

*thuringiensis* VIP3A Insect Control Protein as Expressed in Event COT102 and the genetic material necessary for its production in or on cotton.

Pursuant to section 408(d)(2)(A)(i) of the FFDCA, as amended, Syngenta Seeds, Incorporated has submitted the following summary of information, data, and arguments in support of their pesticide petition. This summary was prepared by Syngenta Seeds, Incorporated and EPA has not fully evaluated the merits of the pesticide petition. The summary may have been edited by EPA if the terminology used was unclear, the summary contained extraneous material, or the summary unintentionally made the reader conclude that the findings reflected EPA's position and not the position of the petitioner.

#### A. Product Name and Proposed Use Practices

Syngenta has developed a new cotton line that expresses an insect control protein designated VIP3A. It has been genetically incorporated into a cotton plant product identified as *Bacillus thuringiensis* (Bt) VIP3A Insect Control Protein as Expressed in Event COT102. VIP3A is one of a novel class of recently discovered insecticidal proteins that occur naturally in *Bacillus thuringiensis*. The VIPs (vegetative insecticidal proteins) are produced during vegetative bacterial growth.

Other than its demonstrated insecticidal activity, VIP3A is not known to have any other biological or catalytic function. Although VIP3A protein shares no homology with known Cry proteins, extensive testing has established that VIP3A is similarly very specific in its activity, and has demonstrated toxicity only to the larvae of certain lepidopteran species, including key pests of cotton. Further, because VIP3A appears to target a different receptor than Cry proteins in sensitive species, it represents a potentially useful tool in the prevention or management of pest resistance to Cry proteins.

Upon commercial introduction, the use of transgenic VIP3A cotton plants is expected to offer an important new option in lepidopteran pest control and integrated pest management programs. Moreover, VIP3A cotton will be an attractive, biologically based alternative to the use of foliar insecticides. The use of VIP3A cotton plants is expected to offer substantial environmental and worker safety benefits associated with the reduced need for broad-spectrum insecticides. Additionally, benefits to cotton growers will likely include greater profitability, convenience and

predictability in producing a high-yielding cotton crop.

VIP3A expressing cotton plants derived from transformation event COT102 have been field tested under U.S. Department of Agriculture (USDA) Notifications and in compliance with the guidelines for USDA regulated plantings in 2000, 2001, and 2002. The overall results of those trials have indicated that cotton plants derived from event COT102 have significant and specific insecticidal activity against several lepidopteran pests including, but not limited to, *Helicoverpa zea* (cotton bollworm), *Heliothis virescens* (tobacco budworm), and *Pectinophora gossypiella* (pink bollworm).

#### B. Product Identity/Chemistry

1. *Identity of the pesticide and corresponding residues.* Cotton, *Gossypium hirsutum*, has been genetically modified to be resistant to selected lepidopteran insect pests. Insect protection was accomplished by the insertion of the VIP3A(a) gene, which was cloned from *Bacillus thuringiensis* strain AB88. The identity of the active pesticidal ingredient in cotton plants derived from transformation event COT102 includes the protein VIP3A and the genetic material necessary for its production in cotton. Research has demonstrated the specific insecticidal properties of VIP3A to certain lepidopteran insects in cotton as well as its lack of effects on nontarget organisms such as mammals, birds, fish, and beneficial insects.

2. *Magnitude of residue at the time of harvest and method used to determine the residue.* A determination of the magnitude of residue at harvest is not required for residues exempt from tolerances. However, the petitioner has provided data on the quantity of VIP3A protein measured in various plant parts including seeds of VIP3A cotton, as measured by enzyme linked immunosorbent assay (ELISA). Additionally, the petitioner has provided data on the quantity or presence of VIP3A protein in processed cottonseed products.

3. *A statement of why an analytical method for detecting and measuring the levels of the pesticide residue are not needed.* An analytical method is not required because this petition requests an exemption from tolerances. However, the petitioner has submitted an analytical method for detection of the VIP3A protein in cottonseed by ELISA analysis.

#### C. Mammalian Toxicological Profile

The VIP3A(a) gene expressed in Event COT102 cotton is very similar (ca. 99%

homology) to VIP3A or VIP3A--like genes that appear to occur commonly in Bt strains from a variety of sources. In addition, it has been determined that the VIP3A protein demonstrates insect specific toxicity and must be ingested to be active. Once in the insect gut, the VIP3A protein binds to specific receptors (different from those bound by Cry1A proteins), inserts into the membrane and forms ion-specific pores. These events disrupt the digestive processes and cause death of the insect. The lack of mammalian toxicity has been confirmed in numerous safety studies conducted in laboratory animals, which are traditional experimental surrogates for humans. These studies, summarized herein, demonstrate the lack of toxicity of the VIP3A protein following high-dose acute oral exposures to mice, rapid degradation of VIP3A upon exposure to simulated gastric fluid, and the lack of amino acid sequence similarity of the VIP3A protein to proteins known to be mammalian toxins or human allergens. It can be concluded from these studies that the VIP3A protein will be non-toxic to humans.

When proteins are toxic, they are known to act via acute mechanisms and at very low doses (Sjoblad, R.D., J.T. McClintock and R. Engler (1992) "Toxicological Considerations for Protein Components of Biological Pesticide Products." *Regulatory Toxicol. Pharmacol.* 15: 3-9). Therefore, when a protein demonstrates no acute oral toxicity in high-dose testing using a standard laboratory mammalian test species, this supports the determination that the protein will be non-toxic to humans and other mammals, and will not present a hazard under any realistic exposure scenario, including long-term exposures.

Studies conducted to assess the mammalian safety of VIP3A protein have demonstrated no toxicity. Four acute oral toxicity studies in mice have been completed. Three of the VIP3A test substances used were produced via microbial expression systems and one prepared by extracting protein from leaves of VIP3A event Pacha-derived corn plants. The four test substances contained VIP3A protein that differed from the VIP3A protein expressed in event COT102 by zero to two amino acids. At maximum dosage the microbially expressed test substance was administered at a level of 5,000 milligrams/kilogram (mg/kg) with an estimated acute lethal dose (LD)<sub>50</sub> by gavage determined to be >3,675 mg VIP3A/kg (mg/kg body weight). Because toxicity was not observed at this dose, it can be concluded that the LD<sub>50</sub> for