UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

JAN 05 2004

OFFICE OF PREVENTION, PESTICIDES AND TOXIC SUBSTANCES

MEMORANDUM

SUBJECT: EPA Review of Syngenta Seed's Vip3A Cotton Insect Resistance Management Plan For Section 3 Full Commercial Registration [Reg. No. 67979-U; MRID: 45835814]

TO: Leonard Cole (PM-90)
Regulatory Action Leader
Microbial Pesticides Branch, Biopesticides and Pollution Prevention Division (7511C)

FROM: Sharlene R. Matten, Ph.D., Biologist
Microbial Pesticides Branch, Biopesticides and Pollution Prevention Division (7511C)

1/5/04

PEER REVIEW: Alan H. Reynolds, M.S., Entomologist
Microbial Pesticides Branch, Biopesticides and Pollution Prevention Division (7511C)

ACTION REQUESTED: To review the adequacy of the Insect Resistance Management Plan and supporting data for Vip3A Cotton submitted by Syngenta Seeds for Section 3 full commercial registration.

CONCLUSIONS:

An appropriate, scientifically-sound and sustainable (includes feasibility) insect resistance management (IRM) plan is required for COT102 cotton expressing the *Bt* vegetative insecticidal protein VIP3A. The registrant, Syngenta Seeds, Inc. proposes that COT102 (cotton expressing VIP3A) be subject to the same IRM requirements which are currently required for registered *Bt* cotton products. However, there are insufficient data provided to formulate a IRM strategy. Syngenta cannot rely on the existing IRM requirements for Bollgard and Bollgard II [1]cotton

[1]Bollgard and Bollgard II are registered trademarks of Monsanto Company.

products without supplying appropriate data. The greatest deficiency is lack of data regarding the Vip3A dose for all the major target pests, i.e., *Heliothis virescens* (tobacco budworm, TBW), *Helicoverpa zea* (cotton bollworm, CBW), and *Pectinophora gossypiella* (pink bollworm, PBW). There is no acceptable high dose information for TBW. There is inadequate high dose information for CBW and PBW. These data are not conclusive. At least one additional high dose verification technique should be tested for both CBW and PBW. In addition no baseline susceptibility data are provided for any of the target pests. Sufficient data have been provided regarding the uniqueness of the mode of action of Vip3A compared to Cry1Ab, but additional cross-resistance data would be useful to confirm the low potential for cross-resistance in the target pests (rather than *Manduca sexta*) and to other toxins, e.g., Cry2Ab2, Cry1F, Cry1Ac.

CLASSIFICATION: UNACCEPTABLE

SPECIFIC RECOMMENDATIONS

1. Syngenta should provide to EPA data and/or published literature to address the pest biology and ecology for each target pest: TBW, CBW, and PBW. Specific information includes: host range, larval and adult movement, reverse migration for CBW and its impact, mating behavior and dispersal, ovipositional preferences, population dynamics, gene flow, overwintering mortality, and life cycle analysis. The varied cropping systems for cotton, including local and regional differences, should also be considered for evaluation the biology, ecology, and population dynamics and genetics of the target pests. Syngenta would have to provide appropriate data to support alternate hosts as effective refuges for CBW (but also TBW) including timing and production of larvae and adults on each alternate host, mating behavior, proximity of alternate hosts to transgenic cotton, survival and fecundity on each hosts, and fitness of adults coming off alternate hosts. Similarly, Syngenta would have to provide appropriate data regarding the effectiveness of supplemental insecticide treatment of transgenic cotton fields to control putative resistant CBW.

2. Additional data are needed to determine if COT102 cotton plant expresses a high dose of VIP3A to meet the high dose requirements for target pests (TBW, PBW and CBW) using at least two of the five high dose techniques as originally described by the 1998 FIFRA Scientific Advisory Panel Subpanel (SAP 1998). There are no acceptable data for TBW and insufficient data for CBW and PBW to make a decision as to whether COT102 produces a high dose of VIP3A. If Syngenta is unable to use two of the five recommended high dose techniques, they may propose alternate techniques to address the SAP's 25X definition of high dose.

3. Additional data are needed to determine LC_{50}'s and LC_{90}'s for the target pests listed on the label. A referenced, unpublished Syngenta report contains LD_{90}s for target pests. This report by Privalle, 2002 "Characterization of VIP3A Protein Produced in Pacha-Derived Maize and Comparison with VIP3A Protein Expressed in Recombinant Escherichia coli" Syngenta Report No.SSB-004-00 may provide additional data and should be submitted to

EPA for review.

4. Baseline susceptibility data for the VIP3A toxin are needed for all of the targeted pests (i.e., TBW, CBW, PBW). Diagnostic concentrations for testing for resistance to VIP3A need to be established for each of the target pests and a detailed resistance monitoring plan for the VIP3A protein is needed.

5. A study of how VIP3A and insect resistance management plans have impacted or will impact secondary lepidopteran pests is recommended.

6. Estimates of initial resistance allele frequency should be provided as well as appropriate models that can simulate the evolution of resistance for Vip3A cotton and impact of appropriate insect resistance mitigation strategies, i.e., appropriate refuge using both non-*Bt* cotton and other alternate hosts.

7. Additional cross-resistance potential data should be provided using Vip3A, Cry1Ac, and Cry2Ab for the target pests. Presumably Vip3A does not confer cross-resistance to Cry1Ac and Cry2Ab2, the Cry proteins expressed in Bollgard (Cry1Ac) and Bollgard II (Cry1Ac/Cry2Ab2) cotton. However, these data are not provided for the specific target insects of interest: tobacco budworm, cotton bollworm, pink bollworm, or fall armyworm. These data would be useful to confirm that Vip3A poses a novel model of action and that cross-resistance to other Cry proteins is not conferred by Vip3A. Use of laboratory-selected resistant colonies would provide some indication of the cross-resistance potential. While the likelihood of cross-resistance appears to be very low, there may be other *Bt* resistance mechanisms to consider other than binding site modification (see recent review Ferré and Van Rie, 2002).

8. Specific monitoring plans, remedial action strategies should resistance to Vip3A occur, grower education program, compliance assurance program, and research activities should be provided for COT102.

BACKGROUND

COT102 cotton expresses the vegetative insecticidal protein (VIP3A) which was isolated from *Bacillus thuringiensis* strain AB88. The cotton line Coker 312 (*Gossypium hirsutum* L. cv Coker 312) was transformed via *Agrobacterium* transformation procedures with a synthetic *vip3A(a)* gene encoding VIP3A protein and the selectable marker gene *aph4* encoding the enzyme APH4. The transformation event that produced the transgenic cotton line, designated COT102, was transformed with plasmid pCOT1. COT102 is intended to protect cotton from feeding by the primary lepidopteran pests: tobacco budworm (*Heliothis virescens*, TBW), cotton bollworm (*Helicoverpa zea*, CBW) and pink bollworm (*Pectinophora gossypiella*, PBW). Based on cotton insect loss data from 1991-2000, the three primary pests, TBW, CBW, and PBW, account for more than 77% of the yield lost and 84% of the insecticide use due to lepidopteran infestation in cotton.

REVIEW OF SYNGENTA'S VIP3A IRM SUBMISSION

1. Pest Biology and Ecology

Knowledge of pest biology and ecology is critical for the development of effective IRM strategies. For example, refuges must be designed with a solid understanding of the target pest to maximize the production of susceptible insects and increase the likelihood of random mating between susceptible and potentially resistant pests. Syngenta has not provided pest biology and ecology data (or cited published literature) for the three primary pests: TBW, CBW, and PBW. Syngenta should provide to EPA data and/or published literature to address the pest biology and ecology for each target pest.

Syngenta should provide to EPA data and/or published literature to address the pest biology and ecology for each target pest: TBW, CBW, and PBW. Specific information includes: host range, larval and adult movement, reverse migration for CBW and its impact, mating behavior and dispersal, ovipositional preferences, population dynamics, gene flow, overwintering mortality, and life cycle analysis. The varied cropping systems for cotton, including local and regional differences, should also be considered for evaluation the biology, ecology, and population dynamics and genetics of the target pests. Syngenta would have to provide appropriate data to support alternate hosts as effective refuges for CBW (but also TBW) including timing and production of larvae and adults on each alternate host, mating behavior, proximity of alternate hosts to transgenic cotton, survival and fecundity on each hosts, and fitness of adults coming off alternate hosts. Similarly, Syngenta would have to provide appropriate data regarding the effectiveness of supplemental insecticide treatment of transgenic cotton fields to control putative resistant CBW.

2. Insecticidal Activity

Insecticidal Activity Against Lepidopteran pests

Syngenta has provided the results of *in vitro* and *in planta* studies of the efficacy of the VIP3A protein against FAW, TBW, CBW and PBW (MRID #45835812 and MRID #45835814). *In planta* studies resulting from natural infestations were provided for TBW and PBW. Table 1 presents *in vitro* diet results for sensitivities to test solutions of VIP3A protein from COT012 derived cotton leaves (LPCOT102-0102) and recombinant *E. coli* (VIP3A-0199). A rank order of sensitivity was found for both test solutions: FAW<BCW<CBW<TBW. FAW was the most sensitive to VIP3A, while TBW was the least sensitive. Corrected mortality (for control mortality) results showed a slightly higher level of sensitivity of the insects to cotton-produced VIP3A over that produced from *E. coli*. This was most obvious for TBW where the mortalities were 55% and 20% obtained from COT cotton and *E. coli* VIP3A, respectively.

EPA Review. This study does not support calculation of LC_{50}'s for primary targeted species (i.e. FAW, TBW, CBW and PBW). Insecticidal activity of VIP3A against additional secondary lepidopteran pests such as beet armyworm (BAW), and soybean looper (SL) is not provided.

This study merely provides a crude indication of the rank order of sensitivity of the target insects to VIP3A produced by COT102 cotton or *E. coli* (VIP3A-099). The rank order indicates that FAW is the most sensitive species while CBW and TBW are the least sensitive. PBW was not included. Additional information is needed to assess the susceptibility of TBW, CBW, PBW, and FAW as well as secondary lepidopteran pests which may come in contact with VIP3A expressing cotton products.

Table 1. Comparison of efficacy of cotton VIP3A (LPCOT102-0102) with *E. coli* VIP3A (VIP3A-0199).

Insect	Treatment	ng VIP3A/cm²	Hours	# Dead / #Tested	% Mortality
FAW	Diet	0	96	2/24	8
	Buffer	0		1/24	4
	LPCOT-102-0102C	0		0/12	0
	LPCOT102-0102	62.5		12/12	100 (100)*
	VIP3A-0199	70.5		10/12	83
BCW	Diet	0	96	0/24	0
	Buffer	0		1/24	4
	LPCOT-102-0102C	0		3/24	12
	LPCOT102-0102	124.0		17/24	70(66)
	VIP3A-0199	141.1		13/24	54
CBW	Diet	0	144	1/24	4
	Buffer	0		2/24	8
	LPCOT-102-0102C	0		3/24	12
	LPCOT102-0102	186.0		11/24	45(38)
	VIP3A-0199	211.6		9/24	37
TBW	Diet	0	144	0/24	0
	Buffer	0		0/24	0
	LPCOT-102-0102C	0		4/24	16
	LPCOT102-0102	496.0		15/24	62(55)
	VIP3A-0199	564.2		5/24	20

*Mortality corrected for LPCOT102-0102C using Abbott's method appears in parenthesis.
FAW - fall armyworm, CBW - black cutworm, CBW - corn bollworm, TBW - tobacco budworm
Data taken from page 4, MRID #45835812.

3. **High Dose Determination**

VIP3A High Dose Determination for TBW, CBW, and PBW
Limited data have been provided to assess the high dose activity of COT102. These studies are summarized below.

CBW

Both field and laboratory studies were conducted to describe the dose of VIP3A expressing cotton event COT102 to CBW (MRID #45835814). The laboratory assessment involved using an older instar lava (L3) with an LD_{50} that was at least 25-fold higher than that of the neonate larvae (L1) and examining mortality following exposure to COT102 leaf tissue. This is one of the 1998 SAP's recommended high dose techniques. However, two techniques are necessary to confirm a high dose.

In order to identify larvae with an appropriate LD_{50}, (i.e., at least 25-fold higher than the neonate LD_{50}), meridic diet bioassays were carried out using purified VIP3A protein to establish LC_{50} values of different instars. Table 2 indicates that L3 CBW larvae were approximately 50-60 fold more tolerant to VIP3A protein than L1 larvae. Accordingly, L1 and L3 larvae were used in the experimental phase of analysis. The experimental phase consisted of placing freshly harvested leaf tissue from COT102 plants into petri dishes and placing an L1 or L3 CBW larvae on the leaf tissue. Mortality was scored at 24, 48, 72, 96 and 120 hours. One hundred replica plates were prepared for the COT102 and the non-transgenic control (Coker 312) and each larval stage.

Results indicate mortality of the L1 larvae placed on COT102 leaf tissue to be 99% after 72 hours. Mortality for L3 larvae was 66% mortality by 72 hours and 93% after 96 hours. Correcting for control mortality in the L3 larvae results in 90% mortality. However, the L3 larvae exhibit increased tolerance (50-60 fold) toward VIP3A which compensates for the discrepancy in mortality indicating that the COT102 exhibited high dose efficacy against CBW in laboratory studies. Results of LC_{50} calculations for L1 and L3 larvae are presented in Table 2.

Table 2. LC_{50} Values for CBW Larval Stages

Instar	LC_{50} (ng/cm^2)	95% Confidence Interval
L1[a]	158.1	109.5 to 228.4
L1[b]	91.6	47.9 to 175.2
L3[a]	9564.7	7814.7 to 11,706.6
L3[b]	7835.5	4559.6 to 13,464.9

[a] bioassays completed on the same day

[b] bioassays completed on the same day

TBW

In 2002, field trials to determine insect control efficacy of COT102 was assessed at seven locations across the cotton belt in AL, AR, GA, LA, MS and TX. Heliothine (*H. zea* and *H. virescens*) damage to square and bolls was monitored throughout the entire flowering period. At each location, similar damage ratings were performed on non-transgenic Coker 312. Table 3 presents the cumulative feeding damage to squares (2145 examined/treatment group) caused by Heliothine across the seven locations. COT102 provided control of the Heliothine compared to

the non-transgenic Coker control.

Table 4 presents the cumulative feeding damage to bolls (1600 examined/treatment group) caused by Heliothine across the seven locations. Similar to the square damage findings, COT102 provided control of the Heliothine compared to the non-transgenic Coker control. For both the square and boll damage findings, the application of conventional insecticide to COT102 further reduced damage to low percentages (Tables 3 and 4). Syngenta concludes that the results from these field trials suggest COT102 likely expresses a high dose VIP3A for both Heliothine pests.

Table 3. Estimates of COT102 square damage by Heliothine in field trials.

Cotton Line	% Squares Damaged (mean)	% Squares Damaged (range)
COT102	4.0	0 - 12.4
Coker	26.0	6.3- 69.8
COT102 +Insecticide	2.1	0 - 7.4
Coker +Insecticide	16.5	6.1 - 52.3

Table 4. Estimates of COT102 boll damage by Heliothine in field trials.

Cotton Line	% Bolls Damaged (mean)	% Bolls Damaged (range)
COT102	4.3	0.7 - 6.5
Coker	27.0	3.3- 52.9
COT102 +Insecticide	2.6	0.2 - 4.9
Coker +Insecticide	15.3	0.9 - 38.7

PBW

High dose status of COT102 toward PBW was evaluated in field trials performed at the University of Arizona in 2002. Bolls of COT102 (n=72) and non-transgenic Coker 312 (n=80) were individually infested with egg masses (~100 eggs/boll). The number of entry holes/boll was assessed and 5-9 days later bolls were dissected to determine the number of surviving larvae. Results indicate that bolls from both COT102 and Coker plants suffered from a large number of entry holes. Upon dissection, 276 live L3 larvae were identified from 75 of the Coker 312 bolls. In contrast, only one boll of COT102 was identified to harbor two larvae. Syngenta concludes that COT102 provides a high dose for PBW.

EPA Review

Syngenta states that COT102 cotton expressing the VIP3A insecticidal protein should be subject to the same IRM requirements as currently stated for Bollgard and Bollgard II cotton products. However, Syngenta has not provided sufficient dose data to warrant this conclusion. The 1998 SAP Subpanel recommended at least two of five techniques be used to confirm high dose. No usable data were presented to address the high dose status of COT102 to TBW. Syngenta comments that attempt to ascertain the high dose status of TBW were "fraught with insurmountable technical challenges that prevented the obtaining of reliable data." Rather their efforts focus on field performance of COT102 toward this pest under natural infestation. Field performance experiments (7 locations) under natural infestation conditions were inadequate to determine whether COT102 produced a high dose to control TBW and CBW. First, one cannot distinguish between square or boll damage caused by TBW or CBW. COT102 showed 4.0% damaged squares while COKER showed 26.0% damaged squares. Similarly, COT102 showed 4.3% damaged bolls while COKER showed 27.0% damaged bolls. Since TBW is less sensitive to the VIP3A toxin than CBW (see MRID #45835812), one can assume there is a differential efficacy shown for TBW and CBW. One cannot determine from these data that the COT102 expressed VIP3A at the $LD_{99.9}$ to control either TBW or CBW. A high dose determination cannot be made from these field performance experiments.

There is limited information for CBW and PBW. Only one technique was used to measure dose for CBW and PBW rather than two techniques as recommended by the 1998 SAP. If the second technique indicates that COT102 expresses a high dose for CBW and PBW, then there would be some certainty that there is a high dose. There is no certainty at present.

As discussed above, field performance experiments were inadequate to determine high dose for CBW. Based on the limited field and laboratory data provided, it is not clear whether COT102 produces a high dose of VIP3A to control CBW. The adjusted mortality for COT102-exposed L3 larvae at 96-hours was calculated to be 90% in contrast to the SAP-recommended value of 95%. However, at 72 hours, there was only 66% mortality of CBW L3 larvae (uncorrected mortality) and 99% mortality of CBW L1 larvae (uncorrected mortality). The tolerance of the L3 larvae towards VIP3A protein is 50- to 60-fold greater than the tolerance of the L1 larvae. The recommended SAP technique suggested that the tolerance of a later-instar larvae should be at least 25-fold greater than the L1 larvae to be a high dose event. That is, a 25-fold dilution of the COT102 tissues should still cause very high levels of mortality, >95% to kill all potential heterozygotes. This isn't the case based on Syngenta's data. Therefore, it isn't clear that COT102 produces a high dose to control CBW according to the recommendations provided by the 1998 SAP Subpanel.

High dose assessment of PBW was not conclusive. There were two PBW larvae identified in a single boll of COT102, but no additional analyses were performed. No immunological analysis was performed to determine whether these larvae were isolated from a VIP3A-expressing cotton plant (COT102 plant) or from a non-VIP3A-expressing cotton plant. There is insufficient information provided to address the high dose status of COT102 to PBW.

4. Biochemical Characteristics of VIP3A: Cross-Resistance Potential and Mode of Action: EPA Review

Mechanism of Action and Binding Characteristics
Syngenta states that "VIP3A has a novel mode of action and a novel binding site" (MRID #45766501). The VIP3A mode of action is proteolytically activated to form a toxin core in the lepidopteran larval midgut which reacts with cell membranes creating pores in the midguts of sensitive species. VIP3A shares no structural homology with delta-endotoxins that would suggest a similar mechanism of action (Estruch et al. 1996). These researchers show that VIP3A, unlike other secreted proteins, is not N-terminally processed during export.

In brush border membrane experiments using *Manduca sexta* (tobacco hornworm), Vip3A did not show binding to isolated Cry1A receptors such as the amino pepsidase N-like and cadherin-like molecules. Both Cry1Ab and Vip3A channels were voltage independent and highly cation specific, but they differed considerably in their principal conductance state and cation specificity. Thus, Vip3A channels cannot be equated with those of the Cry proteins (Lee et al., 2003). Vip3A is gut activated as are the Cry proteins. While direct structural information is lacking for Vip3A, there is no indication of similar domain organization with delta-endotoxins based on primary structure divergence and predicted secondary structure.

Syngenta has provided adequate information about the unique nature of the VIP3A mode of action. The VIP3A will bind to a different binding site(s) from the Cry1Ab toxin. Sufficient data have been provided regarding the uniqueness of the mode of action of VIP3A compared to Cry1Ab using *Manduca sexta*.

Cross Resistance Potential
Cross-resistance is most likely when toxins share key structural features, which allows one resistance mechanism to confer resistance to more than one toxin. This is, if two separate *Bt* toxins bind to the same midgut receptor or share one or more receptors, the likelihood of cross-resistance increases.

Syngenta indicates that the likelihood of cross-resistance is low because of VIP3A's unique mode of action as compared to Cry proteins. Binding data (*Manduca sexta*) indicate the VIP3A has unique binding receptors when recompared to Cry1Ab binding receptors. However, data has not been provided to show that VIP3A does not confer cross-resistance to Cry1Ac and Cry2Ab2, the Cry proteins expressed in Bollgard (Cry1Ac) and Bollgard II (Cry1Ac/Cry2Ab2) cotton for the specific target insects of interest: tobacco budworm, cotton bollworm, pink bollworm, or fall armyworm. These data would be useful to confirm that Vip3A poses a novel model of action and that cross-resistance to other Cry proteins is not conferred by Vip3A. Use of laboratory-selected resistant colonies would provide some indication of the cross-resistance potential. English et al. (1994) concluded that binding characteristics of cotton bollworm to Cry1A and Cry2A toxins were different. Based on the mode of action data for VIP3A and unique binding sites as compared to Cry1Ab, the likelihood of cross-resistance appears to be very low; however, there

may be other *Bt* resistance mechanisms to consider other than binding site modification (see recent review Ferré and Van Rie, 2002). Based on the available information, the introduction of transgenic cotton expressing VIP3A would provide an alternative mode of action that would relieve the continued selection pressure by Cry proteins, thus sustaining their collective longer-term effectiveness and commercial viability depending on market adoption of different transgenic cotton products. Predictive resistance evolution models would be very useful to illustrate how the introduction of VIP3A cotton would reduce the selection pressure and, consequently, resistance evolution by the target pests to VIP3A, Cry1Ac, Cry1F, and Cry2Ab2.

5. Resistance Modeling: EPA Review

Predictive modeling is the only effective way to compare the relate rates of resistance evolution to multiple toxins. Syngenta has provided no specific predictive modeling information by which one can project the delay of onset of resistance to cotton insect pests through the use of VIP3A presumably deployed as a mosaic with Bollgard and Bollgard II. Syngenta references Roush (1998) and Caprio (1998) predictive models that examine the relative delay in resistance evolution from single vs. multiple toxins introduced sequentially, as a mosaic, or as a pyramid. Syngenta claims that mosaics of different transgenic cotton cultivars expressing different toxins (unique binding sites) would be no less effective than insect resistance management than a sequential introduction of different transgenic cotton cultivars expressing different toxins (unique binding sites), but this does not seem to be borne out by Roush (1998), Caprio (1998), and Zhao et al. (2003).

Roush (1998) showed that mosaics select for resistance more quickly than either pyramid or sequential deployment. Both Roush (1998) and Caprio (1998) found that a pyramid was more effective in delaying resistance than either a sequential introduction of single toxins or a mosaic of single toxins. Zhao et al. (2003) has demonstrated that Bt broccoli plants expressing two Bt toxins will delay diamondback moth resistance compared to single toxins used sequentially or in a mosaic (mosaic was the worst case). Syngenta should provide more extensive analysis for VIP3A in the context of these predictive models recognizing that VIP3A will be introduced as a mosaic. The rate of resistance evolution will be very dependent of the initial resistance allele frequency, the genetics of resistance, and the dominance (survival of heterozygotes).

It is generally recognized that the introduction of a new toxin, VIP3A, with an unrelated mode of action to the Cry1A toxins, and presumably the Cry2A toxins, that selection pressure would be reduced and resistance evolution would be delayed because there would be three unique modes of action. Because Syngenta has not provided sufficient dose data for VIP3A, one cannot evaluate the effectiveness of this toxin to delay resistance and consequently, one cannot substantiate Syngenta's claim of delayed resistance.

6. Structured Refuge

Syngenta indicates that the current refuge requirements for Bollgard and Bollgard II cotton are also applicable to it's COT102 expressing VIP3A. Current refuge options are: 1) 5% external,

unsprayed structured refuge (must be within ½ mile of Bollgard fields and at least 150 feet wide, but preferably 300 feet wide), 2) 5% embedded refuge (must be at least 150 feet wide, but preferably 300 feet wide), 3) 20% external, sprayed structured refuge (must be within 1 mile of the *Bt* fields), and 4) community refuge (either 5% external, unsprayed or 20% external, sprayed refuge options allowed).

EPA Review. Without adequate dose information for TBW, CBW, and PBW, it is impossible to determine whether the current refuge options are appropriate for Syngenta's VIP3A (COT102) cotton.

7. Resistance Monitoring

Syngenta notes that COT102 should be subject to the same resistance monitoring requirements as currently stated for Bollgard and Bollgard II products and summarizes the current requirements (EPA 2001).

EPA Review. Baseline susceptibility data to the VIP3A toxin for the key pests, TBW, CBW and PBW has not been provided by Syngenta.. Additionally, susceptibility should be provided for FAW and BCW (black cutworm). No specific resistance monitoring program for VIP3A has been provided. It is recommended that Syngenta provide a specific monitoring program for COT102 to address the three primary target pests: TBW, CBW, and PBW. Consultation with the Southern Insect Management Research Unit (SIMRU) USDA/ARS at Stoneville and other cotton insect entomologists should also be considered as part of the development of a resistance monitoring program. To develop the monitoring program, Syngenta should provide to EPA baseline susceptibility data collected at various locations across the Cotton Belt for at least one growing season prior to commercialization. Following collection of baseline susceptibility data, Syngenta should establish diagnostic concentrations for testing for resistance to VIP3A.

8. Remedial Action Plans

Syngenta notes that it should be subject to the same remedial action program as currently stated for Bollgard and Bollgard II products and summarizes the current remedial action plans required for these two products (EPA 2001).

EPA Review. Without a clear understanding of dose, baseline susceptibility, initial resistance allele frequency etc., an appropriate IRM strategy cannot be devised at this time. It should be noted that Monsanto has submitted a revised remedial action plan for TBW and CBW (as per the terms and conditions of the September 29, 2001 registration agreement for Bollgard cotton) that is still under consideration by the Agency. If the triggers for remedial action plan are appropriate for VIP3A (COT102) cotton, then whatever EPA'approved Remedial Action Plans for TBW, CBW, or PBW should also be applicable to VIP3A cotton.

8. Grower education, compliance, and annual reporting

Syngenta states that it will adher to the grower education, compliance assurance, and annual reporting requirements as stated in EPA's *Bt* Crops Plant-Incorporated Protectant Biopesticides Registration Action Document (EPA 2001).

EPA Review. No specific information was provided; therefore, it is not possible to assess Syngenta's grower education and compliance assurance programs.

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