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EVALUATION OF DOW AGROSCIENCES' CRY1Ac/CRY1F TRAIT FOR IMPROVED LEPIDOPTERAN CONTROL

John J. Adamczyk, Jr. and Jeff Gore
USDA, ARS, SIMRU
Stoneville, MS
John Pellow
Dow AgroSciences
Corcoran, CA

Abstract

In 2001 and 2002, cotton lines containing Cry1Ac, Cry1F, and Cry1F stacked with Cry1Ac were evaluated in field plots and laboratory bioassays for efficacy against various lepidopteran pests. Cotton containing Cry1F stacked with Cry1Ac provided acceptable control of many lepidopteran pests of cotton in the Mid-South.

Introduction

Transgenic cotton containing a single Cry protein from the soil bacterium, *Bacillus thuringiensis* has been commercially available since 1996 in the United States. These cottons provide excellent control against various lepidopterous pests. Future transgenic cottons designed to control Lepidoptera will contain multiple Cry proteins to improve efficacy and delay the spread of resistance. In 2001 and 2002, experimental transgenic cotton lines containing Cry1Ac, Cry1F, and Cry1F stacked with Cry1Ac (Dow Agrosciences) were evaluated in field plots in the Mississippi Delta against various lepidopteran pests. In addition, laboratory bioassays for efficacy against various lepidopteran pests were conducted at the USDA, ARS, Southern Insect Management Research Unit.

Methods and Materials

Plots

Lines of experimental transgenic cottons were planted near Stoneville, MS on May 23, 2001 and May 13, 2002 (Table 1). Plots consisted of 2 rows (1.0 m centers) X 10.67 m (2001) or 4 rows (1.0 m centers) X 12.20 m (2002) arranged in a randomized complete block design (4 replications). Plots were irrigated twice in 2001 and once in 2002.

Field Efficacy

2001. The primary objective was to determine the efficacy of various transgenic cottons against the beet armyworm under field conditions. Because a natural infestation of beet armyworms never occurred, plots were inoculated with beet armyworm egg masses (Plate 1). Eight days after inoculations (DAI), beet armyworm populations were estimated using a standard 1.2 m drop cloth (5 drops/row/plot). Larvae were weighed to determine if detrimental effects on development had occurred.

2002. The primary objective was to determine the efficacy of various transgenic cotton lines against noctuids under field conditions. As in 2001, a natural infestation of beet armyworms never occurred; therefore, plots were inoculated with beet armyworm egg masses (Plate 1). Eight days after inoculation (DAI), beet armyworm populations were estimated using a standard 1.2 m drop cloth (3 drops/row/plot).

A natural population of bollworms occurred after local corn began to senesce. On July 22, 2002, larval populations were estimated by randomly examining 25 terminals and 25 squares per plot.

A late population of soybean loopers occurred in early September 2002. Larval populations were estimated using a standard 1.2 m drop cloth (3 drops/row/plot).

Laboratory Bioassay

Field plots of velvetleaf were grown near Stoneville, MS to attract tobacco budworms. Eggs were collected in mid-June, 2002 and transported to the laboratory. A colony was established an the F_1 generation used in all bioassays.

For all lines, terminal leaves were individually placed into Petri dishes (Falcon). Each dish was inoculated with either a single neonate or a single second instar. Mortality was determined at 3 days after exposure (DAE) and 5 DAE.

Damage and Yield

To determine fruit damage by bollworms, 50 squares and 50 bolls were randomly examined per plot on July 30, 2002. Seedcotton yields were measured by hand harvesting 4.0 m of row from each plot.

Statistics
All data were analyzed using REML-ANOVA and means separated using LSD.

Results and Discussion

Beet Armyworms
In 2001, cotton containing either Cry1F alone or stacked with Cry1Ac significantly reduced larval populations compared to the non-transgenic control (Figure 1). However, there were no significant differences in larval populations of beet armyworms on cotton containing only Cry1Ac compared to the non-transgenic control. In 2002, populations never became well established following inoculation. Consequently, no differences in the numbers of larvae were observed among the cotton lines. In addition, larval weights indicated that all Cry proteins had a detrimental effect on larval development (Table 2).

Bollworms
Cotton lines containing Cry1Ac alone or stacked with Cry1F significantly reduced larval populations compared to the non-transgenic control (Figure 2). However, there were no significant differences in larval populations of bollworms on cotton containing only Cry1F compared to the non-transgenic control.

Soybean Loopers
Cotton lines containing Cry1F alone or stacked with Cry1Ac significantly reduced larval populations compared to the non-transgenic control (Figure 3). However, there were no significant differences in larval populations of soybean loopers on cotton containing only Cry1Ac compared to the non-transgenic control.

Tobacco Budworms
In laboratory bioassays, all transgenic cotton lines had significantly higher mortality compared to the non-transgenic control (Figure 4).

Damage and Yield
In 2002, all transgenic cotton lines had significantly fewer damaged squares and bolls, and significantly higher seedcotton weight compared to the non-transgenic control (Figures 5 and 6). Furthermore, cotton containing Cry1Ac alone or stacked with Cry1F had significantly higher seedcotton weight compared to cotton containing only Cry1F (Figure 6).

Acknowledgments

The authors would like to thank the numerous student interns that assisted in field maintenance and bioassays. Special thanks to Larry C. Adams, Don Hubbard, and O. Houston.

Disclaimer

Mention of a commercial or propriety product does not constitute an endorsement by the U.S. Department of Agriculture for its use.

Table 1. Experimental Transgenic Cotton Lines Evaluated in 2001 and 2002.

Lines	Cry Genes
MXB-7	1Ac
MXB-9	1F
MXB-13	1Ac / 1F
Non-Bt	None

Table 2. Weights of Beet Armyworm Larvae from Plots of Transgenic Cotton Lines in 2001.

Lines	N [1]	Mean Wt (mg) ± SE
Cry1Ac	114	6.09 ± 0.385 b
Cry1F	19	3.39 ± 0.451 c
Cry1Ac / Cry1F	21	2.33 ± 0.399 d
Non-Bt	94	18.23 ± 1.476 a

[1]No. of larvae weighed.
Means were log-transformed prior to analysis with REML-ANOVA and separated using LSD (PROC MIXED). Means followed by the same letter are not significantly different ($\alpha = 0.05$).

Inoculation dates	No. egg masses/plot
July 10, 2001	12
July 11, 2001	12
July 12, 2001	18
Aug. 1, 2002	26
Aug. 2, 2002	30

Plate 1. Inoculation of Beet Armyworm Egg Masses

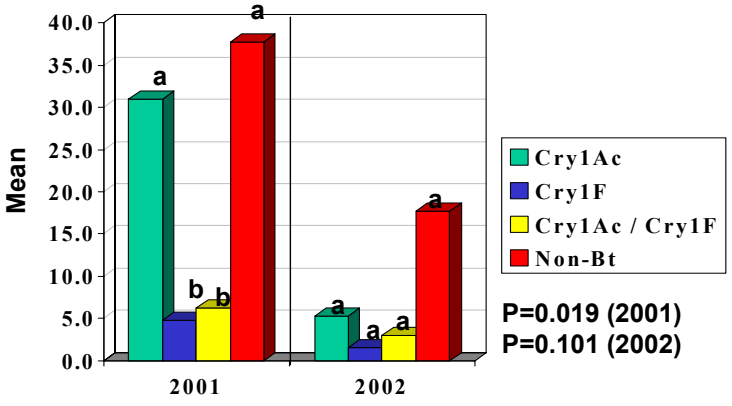

Figure 1. Mean number of beet armyworms per plot during 2001 and 2002. Means followed by the same letter are not significantly different (REML – ANOVA; LSD, $\alpha = 0.05$).

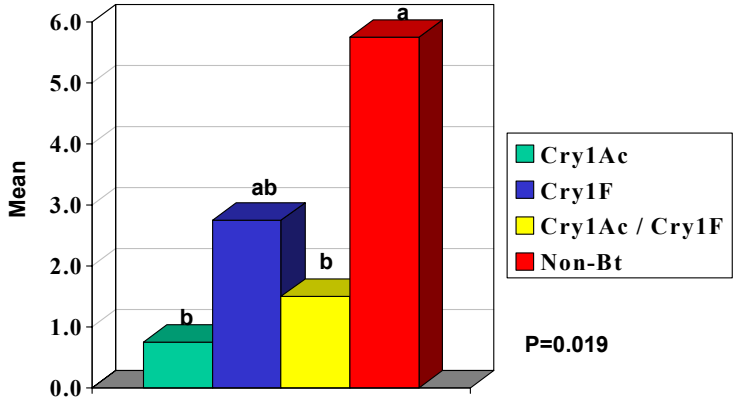

Figure 2. Mean number of bollworms per plot in 2002. Means followed by the same letter are not significantly different (REML – ANOVA; LSD, $\alpha = 0.05$).

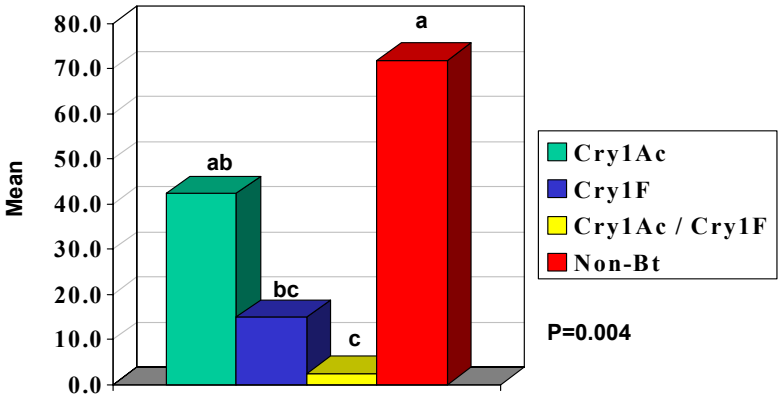

Figure 3. Mean number of soybean loopers per plot during 2002. Means followed by the same letter are not significantly different (REML – ANOVA; LSD, $\alpha = 0.05$).

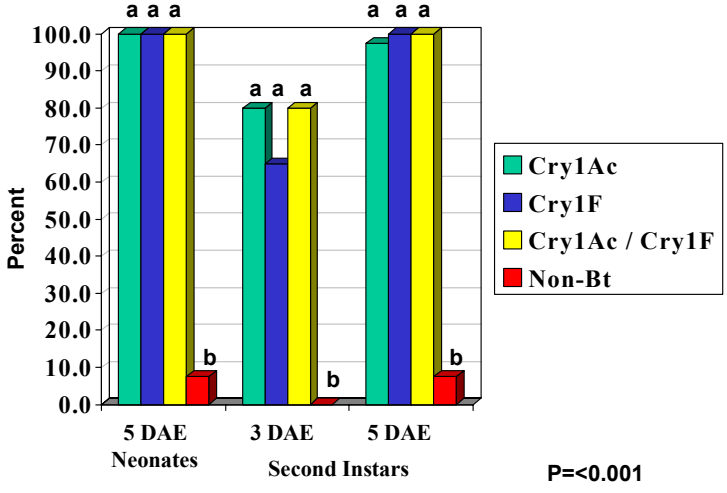

Figure 4. Mortality of tobacco budworm neonates at 5 days after exposure (DAE) and second instars at 3 and 5 DAE when fed terminal leaves from various transgenic cotton lines in laboratory bioassays. Means followed by the same letter are not significantly different (REML – ANOVA; LSD, $\alpha = 0.05$).

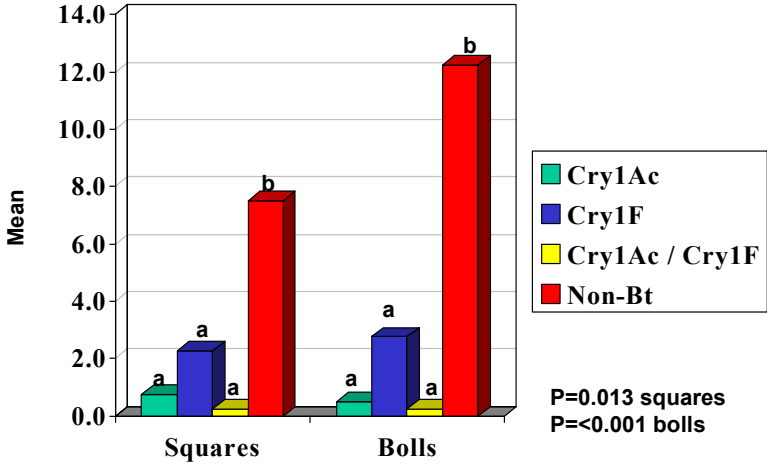

Figure 5. Mean number of damaged squares and bolls found per plot during 2002. Means followed by the same letter are not significantly different (REML – ANOVA; LSD, $\alpha = 0.05$).

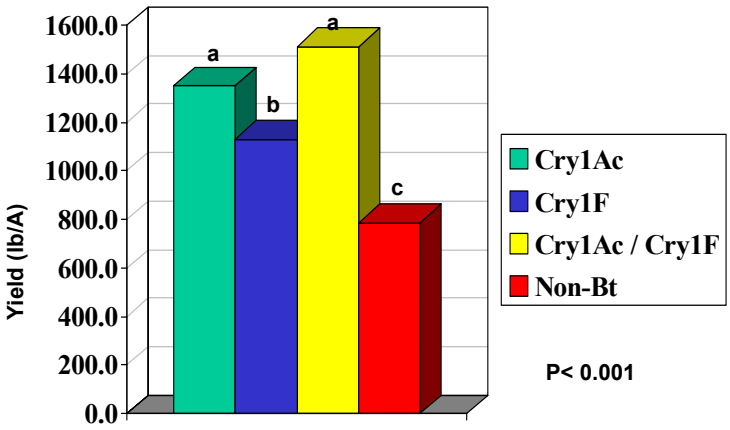

Figure 6. Seedcotton weight during 2002.