

## The Development of Transgenic Cotton Containing a Bt Gene for Control of Lepidopteran Pests

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Monsanto Co., based in St. Louis, Missouri, US, is currently developing new cotton lines which resist damage from lepidopteran pests. The source of resistance is a protein produced by *Bacillus thuringiensis*. The protein is generally referred to as Bt.

The Bt protein was discovered approximately 80 years ago. The insecticidal activity was related to a protein in the 1950s. The protein is a stomach toxin which is specific for certain insects. This specificity accounts for a considerable portion of the value since the protein is toxic to certain insect pests, but will not harm beneficial insects. Its half life is only 2 to 3 days, so it will not persist in the environment. Also, it is safe for both humans and animals.

The first commercial insecticidal products were produced in the early 1960s and have been used to a limited extent for the last 30 years. Recently, considerable research effort has focused on Bt. This renewed interest is due to three major reasons, First, improved bacterial strains have been discovered which are more efficient at producing the Bt protein. Second, improved pesticidal formulations have increased the efficacy and persistence of the product. Finally, recent developments in biotechnology have provided an improved delivery system for the Bt. With genetic engineering, the gene which produces the Bt protein can be added to plants. Once a plant has this gene incorporated into its DNA, the plant will then produce the protein itself.

Having the Bt protein produced within the plant provides several advantages over topical applications of formulations of Bt insecticides. Since the Bt protein is produced in the plant throughout the growing season, full season lepidopteran pest control is achieved. Also, since the plants produce the protein, this will provide cost effective lepidopteran control. In addition to those benefits, Bt produced within the plants still maintains the specificity of control (no harm to beneficials) and the safety of topical applications.

The first stage in development of Bt cotton lines is to isolate and develop the gene so that it will function properly in cotton. Then, the gene is introduced into cotton using tissue cultures of cotton with transformation mediated by a common soil bacteria, *Agrobacterium tumefaciens*. Several hundred cotton lines are then screened to identify those lines which are suitable from an insect control and an agronomic standpoint. The most efficacious lines are then backcrossed into commercially useful varieties. Additional testing confirms the safety of the product and the optimum systems for utilization of the technology.

The data presented is from one set of trials conducted in the United States in 1992. The objectives of these trials were to identify the Bt lines with the best insect control and to obtain an initial assessment of commercial potential of the best lines. Other studies, which will not be discussed, involved optimizing resistance management strategies, determining economic thresholds, and analyzing the effect on other, non-lepidopteran, insect pests.

Tests were conducted in 6 locations in the southern United States and covered the entire US cotton belt. These tests were conducted in the following

states: Georgia, Alabama, Mississippi, Louisiana, Texas and Arizona. All tests, with the exception of the Alabama site, were done with academic cooperators.

Results from all sites demonstrate that certain Bt cotton lines provided season-long protection of squares and bolls. The primary pests were *Heliothis zea* and *Heliothis virescens*. The Arizona site was infested with *Pectinophora gossypiella*. The protection from lepidopteran damage afforded by the Bt lines, which were not treated with chemical insecticides for lepidopteran control, was at least as effective as, and generally more effective than, the protection provided by weekly applications of insecticides. Control of these pests was consistent throughout the growing season. The square and boll protection provided by the Bt protein also translated into yield protection; the Bt lines yielded the same whether or not they were sprayed with chemical insecticides or left untreated.

The 1992 results confirmed the results of earlier trials. Transgenic cotton lines have been identified which provide protection for *Heliothis* species and *P. gossypiella*. All flower and fruiting structures of the Bt lines were

protected throughout the growing season at all test sites and this also resulted in yield protection.

In addition to these trials, Monsanto is actively developing strategies to delay the development of resistance by these insect pests to the Bt protein. The strategies include, but are not limited to:

- Refugia, whereby sources of non-Bt cotton are grown with or near the Bt cotton to supply an alternate food source for the insects.
- The development of other Bt genes with different sites of action.
- The development of non-Bt proteins with insecticidal activity. The various strategies are being investigated prior to commercialization and will be implemented in stages after commercialization.