EVALUATION OF AVERMECTIN B_1 (MK 936) AGAINST HELIOTHIS SPP.[1/] IN THE LABORATORY AND IN FIELD PLOTS AND AGAINST THE BOLL WEEVIL[2/] IN FIELD PLOTS[3/]

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ABSTRACT

The activity of avermectin B_1 (MK 936) was assessed in the laboratory, by exposure of 1st stage larvae of *Heliothis virescens* (F.) and *H. zea* (Boddie) to treated cotton seedlings. In addition, larvae (280-300 mg) of *H. virescens* were topically treated with acetone and aqueous solutions of avermectin and contact activity was determined with residues on glass petri dishes. The LC_{50} for the 1st stage larvae for both species was 0.00002% whereas for *H. virescens*, the LC_{95} = 0.00052%, and for *H. zea*, was 0.00234%. Mortality responses for the topical applications on *H. virescens* (avg. wt. 306 mg) was LD_{50} = 0.0154 µg and LD_{95} = 0.1583 µg/larva for acetone and LD_{50} = 0.0024 µg and LD_{95} = 0.172 µg/larva for aqueous solutions. In field evaluations crop yields were significantly greater in the avermectin-treated plots that were infested either artificially or naturally with *H. virescens*. Oviposition damage in plots naturally infested with high populations of the boll weevil, *Anthonomus grandis* Boheman, also was significantly lowered by avermectin treatment.

INTRODUCTION

The avermectins are macrocyclic lactones naturally produced by the soil microorganism, *Streptomyces avermitilis*. These structurally-related products have a broad spectrum of activity against mites, insects, and plant parasitic nematodes (Putter et al. 1981). Studies on the mode of action of the avermectins suggest that the transmission of signals from interneurons to excitatory motoneurons are blocked by the interference with the neurotransmitter, gamma-aminobutyric acid (Fritz et al. 1979, Kass et al. 1980). The result of this activity is that insects are quickly immobilized and eventually paralyzed leading to death as reported by Wright (1984) in studies with the boll weevil, *Anthonomus grandis* Boheman. Applications of avermectin significantly reduced feeding activity by the alfalfa weevil, *Hypera postica* (Gyllenhal), and in some instances deterred feeding (Pienkowski and Mehring 1983). Schuster and Everett (1983) reported that avermectin reduced egg deposition and egg hatch in *Liriomyza trifolii* (Burgess) when females were exposed to tomato leaflets dipped in an aqueous solution. Glancey et al. (1982) demonstrated that avermectin B_1a caused irreversible damage in the ovaries of the imported fire ant, *Solenopsis invicta* Buren. In this report we discuss experiments to assess the relative activity of MK 936 against the tobacco budworm, *Heliothis virescens* (F.), in the

1/ Lepidoptera: Noctuidae.
2/ Coleoptera: Curculionidae.
3/ Report of research results only. Mention of a proprietary product does not constitute a recommendation or an endorsement by USDA.

laboratory and in small plots, and against the boll weevil in small plots. Activity against 1st stage larvae of the bollworm, Heliothis zea (Boddie), was determined in laboratory studies.

MATERIALS AND METHODS

Avermectin B_1 (MK 936) was obtained as a technical grade product and as a 0.15 EC formulation from Merck, Sharp and Dohme Research Laboratories, Rahway, NJ. The MK 936 formulation (hereafter called avermectin) is a mixture containing ca. 80% avermectin B_1a and 20% avermectin B_1b. Cottonseed oil (hexane extracted) was obtained fresh and was stored at 6°C. The tobacco budworm (TBW) and bollworm (BW) strains were from the Southern Field Crop Insect Management Laboratory, ARS, USDA, at Stoneville, MS.

Laboratory Tests. Cotton seedlings (Stoneville 213) at the four leaf stage were sprayed using a Preval Power Unit®. The 0.15 EC formulation of avermectin was diluted in water, sprayed onto the leaf surface, and allowed to dry for 3 h. First stage TBW or BW larvae (10/each) were placed on the leaf surface and enclosed within a 2.54 cm square plastic box. Ten plants used for each treatment level and the experiment was replicated six times. The following concentrations (% w/v) were evaluated: 0.5, 0.05, 0.005, 0.0005, 0.00005, and 0.000005. Seedlings sprayed only with water served as controls. Microscopic observations of the larvae on the seedlings were made at 24 h intervals.

In other tests, TBW larvae (avg. wt. 280 mg) were confined for 1 h in 100 mm petri dishes coated with the residue left by the evaporation of an acetone solution of the technical avermectin. The rates of application were 6.3, 2.8, 0.63, and 0.28 μg/dish. Following exposure to these residues, larvae were placed on untreated diet and observed for development.

In addition, TBW larvae (avg. wt. 306 mg, 10/treatment) were topically treated on the dorsal surface of the abdomen with 1 μl of either an acetone solution of the technical avermectin or an aqueous solution of the 0.15 EC formulation, and then held individually in diet cups where subsequent development was monitored. Controls were treated with acetone or water only. Data for the efficacy of avermectin against the TBW and BW first stage larvae were analyzed by the probit procedure of Finney (1971).

Field Plots. The cultivar Stoneville 213 was planted in 12.8 m single row plots -- in a two and one skip row pattern in area one, and in single row plots spaced 1 m apart in area two. Four replications were used in area one and eight replications in area two. First stage TBW larvae (12/terminal area of each plant) were applied weekly beginning at 2 wks prior to treatment and then during treatment for 3 wks, using the method of Jenkins et al. (1982) with the equipment developed by Davis and Oswalt (1979). Natural infestations of TBW were also present in the cotton acreage. The rate of application of avermectin was 28.3 g/ha in a total volume of 22.7 L/ha. The total volume included 3.78 L of cottonseed oil that was emulsified with Tween 80® and Span 80®. The material was applied with a backpack CO_2-powered sprayer; six treatments at 7 day intervals were made in each area. Plots were harvested and the yield of seed cotton was determined to evaluate the efficacy of avermectin applications.

Cotton plots with heavy boll weevil infestation (ca. 40% oviposition damage) were treated semiweekly with avermectin at a rate of 28.3 g/ha. Four 0.1-ha plots were used; two were treated and two served as untreated control plots. Application was from a tractor-mounted spray rig with a 12 nozzle boom (no. 1 nozzles). Six treatments were made in experiment 1 and nine treatments were made in experiment 2. Oviposition damage (%) to 100 squares/plot/sampling date was the criterion used for evaluation of avermectin.

RESULTS AND DISCUSSION

In preliminary evaluations, 1st stage larvae (TBW) were placed on treated cotton seedlings and were not restricted in their movement. The larvae left the seedlings and no feeding was observed to occur in either the controls or the treatments. Subsequently, larvae were confined within 2.54 cm plastic boxes and mortality and feeding activity were easily determined with a dissecting microscope. The first visible symptom of avermectin against the 1st stage larvae was paralysis at the 24 h observation; eventually death occurred with the larva being in a swollen and elongated condition similar to that observed in a lepidopteran larva dying from viral infection. Mortality of the TBW larvae did not differ from the BW larvae at the LD_{50} level (Table 1). Limited feeding activity was observed at the upper dose levels, whereas untreated larvae fed voraciously on the young leaves.

TABLE 1. Comparative Mortality Response of 1st Stage Larvae of H. virescens and H. zea when Placed on Cotton Seedlings Treated with Avermectin.[a]

	LC_{50}	LC_{95}	Slope (±SE)[b]
H. virescens	0.00002(.00001-.00003)	0.00052(.0002-.001)	1.219(.181)
H. zea	0.00002(.00001-.00014)	0.00234(.001-1.019)	0.667(.128)

a/ LC_{50} and LC_{95} (% solution of avermectin) applied to cotton seedlings as an aqueous spray. Values for 95% confidence limits are given in parentheses.
b/ Slope of the regression between dose and mortality, with standard error (SE) given in parentheses.

Analyses of data for mortality of TBW larvae (avg. wt. 280 mg) exposed for 1 h to residues of avermectin on glass petri dishes indicated a nonsignificant regression at the levels tested. The results for the contact activity were:

dose (μg/dish)	% mortality
6.3	100
2.8	80
0.63	60
0.28	0
0	0

The onset of mortality at the 6.3 μg treatment level was observed within 24 h. Indications of delayed neurotoxicity, such as the failure of larvae to pupate or failure of pupae to eclose, were observed at the other dosage levels.

The topical LD_{95} of avermectin-treated TBW larvae (avg. wt. 300 mg) was similar whether applied topically in an aqueous solution or in an acetone solution. The aqueous solution was more active at the LD_{50} level (Table 2). Similar to the results obtained with the contact evaluation, delayed mortality was observed in the larvae and pupae in the test.

Field Plots. The field evaluation of avermectin as a spray for TBW larvae showed it to be effective in area 1 where the test plots were in a two and one skip row configuration. Treated cotton produced significantly more (1887 kg/ha) seed cotton than the untreated control (1086 kg/ha) (Table 3). The treatment in area 2 did not result in increased yields; apparently the oil and the environment interacted and defoliated treated plots (Table 3). This area was on an upland slope and was drier and thus the cotton matured earlier than in area 1 and treatments were begun after peak boll load in these plots.

TABLE 2. Mortality Response of *H. virescens* Larvae (avg. wt. 306 mg) to Avermectin when Applied Topically in Acetone Solution or an Aqueous (EC) Suspension.[a]

Application mode	LD_{50}	LD_{95}	Slope (SE)[b]
Acetone	0.0154(.0352-.0035)	0.1583(3.216-.0613)	1.629(.503)
Aqueous	0.0024(.0076-.0003)	0.1720(6.03-.0427)	0.894(.234)

a/ LD_{50} and LD_{95} of avermectin in µg. Values for 95% confidence limits are given in parentheses.
b/ Slope of the regression between dose and mortality, with standard error (SE) given in parentheses.

TABLE 3. Cotton Harvested from Plots Treated with Avermectin (0.028 kg/ha) as a Spray Against *H. virescens*.

Reps	Kg seed cotton/ha: Treated	Control
	Experiment 1	
1	1835	803
2	1776	605
3	1457	1235
4	1599	1229
$\bar{X}$	1667[a]	968
	Experiment 2	
1	2494	2438
2	1864	2335
3	2057	2643
4	2687	2694
5	1564	2164
6	2254	2090
7	2378	2711
8	2313	2504
$\bar{X}$	2201[b]	2447

a/Means are significantly different at 0.01 level.
b/Means are significantly different at 0.05 level.

Of the two experiments conducted against the boll weevil, one was terminated after 21 days and the other was terminated after 42 days. There was a clear difference between the levels of oviposition damage in each of the test plots as indicated by data illustrated in Figs. 1a and 1b. After treatment was discontinued in test plot 2 at 30 days, the oviposition damage began to increase. Adult weevils were collected from the treated plots and observations in the laboratory confirmed the results reported by Wright (1984) in that paralysis developed in these adults, their abdomens became swollen and distended beyond the elytra, and then death occurred. In the treated plots we observed a considerable reduction of other insect activity when compared to the controls.

Our laboratory tests showed that avermectin is highly effective against TBW and BW larvae in the 1st instar, as well as against larger TBW larvae. Laboratory evaluations against the boll weevil by Wright (1984) indicated that avermectin significantly inhibited pheromone production of adult weevils before delayed neurotoxicity occurred. In our field evaluations, crop yield was significantly increased by avermectin treatment of TBW infested plots. Additionally, late season applications of avermectin were effective in suppressing very heavy infestations of native boll weevils.

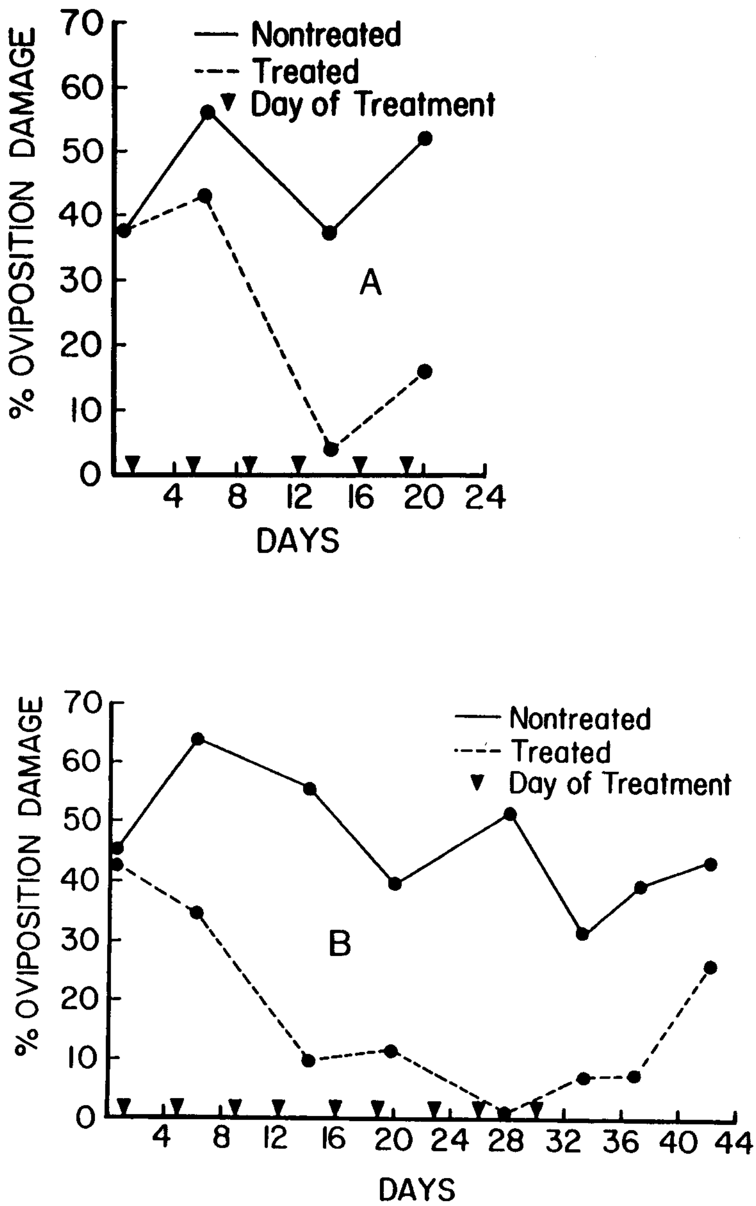

FIG. 1. Comparison of oviposition damage by the boll weevil in avermectin-treated and untreated cotton plots in two experiments.

Although avermectin has been evaluated extensively against many insects and has been shown to possess a broad spectrum of activity against mites, insects, and nematodes (Putter et al. 1981) our research yielded data that indicates high activity against two of the more important insect pests in cotton. Treatments with avermectin in the laboratory and field were also evaluated for possible insect growth regulator effects. A different time interval was used because of the unique mechanism of action of avermectin that involves its interference with gamma-aminobutyric acid in neuromuscular transmission sites (Fritz et al. 1979, Kass et al. 1980).

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