CONTROL OF BOLLWORMS IN BT COTTON USING GEMSTAR LC BIOLOGICAL INSECTICIDE

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Abstract

Results of a study in 1999 were reported that assessed the effect of B.t.-transgenic cotton on the cotton bollworm-*Helicoverpa zea* nucleopolyhedrovirus interaction, and evaluated the effectiveness of lower rates of Gemstar against cotton bollworms on B.t.-transgenic cotton. The B.t.-treated diet used in the laboratory study reduced the growth rate of cotton bollworms and caused an additive mortality response when combined with Gemstar. Larval counts of *H. zea* in field studies were significantly lower on B.t. cotton for all treatments, while *H. zea* larvae in all virus treatments showed lower survival.

Introduction

Since 1996, when commercial B.t.-transgenic cotton varieties were first introduced, the planted acreage has steadily increased each year. In 1998, approximately 55% of the cotton acreage planted in Mississippi were B.t.-transgenic varieties (Layton et. al., 1999). Adoption of the B.t.-transgenic cotton technology by growers will most likely continue to increase in Mississippi as the Boll Weevil Eradication Program is implemented in the Delta. As this technology becomes more widespread, the potential for tolerance to B.t. in target pest species will be a critical issue. Already recent studies have shown that the cotton bollworm, *Helicoverpa zea* Boddie may survive on B.t.-transgenic cotton (Meyers et al., 1997).

Foliar sprays that would control cotton bollworm and allow for the preservation of beneficial insects would be useful. Gemstar LC is a microbial insecticide used to control Heliothine species on several major crops, including cotton (Dimock, 1997). The active ingredient is the *Helicoverpa zea* nucleopolyhedrovirus (HzSNPV), a naturally-occurring pathogen originally isolated from the cotton bollworm. Gemstar is recommended for use against heliothine species on conventional cotton prior to flowering. However, potential B.t./virus interactions may make foliar applications of Gemstar more effective at controlling cotton bollworms on B.t. cotton.

Foliar applications of a mixture of *Bacillus thuringiensis* and the *Autographa californica* nucleopolyhedrovirus (AcMNPV) have been shown to control tobacco budworm, *Heliothis virescens* (F.) populations on cotton (Bell and Romine, 1980). Young et al. (1980) reported additive mortality among 3rd instar cabbage looper, *Trichoplusia ni* (Hübner) treated with B.t. and high concentrations of nucleopolyhedrovirus. Furthermore, a synergistic response was reported for mixtures of a low level of B.t. endotoxin and the *Helicoverpa armigera* nucleopolyhedrovirus (HaMNPV) against *H. zea* (Bell and Romine, 1986).

The objective of this study was to examine the effect of B.t. cotton on the cotton bollworm-HzSNPV interaction and to determine whether lower rates of Gemstar might be effective against cotton bollworms on B.t.-transgenic cotton.

Materials and Methods

Laboratory

Gemstar LC was used to prepare HzSNPV suspensions and virus concentrations were determined by making direct counts with a hemacytometer. Each virus suspension was applied to diet discs (5mm dia., 1mm thick) in ten-microliter amounts to produce virus concentrations of 1×10^5, 1×10^4, and 1×10^3 occlusion bodies per larva. Control larvae were fed untreated diet discs.

Control and virus-contaminated diet discs for each treatment group were individually fed to approximately eighty second-instar *H. zea* larvae that had been starved overnight. The first 40 larvae in each treatment group that consumed the entire diet disc were divided equally into two subgroups, and transferred individually to either untreated or B.t.-treated diet cups. Untreated diet consisted of the standard soyflour diet, whereas the B.t.-treated diet was prepared using the MVP II bioinsecticide (Mycogen Corp.) with the delta endotoxin of *Bacillus thuringiensis* variety kurstaki. The MVPII-treated diet consisted of sufficient MVP II added to 300 ml of molten diet to achieve 500 ng delta endotoxin per ml diet, and then blended for three minutes. Five mls of the final mixture was added to 40 one oz. cups and allowed to harden prior to infesting with insects. Larvae were reared at 26EC, and examined daily for mortality until the experiment was terminated after six days. The mean mortality time for H. zea exposed to various virus concentrations and maintained on control or Bt-treated diet were separated using a means-Tukey statement with the GLM procedure (SAS Institute, 1989).

Field

All tests were conducted on the B.t.-transgenic cotton variety DeltaPine 33B and the conventional cotton variety, Stoneville

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BXN 47. Treatments were applied by ground application at 6 gal/ acre . The recommended rate for Gemstar LC on cotton is 10 fl. oz. per acre (750 ml per hectare). In this study, HzSNPV (Gemstar™ LC Thermo Trilogy, Inc.) was applied at rates of 10 fl. oz., 5 fl. oz., and 2.5 fl. oz. per acre.

Each of the virus formulations and the untreated control were replicated six times in plots that were 4 rows (13.3 ft.) X 125 ft in length. Twelve plant sleeves (8 in.X19 in X 22 in) were randomly placed on individual cotton plants in the center two rows of each plot. Six -four-day-old cotton bollworms were added to each plant sleeve and sealed to prevent escape. Six plant sleeves were removed on day 3 (July 26th) and day 4 (July 27th) post-application and the number of dead, missing, and live larvae recorded. The presence or absence of arthropod predators for each cage was also recorded. All live larvae were reared at 26EC on insect diet and examined for virus infection after 14 days. The mean number of *H. zea* that were dead, missing and alive after exposure to the various virus treatments on conventional and B.t.-transgenic cotton were analyzed after removing predator-infested cages using the GLM procedure, and the means were separated by LSD (P=0.05) (SAS Institute, 1989).

Results and Discussion

Results from the laboratory study are shown in Figure 1. Exposure of *H. zea* to diet containing 500 ng endotoxin/ ml diet resulted in an average mortality of 5%, whereas larvae fed on untreated diet had 0% average mortality. The endotoxin caused limited feeding that reduced the *H. zea* growth rate, and caused the *H. zea* reared on the B.t.-treated diet to be stunted in comparison to *H. zea* larvae reared on untreated diet. Among the live larvae at the termination of the study, the average weight of uninfected larvae was 0.422 ±0.09gm on standard soyflour diet, whereas uninfected larvae had an average weight of 0.041±0.01 gm on B.t.-treated diet(data not shown). This represents nearly a 10-fold difference in weight. The HzSNPV fed to 4-day-old *H. zea* larvae produced a dose dependent mortality response. An additive mortality response was observed for early instar larvae of *H. zea* exposed to HzSNPV and B.t. endotoxin. These results are comparable to those reported by Young et al. (1980) for cabbage loopers exposed to B.t. and virus. In contrast, Luttrell et al. (1982) in a laboratory study did not observe any increase in heliothine mortality from combinations of virus and B.t. These observed differences reported in the literature may be due to the use of different viruses and B.t. formulations, variations in the B.t. and virus concentration, and the age of the insect.

Results from the field study for July 26th and July 27th are given in Table 1 and Table 2, respectively. The number of live insects were low and significantly reduced in all treatments on B.t. versus non-B.t. cotton. No significant differences were found in the number of live larvae among the virus treatments. However, the mean number of live insects decreased as the virus application rate increased. The infection rate among live insects on the non-B.t. cotton increased as the virus rate increased, whereas the infection rate among live insects on B.t. cotton did not differ significantly. Dead larval numbers for virus rates on B.t. and non-B.t. cotton showed a similar trend. Surprisingly, live larvae found on B.t. cotton for all virus application rates had a similar virus infection level.

Bell and Romine (1980) reported the highest yields of seed cotton in plots treated with a mixture of the *Autographa californica* nucleopolyhedrosis virus (AcMNPV) and B.t. with an adjuvant. Bell and Romine (1986) reported a synergistic response for *H. zea* exposed to a low level of B.t. endotoxin and the *Helicoverpa armigera* nucleopolyhedrovirus (HaMNPV). A similar phenomenom has been observed for some chemical insecticides against cotton bollworms on B.t. cotton. Laboratory studies have demonstrated that insects with a prior exposure to B.t. are more sensitive to a subsequent Karate spray treatment (Harris et. a., 1998). Brickle et al. (1999) also reported that several insecticides were highly effective at lower rates on B.t. cotton.

Conclusion

Laboratory results showed an additive mortality response for cotton bollworms exposed to virus and B.t. endotoxin. The mean number of live insects in the plant sleeves decreased as the virus application rate increased. However, some caution should be used in extrapolating these results to the field since larvae would not be restricted to a plant sleeve after virus application. Further field studies are needed to assess the impact of virus application rates on field populations of cotton bollworms in B.t. cotton.

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Disclaimer

Mention of a proprietary product does not constitute an endorsement by the USDA.

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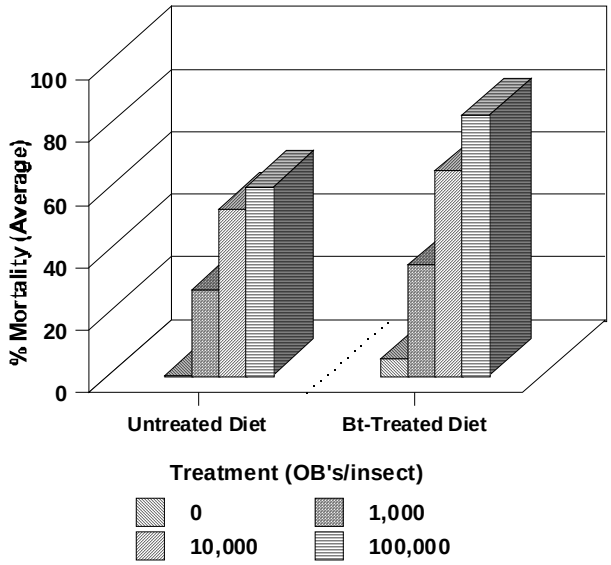

Figure 1. Effect of B.t. and Virus on the Mortality Response of Cotton Bollworm.

Table 1. Effect of Gemstar on the Interaction of Cotton Bollworm and B.t. Cotton (July 26, 1999).

	Mean (%./Plant Sleeve)[1]					
	Non-Bt			Bt		
Treatment	Live	Dead	Infected	Live	Dead	Infected
Check	30[a]	35[a]	4[a]	13[a]	42[a]	7.5[a]
2.5 oz/A	35[a]	41[a]	38[b]	12[a]	57[a]	21[b]
5.0 oz/A	15[a]	51[a]	63[b]	9.5[a]	51[a]	25[b]
10 oz/A	16[a]	60[a]	80[b]	3[a]	60[a]	22[b]

[1]Means within a column followed by the same letter are not significantly different (*P*=0.05).

Table 2. Effect of Gemstar on the Interaction of Cotton Bollworm and B.t. Cotton (July 27, 1999).

	Mean (%./Plant Sleeve)[1]					
	Non-Bt			Bt		
Treatment	Live	Dead	Infected	Live	Dead	Infected
Check	27[a]	52[a]	16[a]	8[a]	50[a]	5[a]
2.5 oz/A	23[a]	43[a]	52[b]	6[a]	49[a]	35[b]
5.0 oz/A	18[a]	57[a]	61[b]	7[a]	58[a]	33[b]
10 oz/A	17[a]	66[a]	79[b]	4[a]	57[a]	33[b]

[1] Means within a column followed by the same letter are not significantly different (*P*=0.05).