

Exploring Resistance in Bt Crops

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INTRODUCTION:

Bacillus thuringiensis is a natural occurring, soil-borne bacteria that has been used for natural insect control. It consists of a spore and a toxic protein crystal within the spore. Cry1Ac protoxin is a crystal protein produced by the bacterium *Bacillus Thuringiensis* during sporulation. Cry1Ac is one of the delta endotoxins produced by this bacterium which have insecticidal properties. When the bacterium is consumed by certain insects, the toxic crystal is released and blocks the system which protects the pest's stomach from its own digestive juices. Once the stomach is penetrated, the insect dies by poisoning from the stomach contents. Because of this, it has been introduced into commercially important crops and by genetic engineering in order to confer pest resistance on those plants.

Since 1996 plants have been modified with short sequences of genes from *Bt* to express the crystal protein *Bt* makes. With this method, plants themselves can produce the proteins and protect themselves from insects without any external *Bt* and/or synthetic pesticide sprays. *Bt* crops are protected specifically against European corn borer, southwestern corn borer, tobacco budworm, cotton bollworm, pink bollworm and the Colorado potato beetle.

Due to limited access to purified proteins, SIMRU conducts studies to develop new approaches to measuring susceptibility in insects targeted by crops. This information will be attained by comparative assays between certain strains of *Bacillus thuringiensis*, such as Dipel and lyophilized tissue of Bt commercial crops.

“Dual-toxin or dual-gene Bt cottons have been widely used. The cottons controls a broader spectrum of lepidopteran pests and provide higher levels of insect control. This has implications to resistance management because of higher effective dose (Luttrell, Jackson 2012).”

MATERIALS AND METHODS:

Assays in 2014 concentrated on developing baseline data for laboratory and field populations exposed to Dipel, a commercial formulation of *Bacillus thuringiensis*. Diet incorporated and diet overlay assays were conducted with laboratory susceptible colonies of bollworm, fall armyworm and tobacco budworm. Separate concentration-mortality regressions were calculating using mortality and mortality + stunting as response variables. Additional tests were conducted with a 2013 field colony of bollworm not selected and selected via exposure to Dipel on diet. Field colonies of bollworm collected from clover in May and June from Natchez, Leland, Vicksburg, Warren, and Yazoo County were compared to the SIMRU susceptible colony.

Bio Assay trays are crucial for completing the experiment. The trays come equipped with eight compartments, each with sixteen slots each, filled with bollworm diet. Dipel and various versions of Bt plants are used (Wildstrike Cotton, Conventional Cotton, BGII Cotton, VT Pro III Corn, and Conventional Corn). Live Tissue from the plants are collected and lyophilized, mixed with water and placed on top of bollworm diet. This method is referred to as “The Overlay Method.” Bollworms are placed on the diet and rated for mortality after seven days. The response is a comparison to concentration of toxin within the diet. Challenges may occur if measurements are associated with insect development, weight gain or other process related to food intake (Luttrell, Jackson 2012).

RESULTS:

Average, minimum and maximum LC50s calculated for different assays are summarized in the table below. Fall armyworm and bollworm are much less susceptible to Dipel than tobacco budworm. Variability among different assays for a given colony conducted on different days ranged from less than 2 to greater than 9 fold, but variability among assays against the same test populations were generally 2 to 3 fold. Field populations of bollworm collected from clover in 2014 were as susceptible as the SIMRU laboratory colony. Selection of a 2013 field colony reared in the laboratory via exposure to Dipel in diet incorporated assays elevated the LC50 about 50%, but variability in assays was as great as the difference between selected and not selected strains.

Additional assays are planned with lyophilized plant tissue and purified Cry1Ac protein to confirm the possible use of Dipel as a source of Bt toxins to monitor levels of susceptibility in field populations.

REFERENCES:

Luttrell, Jackson. September 2012, *Helicoverpa zea* and Bt cotton in the United States. Volume 3(Issue 3): 213-225

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