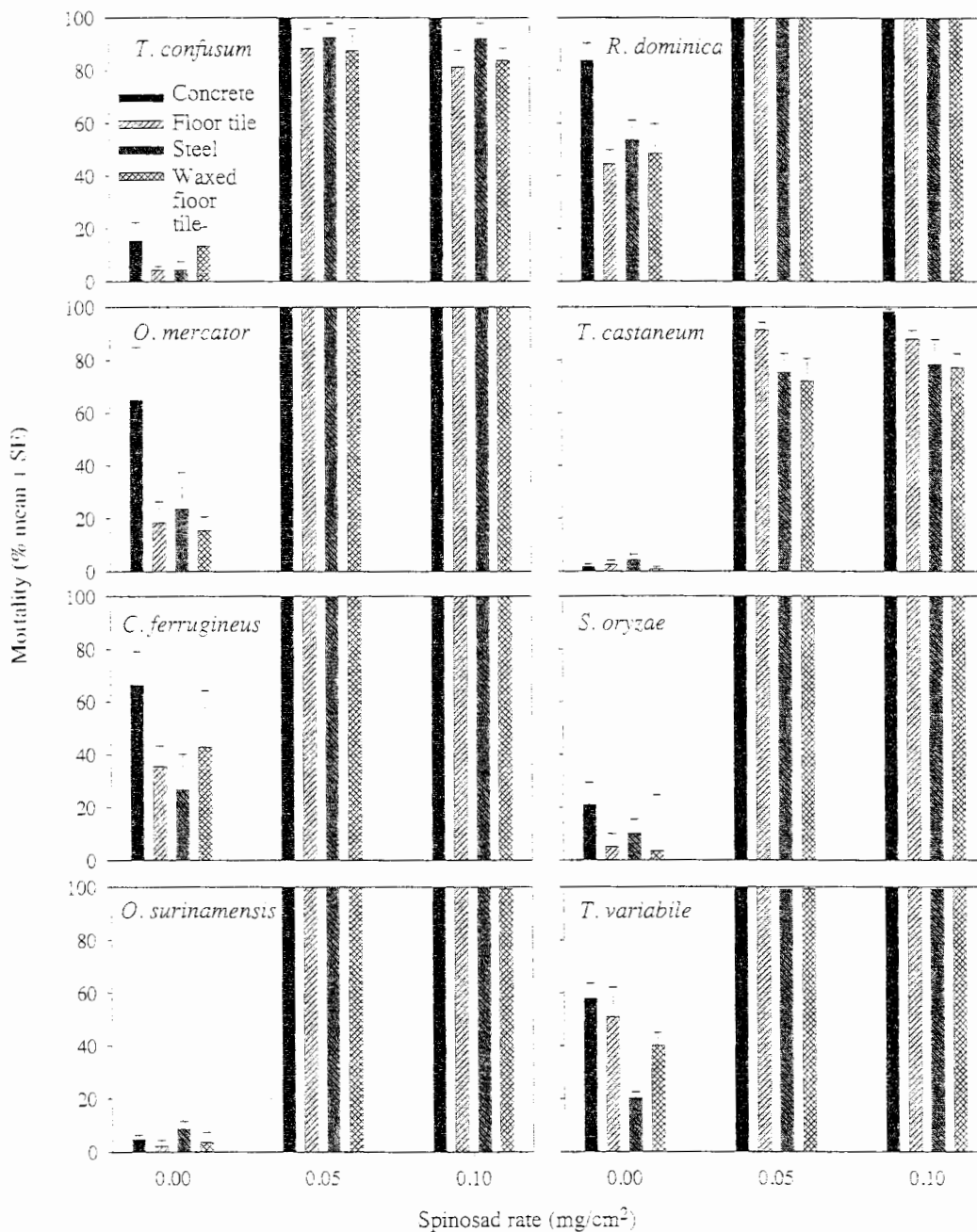


Fig. 1. Mortality of stored-product insects exposed to untreated concrete, steel, and unwaxed and waxed floor tile panels and panels treated with spinosad at 0.05 and 0.1 mg/cm². Adults were exposed for 24 h to panels and mortality assessments were made after 24-h recovery on diets.

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