GROWTH OF WILD *PSEUDOPLUSIA INCLUDENS* (LEPIDOPTERA: NOCTUIDAE) LARVAE COLLECTED FROM BT AND NON-BT COTTON

DOUGLAS V. SUMERFORD AND WALTER L. SOLOMON
USDA-ARS, Southern Insect Management Research Unit, Stoneville, MS 38776

Cotton varieties genetically engineered to express Cry1Ac, a delta-endotoxin protein from *Bacillus thuringiensis* with insecticidal properties to many Lepidopterans, are now commercially available to aid in the control of the tobacco budworm, *Heliothis virescens* F. (Lepidoptera: Noctuidae). Most of the resistance-management strategies for Bt cotton have focussed on this primary pest, and *Helicoverpa zea* (Boddie) (Lepidoptera: Noctuidae), to a lesser degree. Because Cry1Ac is continually expressed in all tissues of the cotton plant, many Lepidopteran insects feeding on Bt cotton may experience selection pressure for improved tolerance of this insecticide. In addition to the major pests of cotton, secondary pests could also develop resistance to Cry1Ac when feeding on Bt cotton. Currently, there is little available information concerning the best strategy to manage resistance to Cry1Ac in these secondary pests (Gould and Tabashnik 1998).

Soybean loopers, *Pseudoplusia includens* (Walker) (Lepidoptera: Noctuidae) are occasional foliage feeders of cotton and have the potential to develop resistance to Cry1Ac (Mascarenhas et al. 1998). We took advantage of a natural infestation of soybean loopers (SBL) in the Mississippi Delta to investigate how well SBL larvae tolerated Cry1Ac expressed in the leaves of Bt cotton. Comparisons were made of the numbers of larvae found in Bt and non-Bt cotton and also comparisons of the growth of larvae feeding on Bt and non-Bt cotton. We also looked at the variability in tolerances of individuals feeding on Bt cotton.

Two varieties of cotton were used in this study. NuCOTN 33B (Bt cotton, treatment = 'BT') and SG125 (conventional cotton, treatment = 'NBT') were planted (9 May 1998) in a randomized design totaling 7 plots of each variety. Plot dimensions were approx. 9 × 9 m and plots were arranged in a 3 × 5 plot grid. Each plot was separated from the others by 2 m of unplanted space. On 6 July 1998, we observed a sizeable infestation of SBL larvae (first and second instars in BT and NBT treatments). Larvae from all plots were manually removed (23 July 1998), placed in plastic bags containing foliage from the plant on which each larva was feeding, and immediately brought back to the lab to be scored and weighed. The number of larvae from each plot was tabulated. Counts of larvae were log-transformed to enhance normality and to homogenize the variances in the BT and NBT treatments. A t-test was used to compare NBT and BT counts. Data are presented as untransformed average numbers per plot. In addition, larvae from 5 and 4 randomly chosen BT and NBT plots, respectively, were weighed to the nearest hundredth of a mg to look for developmental differences in SBL larvae feeding on BT and NBT foliage. Log-transformed weight (mg) data were subjected to ANOVA to determine if there were any differences in the developmental rates of larvae collected from BT and NBT plots. Bt treatment (BT vs. NBT cotton) was considered a fixed effect and plots nested within treatments [plots(treatment)] effects were considered a random source of variation (PROC GLM; SAS 1985). Satterthwaite's approximation was used to estimate the denominator degrees of freedom.

Significantly more SBL larvae were collected from NBT plots than BT plots (Fig. 1; $t = 3.336$, df = 12, $P = 0.006$). Larvae from NBT plots were also significantly larger than larvae from BT plots ($F = 238.96, df = 1, 104.5, P < 0.0001$). The weights of larvae

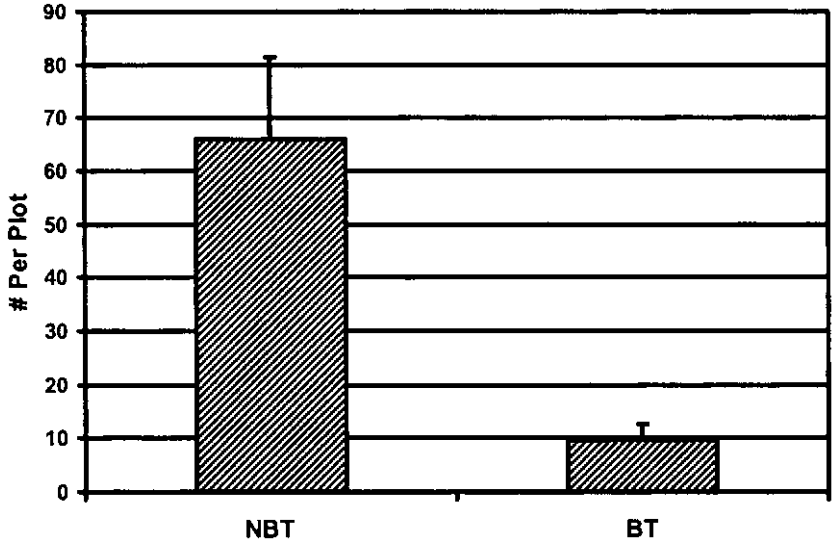

Fig. 1. The average number (±SD) of SBL larvae per plot on non-Bt cotton and Bt cotton.

for plots within treatments did not differ (Plots(treatments), $F = 0.506$, df = 7, 268, $P = 0.830$). The average weight of larvae collected from NBT plots was an order of magnitude larger than the mean weight of larvae collected from BT plots ($\bar{x}$ = 189.6 and 21.2 mg, NBT and BT, respectively). There was very little overlap in the log weights (mg) of larvae collected from NBT and BT cotton (Fig. 2). In addition, there was a great deal of variation in log weights among individuals collected from BT cotton (Fig. 2; CV = 30.4% vs. 15.4% for BT and NBT, respectively).

Although Bt cotton was developed primarily for the control of *H. virescens* in southeastern and mid-south cotton, it does have some effect on SBL. Most larvae collected from non-Bt cotton were close to pupation, in contrast to the range of sizes we observed in larvae collected from Bt cotton. Not only were SBL larvae collected from Bt cotton an order of magnitude smaller than larvae collected from non-Bt cotton, but there was also an order of magnitude difference in the log weights of the smallest and largest larvae collected from Bt cotton. The size (almost 80mg) of some of the SBL larvae feeding on Bt cotton was rather striking. Because SG125 is not the parental variety of NuCOTN 33B, it should be noted that other causes, in addition to Bt expression, might account for the differential growth of larvae on the two cotton varieties. However, in leaf tests with beet armyworms, we have seen similar differences between the growth of larvae feeding on NuCOTN 33B and its parent variety DP5415 (Sumerford, in prep).

Many quantitative traits governed by polygenes are normally distributed and populations often exhibit a large amount of variation in the trait of interest (Falconer and Mackay 1996). The log-weight distribution (mg) of collected larvae from Bt cotton was also normally distributed (Kolmogorov-Smirnov, D = 0.108, P > 0.15; Sokal and Rohlf 1996). We had planned to select for tolerance (based on growth of larvae as determined by weight) in both directions using the weighed individuals from our BT treatment. This would have given an estimate of the response to selection, and therefore

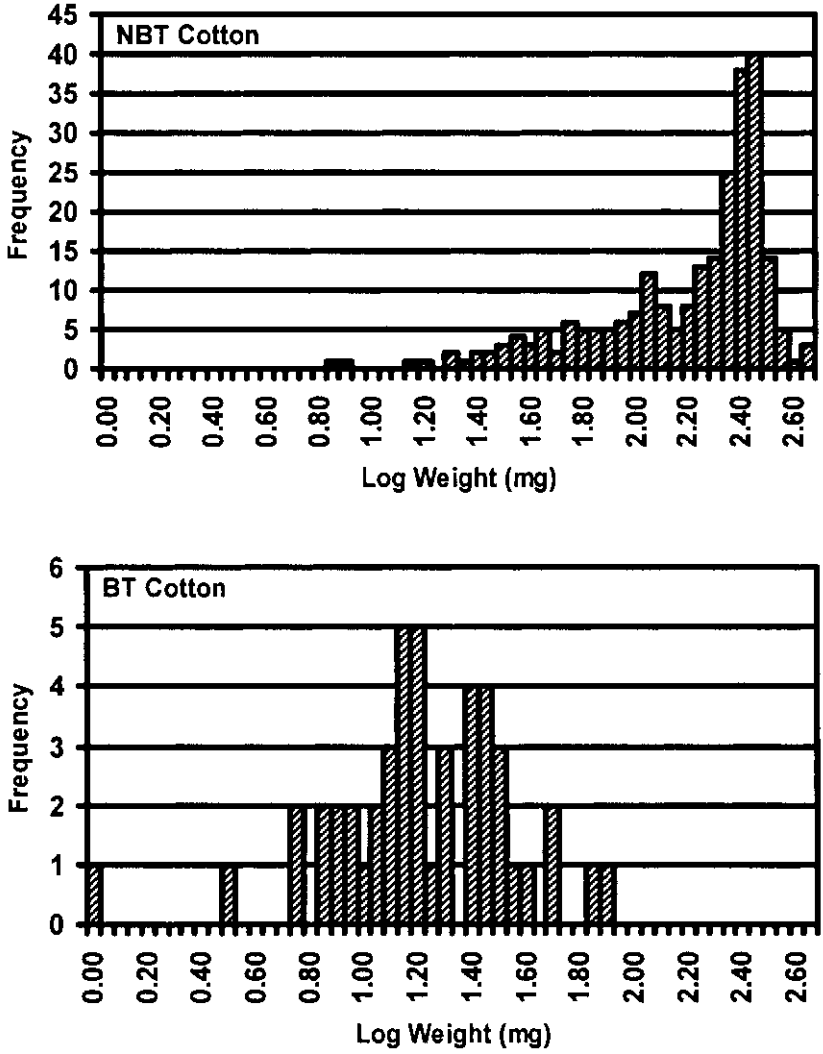

Fig. 2. Frequency distributions of log weights (mg) from SBL larvae collected from non-Bt and Bt cotton.

an estimate of the heritability of growth on the Cry1Ac toxin. However, we had difficulty obtaining viable eggs from adults in our BT and NBT treatments. If the developmental differences observed in SBL larvae from our Bt cotton are a consequence (at least in part) of oligo- or polygenic inheritance, it is unclear whether strategies designed to retard the evolution of resistance in *H. virescens* will also slow resistance development in SBL populations. Selection experiments and quantitative genetic studies would help in understanding the genetic architecture of Cry1Ac tolerance in SBL populations.

SUMMARY

Soybean looper larvae from the Mississippi Delta were collected from Bt and non-Bt cotton to determine the numbers of loopers in each variety and also to compare the rate of larval development on the two cotton varieties. There were significantly fewer larvae collected from Bt cotton than non-Bt cotton ($P < 0.01$) and the weights of these larvae were, on average, an order of magnitude smaller than larvae collected from non-Bt cotton ($P < 0.0001$). There was also an order of magnitude difference among the weights of larvae collected from Bt cotton, indicating considerable variability in the tolerance of the Bt toxin, Cry1Ac.

REFERENCES CITED

FALCONER, D., AND T. F. C. MACKAY. 1996. Introduction to quantitative genetics. 4th ed. Longman, Essex, England.

GOULD, F., AND B. TABASHNIK. 1998. Bt-cotton resistance management. Pp. 67-105 *in* M. Mellon and J. Rissler (eds.). Now or never: serious new plans to save a natural pest control. Union of Concerned Scientists, Washington, D.C.

MASCARENHAS, R. N., D. J. BOETHEL, B. R. LEONARD, M. L. BOYD, AND C. G. CLEMENS. 1998. Resistance monitoring to *Bacillus thuringiensis* insecticides for Soybean Loopers (Lepidoptera: Noctuidae) collected from soybean and transgenic Bt-cotton. J. Econ. Entomol. 91: 1044-1050.

SAS. 1985. SAS procedure guide for personal computers, vers. 6th ed. SAS Institute, Cary, NC, U.S.A.

SOKAL, R. R., AND F. J. ROHLF. 1995. Biometry. 3rd ed. Freeman, New York, NY.