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Compatibility of *Bacillus thuringiensis* and Captan When Used in a Mixture for Treating Seed Corn for Moth Control[1]

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ABSTRACT Captan (Orthocide) in aqueous suspension at 58.6 mg of AI/ml prevented germination of *Bacillus thuringiensis* Berliner (Dipel) spores but did not kill the spores. Upon dilution with water, spores that had been incubated with captan for 4 h germinated and produced colonies on agar plates. Captan had no effect on the toxicity of *B. thuringiensis* to almond moths, *Ephestia cautella* (Walker), and only a slight negative effect on toxicity to Indianmeal moths, *Plodia interpunctella* (Hübner), perhaps because of its effects on the spores to which this species is susceptible. Captan alone was slightly toxic to these insect species, but it did not increase the toxicity of *B. thuringiensis* in mixtures.

Bacillus thuringiensis Berliner has been shown to be effective for preventing moth infestations in stored grains, such as wheat and corn (McGaughey 1976, 1978b, 1980), and to a lesser extent in stored peanuts (McGaughey 1982). As a result, producers and distributors of crop seed, a high-value commodity, have shown considerable interest in using *B. thuringiensis* to protect bagged seed from moth infestation. Crop seed poses a high risk of moth infestation because it is usually bagged and frequently is stored for unpredictable time periods in warehouses that may also contain other grain products, such as animal feeds. This potential use of *B. thuringiensis*, however, has raised questions regarding the compatibility of the bacterial preparation with commonly used seed fungicides, such as captan. The compatibility of *B. thuringiensis* and captan has not been previously investigated on stored seed, and literature on their compatibility in other situations is scant. Heimpel (1967) concluded that they were compatible, that mixing was possible, and that no increased effect of *B. thuringiensis* had been observed. This paper reports results of tests made specifically to determine whether mixing captan with *B. thuringiensis* in aqueous suspensions would affect the subsequent toxicity of *B. thuringiensis* on corn seed to the Indianmeal moth, *Plodia interpunctella* (Hübner), or the almond moth, *Ephestia cautella* (Walker).

Materials and Methods

Captan and *B. thuringiensis* were mixed with water, agitated for ca. 4 h as would be likely to occur in a spray tank, and then applied to corn or laboratory insect diet to determine effects on insect toxicity, and plated to determine effects on spore viability. Orthocide 75 seed protectant containing 75% captan is normally applied to seed corn at a rate of ca. 781.25 mg/7.8 to 13.0 ml of water slurry per kg of grain. Dipel WP containing 16,000 international units of *B. thuringiensis* per mg is labeled for application at a dosage of ca. 125 mg/10 ml of water per kg. For this study the products were mixed with water individually and together at rates of 781.25 mg of captan formulation (586 mg of AI) and 125 mg of Dipel per 10 ml water, and agitated at room temperature for 4 h.

Spore viability was checked on each suspension by making serial 1:10 dilutions and spreading 0.1 ml of each dilution on half-strength nutrient agar plates and counting the number of bacterial colonies after incubation of the plates at 27°C for 48 h. Two sets of dilutions were prepared for the captan + *B. thuringiensis* suspension, one with captan suspension and one with water. The *B. thuringiensis* suspension was diluted with water.

Insect toxicity of the suspensions was determined by treating samples of corn and laboratory insect diet and infesting them with Indianmeal moths and almond moths. Seven serial 1:2 dilutions of the captan-*B. thuringiensis* mixture were prepared for treating 500-g samples of corn at a rate of 5 ml/500 g. Captan suspension was used as the diluent so that the captan dosage remained constant in the dilutions at 586 mg of AI/kg, whereas the Dipel dosage decreased from 125 to 1.95 mg/kg. Similar dilutions, but diluted 10-fold with water, were prepared and used to treat samples of laboratory insect diet so that an application rate of 6 ml/60-g sample could be used, providing captan and Dipel dosages equal to those tested on corn. Dipel suspension (without captan) was diluted in the same manner and applied to samples of corn and diet, except that water, instead of a captan suspension, was used as the diluent. Three check samples of corn and diet were treated with water or with captan suspension at 586 mg of AI/kg. The laboratory diet consisted of cracked wheat and wheat bran supplemented with wheat germ, brewer's yeast, glycerol, honey, water, and fungistatic agents. Yellow corn with an initial moisture content of ca. 14% was used.

The diluted suspensions were applied by pouring them over corn samples in 3.8-liter jars and mixed by tumbling the jars on a mechanical jar roller until all the moisture was absorbed by the grain (ca.. 5 min). Diet samples were treated similarly, but were mixed with a motor-driven polyethylene stirrer in a small bowl. After treatment, samples were divided into two mason jars and infested with 50 eggs of either the Indianmeal moth or the almond moth obtained from laboratory colonies. The

[1]Mention of a proprietary product does not constitute an endorsement by the USDA. Received for publication 22 October 1982; accepted 4 April 1983.

Table 1. Viable spores in suspensions of *B. thuringiensis* and captan[a]

Suspension	Spores/mg of Dipel
B. thuringiensis + captan, aged 4 h, then diluted with water for plating	14.6×10^7
B. thuringiensis in water, aged 4 h before plating	3.5×10^7
B. thuringiensis + captan, aged 4 h, then diluted with captan suspension for plating	No colonies

[a]58.6 mg (AI) of captan per ml of water; 5 mg of Dipel per ml of captan suspension or water.

Table 2. Toxicity of *B. thuringiensis* to Indianmeal moths and almond moths on laboratory diet and corn when applied alone and in a mixture with captan at 586 mg (AI)/kg

Moth	Medium	Treatment	LC_{50} (mg/kg)	95% CI (mg/kg)
Indianmeal	Diet	With captan	24.3	27.4–21.6
		Without captan	16.3	18.5–14.3
	Corn	With captan	4.7	9.9– 0.9
		Without captan	5.5	9.9– 2.0
Almond	Diet	With captan	20.6	24.1–17.6
		Without captan	20.0	27.9–14.4
	Corn	With captan	9.2	16.5– 4.4
		Without captan	5.2	11.2– 1.1

infested samples were held at 25°C and 65% relative humidity (RH), where the corn equilibrated to ca. 13% moisture content and the diet to about 20%. Mortality was calculated based on the number of F_1 adults that emerged from each sample. The experiment was done twice, and the data were pooled to calculate LC_{50} values and 95% confidence intervals (CIs) for *B. thuringiensis* alone and with captan.

Results and Discussion

The effects of captan on *B. thuringiensis* spore viability are summarized in Table 1. No colonies were produced on plates where a high concentration of captan (58.6 mg of AI/ml) was maintained by preparing dilutions with captan suspension. However, colonies did grow when the suspensions were diluted ≥100 times with water. Apparently, high concentrations of captan prevent germination but do not kill spores. Furthermore, plate counts indicate that, after being agitated at room temperature for 4 h, more viable spores were present in the *B. thuringiensis*-captan mixture than in the *B. thuringiensis* suspension lacking captan. Captan may have effectively stabilized the *B. thuringiensis* suspension by preventing spore germination in the aqueous suspension.

Toxicity data for *B. thuringiensis* with and without captan are summarized in Table 2. Captan had little or no effect on the toxicity of *B. thuringiensis* to almond moths in diet or corn, or to Indianmeal moths in corn. Confidence intervals for the LC_{50} values with and without captan overlap for these bioassays. Captan did appear to reduce slightly the toxicity of *B. thuringiensis* to Indianmeal moths in the diet bioassays. This may have resulted because captan prevents spore germination. Spores have an important role, perhaps synergistic, in the toxicity of *B. thuringiensis* to Indianmeal moths (McGaughey 1978a).

Table 3. Mortality of Indianmeal moths and almond moths in check samples of laboratory diet and corn and in samples treated with captan at 586 mg (AI)/kg[a]

Insect	Treatment	% Mortality in: Diet	Corn
Indianmeal moth	With captan	13.3	61.4[b]
	Without captan	11.7	20.4
Almond moth	With captan	21.6	60.6[c]
	Without captan	21.6	37.7

[a]Means of six samples, with 50 insects per sample.
[b,c]Mortalities in corn samples with captan were significantly greater than in the respective samples without captan at the 1% (*b*) or 2.5% (*c*) level (Student's *t* test).

The effects of captan alone on Indianmeal moths and almond moths are summarized in Table 3. Captan significantly increased the mortality of both insect species on corn but not on the laboratory diet. Evidently its very low level of insecticidal activity is more pronounced when the insects are exposed under suboptimal conditions. In grains such as corn we have consistently observed slower larval development and higher mortality levels than in the laboratory diet. Therefore, in the Indianmeal moth bioassays in corn, captan's toxicity apparently compensated for its negative effects on *B. thuringiensis*, but it did not do so in the diet (Table 2). The fact that the mixture was not more toxic than *B. thuringiensis* alone indicates that the same individuals in the insect populations, probably the less vigorous ones, are susceptible to both materials.

These data indicate that *B. thuringiensis* and captan can be applied together to corn seed in an aqueous suspension without noticeable detrimental effects on control of Indianmeal moths or almond moths in the stored seed.

REFERENCES CITED

Heimpel, A. M. 1967. A critical review of *Bacillus thuringiensis* var. *thuringiensis* Berliner and other crystalliferous bacteria. Annu. Rev. Entomol. 12: 287–322.

McGaughey, W. H. 1976. *Bacillus thuringiensis* for controlling three species of moths in stored grain. Can. Entomol. 108: 105–112.

1978a. Response of *Plodia interpunctella* and *Ephestia cautella* larvae to spores and parasporal crystals of *Bacillus thuringiensis*. J. Econ. Entomol. 71: 687–688.

1978b. Moth control in stored grain: efficacy of *Bacillus thuringiensis* on corn and method of evaluation using small bins. Ibid. 71: 835–839.

1980. *Bacillus thuringiensis* for moth control in stored wheat. Can. Entomol. 112: 327–331.

1982. Evaluation of commercial formulations of *Bacillus thuringiensis* for control of the Indianmeal moth and almond moth (Lepidoptera: Pyralidae) in stored inshell peanuts. J. Econ. Entomol. 75: 754–757.