(10)

A
cf

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

27PP

OFFICE OF
PREVENTION, PESTICIDES AN
TOXIC SUBSTANCES

12/19/96

MEMORANDUM

SUBJECT: Consideration of Section 3(c)(7)(B) Conditional Registration for Monsanto's *Bt* Corn Plant-pesticide: *Bacillus thuringiensis* CryIA(b) Delta-Endotoxin and the Genetic Material Necessary for It's Production in Corn (EPA File Symbol 524-UIO)

-DECISION MEMORANDUM-

FROM: Janet L. Andersen, Director
Biopesticides and Pollution Prevention Division

TO: Daniel M. Barolo, Director
Office of Pesticide Programs

I. ISSUE

Should the Agency conditionally register Monsanto's Bt corn product to expand the use of this plant-pesticide from limited plant propagation to full commercial use in field corn pursuant to FIFRA §3(c)(7)(B)?

The limitations currently placed upon the use of the active ingredient [which controls the European corn borer *Ostrinia nubilalis* (Huebner)] include but are not limited to the acreage which may be planted, the duration of the registration, geographic areas where the product may be used, and post-harvest agricultural practices.

II. SUMMARY AND REGULATORY BACKGROUND

A. Registration Applications

On 5/29/96, BPPD registered *Bacillus thuringiensis* delta-endotoxin as produced by the *cryIA(b)* gene and the genetic material for its production (PV-ZMCT01) in corn, EPA Reg. No. 524-492. This was a plant propagation registration. Although this new active ingredient is not limited to a particular corn line, the registration was originally limited to corn line MON 801. On 7/16/96, BPPD amended this registration to allow plantings of corn line MON 810.

MON 810 and MON 801 were each transformed with the same plasmid construct (PV-ZMCT01). The MON 810 progeny express a slightly truncated version of CryIA(b) compared to MON 801, but the active site is still retained. The MON 810 progeny do not express in detectable levels the marker gene products found in MON 801 progeny.

Monsanto has submitted an application which broadens the current use of their plant-pesticide registered for seed increase/hybrid production of the MON 810 corn line. This new registration would expand the use of this plant-pesticide to include the full commercial use for field corn for corn line MON 810 only. Per Monsanto's request, the plant-pesticide labeling will not specify plasmid construct PV-ZMCT01. The Agency herewith considers the active ingredient to be *Bacillus thuringiensis* CryIA(b) delta-endotoxin and the genetic material for its production in corn. The registration is currently limited to corn line MON 810, but could be amended in the future to include other corn lines and plasmid constructs provided sufficient supporting data was submitted.

B. Tolerance Petitions

Monsanto submitted a petition (**PP 5F4473**) which requested that EPA establish an exemption from the requirement of a tolerance for residues of the plant-pesticide active ingredients *Bacillus thuringiensis* CryIA(b) delta-endotoxin and the genetic material necessary for its production in all plants. This regulation eliminates the need to establish a maximum permissible level for residues of these plant-pesticides in all plant raw agricultural commodities. This tolerance exemption was made final on 8/2/96.

C. Plant-Pesticide Policy

The Agency published its proposed position on the regulation of pesticidal substances produced in plants (**59 FR 60496, November 23, 1994**). In the proposal, the Agency would designate the pesticidal substances produced by plants as plant-pesticides. In addition, the Agency issued proposed regulations that define certain categories of plant-pesticides that would be exempt from regulation under FIFRA and FFDCA. Plant-pesticides not exempt would be subject to regulation. The *Bacillus thuringiensis* delta-endotoxins are examples of plant-pesticides that would continue to be regulated under the proposal.

The Agency issued a registration limited in scope and duration that allowed Monsanto to

2821

produce field corn seed for seed increase and hybrid production only.

D. Conclusion

The Biopesticides and Pollution Prevention Division (BPPD), the Biological and Economic Analysis Division (BEAD), and the Pesticide Resistance Management Workgroup (PRMW) have evaluated the data submitted and/or cited by Monsanto. Based on these data and other relevant information, OPP believes that: 1) the applicant has submitted/cited satisfactory data pertaining to the proposed additional use, 2) the product will perform its intended function, and 3) the new use, as proposed by the registrant, would not significantly increase the risks of any unreasonable adverse effects to humans, nontarget organisms, or the environment from the use of this product in field corn in that potential benefits of use of the product outweigh the potential risks and residues that result from use of the product are safe within the meaning of FFDCA section 408. BPPD scientists have reviewed the information submitted with respect to health effects, and these data show that the product will be digested like any other protein and genetic material and will have no significant effects on human health. Likewise, the data submitted for ecological effects have identified no significant hazards to non-target organisms. The pesticide resistance management plan submitted by Northrup King and cited by Monsanto has been reviewed. Certain terms and conditions were deemed necessary for these registrations in order to mitigate the risk of insect resistance to *Bacillus thuringiensis*.

The registration for use of this product in field corn, if granted, would be conditional under section 3(c)(7)(B) of FIFRA. The Agency is imposing terms and conditions as outlined in Section VI of this document to address resistance management concerns. These conditions and terms are being imposed to address concerns that the target insects may develop resistance to *Bt* plants and sprays which can be used on corn, cotton, and a variety of vegetable crops. The ecological effects data requirements imposed on the existing FIFRA § 3(c)(7)(C) registration for this plant-pesticide are still required to address *Collembola* and *Daphnia* data gaps.

III. PUBLIC COMMENTS TO NOTICE OF RECEIPT

A. Comment in Response to Seed Increase and Full Commercial Use of this Plant-Pesticide

The Agency received one comment opposing tolerance levels that were described by the commentor as being proposed in text attached to the end of her letter. The notice upon which comment was received did not give notice of filing or propose a tolerance level. Rather, it announced the receipt of applications to register new active ingredients including the *Bt* corn that is the subject of this Decision Memorandum.

The commentor raised the issue of potential roles of pesticides in neurotoxicity and supported "safe/safer" alternatives to toxic chemicals. The Agency believes that the subject active

ingredient does not pose the risk of neurotoxicity. The data submitted by Monsanto support the prediction that the CryIA(b) protein would be non-toxic to humans. When proteins are toxic, they are known to act via acute mechanisms and at very low dose levels [Sjoblad, Roy D., et al. "Toxicological Considerations for Protein Components of Biological Pesticide Products," Regulatory Toxicology and Pharmacology 15, 3-9 (1992)]. Therefore, since no significant acute effects were observed, even at relatively high dose levels, the CryIA(b) delta-endotoxin is not considered toxic.

For human health, this product falls into the category of "safe/safer" pesticides, as supported by the response of the commentor.

B. General Comments in Response to *Bt* Corn

Several comments were received and addressed in the Ciba/Mycogen *Bt* corn decision document of August, 1995. The main concern expressed by commentors was that of insect resistance development. In addition, over 800 comments calling for the Agency to have "workable resistance management plans" before allowing further *Bt* plant-pesticide registrations have been received by the Agency. The Agency believes that the terms and limitations of this registration relative to resistance management adequately mitigate the risk of resistance developing during the duration of this conditional registration.

Margaret Mellon and Jane Rissler of the Union of Concerned Scientists and Rebecca Goldberg of the Environmental Defense Fund sent a 10/10/96 to Assistant Administrator Dr. Lynn Goldman. The letter stated that Bt resistance management is at a critical stage and they requested that EPA: (1) convene a meeting of the FIFRA Scientific Advisory Panel (SAP) in the Fall of this year to evaluate current Bt resistance management efforts, (2) prepare a report on the status of resistance management, and (3) suspend registrations of Bt-corn and Bt-cotton and delay approvals of other Bt crops.

The request that the EPA should convene a meeting within the FIFRA SAP to evaluate resistance management efforts has merit. However, SAP meetings are planned several months in advance. OPPTS is exploring having either a SAP meeting or similar forum in 1997. In order to have a productive meeting several preparatory steps must occur before to such a meeting is convened.

First, the EPA has to evaluate the information/data being generated by the Monsanto Company on the incidents involving the company's Bt-cotton and the cotton bollworm in 1996. The Agency understands Monsanto has just submitted their information to EPA and it must clear our front end processing before staff analysis can begin. EPA's analysis of these data along with actual data that are not considered confidential business information (CBI), should be made available at a SAP meeting. It is EPA's understanding that Monsanto conducted a number of assays and studies to investigate the lack of effective cotton bollworm control in Texas.

Second, EPA must have an opportunity to evaluate the use pattern data and research data being supplied by the registrants to EPA for Bt corn and Bt-cotton. Annual information reports are due to EPA each January 31st. EPA believes these reports will serve as the basis for any presentation at such a public meeting. It will take the Agency a few months, from the time the material is submitted, to conduct an evaluation, write the report, and then plan and organize the public meeting.

The Agency believes that, absent a thorough analysis of the situation, suspension of the current Bt-corn and Bt-cotton registrations and delaying the registration of any additional Bt crops would be a precipitous action. EPA will consider the value of such actions only after we have obtained and analyzed pertinent information. Agency scientists communicate frequently with academicians, extension personnel, and commodity groups regarding resistance management practices and experiences in the field. In addition, registrants of Bt crops are required by EPA to report confirmed insect resistance within 30 days and take corrective action after consultation with EPA. Should any of this information demonstrate that a suspension is necessary, EPA will act to suspend the registration of Bt corn and cotton products.

A subpanel of the independent FIFRA SAP met on March 1, 1995 to discuss in part, resistance management of Bt crops. Although that meeting focused primarily on Bt potatoes, the FIFRA SAP subpanel also discussed Bt corn and Bt cotton. The subpanel members recommended that in order to refine existing resistance management plans, large scale use of these plant-pesticides was needed. The Agency agrees with this approach and is allowing such large-scale use, with appropriate safeguards to protect against the development of resistance, while requiring registrants to conduct research necessary to develop acceptable long-term resistance management plans.

Moreover, the Agency has raised, in general, issue of pesticide resistance management to its Pesticide Program Dialog Committee (PPDC). The PPDC supports EPA's continued efforts to protect the use of Bt foliar pesticides and plant-pesticides.

EPA is pursuing a public meeting to discuss lessons learned to date and what activities are appropriate for the future.

C. Response to van Duyn Letter Regarding the Effects of *Bt* Corn Producing Toxin in the Silks and Kernels on the Corn Earworms

On April 22, 1996 Dr. John Van Duyn, North Carolina Cooperative Extension Service, N.C. State University sent a letter to the Agency in regard to restricting the use of *Bt* corn in areas that also grow cotton (or vice-versa). He concludes that sales should not be restricted because there are adequate refugia provided by alternate crops and weeds that would be provide alternate hosts for European corn borer (ECB) and CEW (corn earworm, also known as the bollworm with the scientific name of Helicoverpa zea (Boddie)) other than just corn or cotton.

While EPA agrees that market driven refugia, alternate crops, and weed hosts will provide adequate refugia in the corn belt for the duration of this registration, EPA does not agree that alternate crops and weed hosts provide adequate refugia in cotton growing states. Section IV. E. Resistance Management of this document discusses the risk and benefit considerations made by the Agency which will allow for limited use of this product in cotton growing states. Dr. Van Duyn's letter, along with other public comments and registrant submissions were included in the Agency's registration decision.

IV. SCIENCE ASSESSMENT

The discussion that follows summarizes BPPD's reviews of the data available to the Agency on these plant-pesticide active ingredient. A more detailed discussion of this assessment is provided in the Data Evaluation Records for the studies summarized below.

Monsanto's MON 810 Corn Line

Monsanto's corn line MON 810 was produced by ballistically transforming another proprietary corn line with plasmid construct PV-ZMCT01. Plasmid construct PV-ZMCT01 consists of plasmids PV-ZMBK07 & PV-ZMGT10 ballistically introduced together. The MON 810 line of corn is similar to MON 801 corn in that they both were derived from transformation events utilizing PV-ZMCT01. The MON 810 only expresses a truncated version of CryIA(b) delta-endotoxin rather than the full length version of CryIA(b) and the marker gene products found in MON 801.

MON 810 and MON 801 were each transformed with the same plasmid construct (PV-ZMCT01). The MON 810 progeny express a slightly truncated version of CryIA(b) compared to MON 801, but the active site is still retained. The MON 810 progeny do not express in detectable levels the marker gene products found in MON 801 progeny.

The level of CryIA(b) produced in corn line MON 801 progeny has apparently decreased with breeding over time.

A. Human Health

1. Product Analysis - CryIA(b)

Data were presented which showed that the truncated *CryIA(b)* toxin can be extracted from corn leaf tissue and this purified material displays characters and activities similar to that produced in *E. coli* which has been modified to produce *CryIA(b)*. The similarities are shown for the tryptic core proteins in molecular weight after SDS-PAGE, immunorecognition in Western blots and ELISA, partial amino acid sequence analysis, lack of glycosylation and bioactivity against either European corn borer or corn earworm. This analysis supports the

use of the microbially produced toxin as an analogue for the plant produced protein in mammalian toxicity testing.

2. Toxicology Assessment

There is a reasonable certainty that no harm will result from aggregate exposure to the United States population, including infants and children, to the CryIA(b) protein and the genetic material necessary for its production. This includes all anticipated dietary exposures and all other exposures for which there is reliable information.

The data submitted regarding potential health effects of CryIA(b) include information on the characterization of the expressed protein in corn, the acute oral toxicity of CryIA(b) , and *in vitro* digestibility studies of the protein. The results of these studies were determined applicable to evaluate human risk and the validity, completeness, and reliability of the available data from the studies were considered. The acute oral toxicity test of bacterially-derived CryIA(b) protein showed no test substance related deaths at a dose of 4000 mg/kg. This doses represents the highest amounts that could be administered with the microbially produced test substances.

Although CryIA(b) expression level data was required for an environmental fate and effects assessment, residue chemistry data were not required for a human health effects assessment of the subject plant-pesticide ingredients because of the lack of mammalian toxicity. Both (1) available information concerning the dietary consumption patterns of consumers (and major identifiable subgroups of consumers including infants and children) and (2) safety factors which, in the opinion of experts qualified by scientific training and experience to evaluate the safety of food additives, are generally recognized as appropriate for the use of animal experimentation data were not evaluated because the lack of mammalian toxicity at high levels of exposure demonstrate the safety of the product at levels above possible maximum exposure levels. This is similar to the Agency position regarding toxicity and the requirement of residue data for the microbial *Bacillus thuringiensis* products from which this plant-pesticide was derived. [See 40 CFR Sec. 158.740(b).] For microbial products, further toxicity testing to verify the observed effects and clarify the source of the effects (Tiers II & III) and residue data are triggered by significant acute effects in studies such as the mouse oral toxicity study.

The acute oral toxicity data submitted support the prediction that the CryIA(b) protein would be non-toxic to humans. When proteins are toxic, they are known to act via acute mechanisms and at very low dose levels [Sjoblad, Roy D., *et al.* "Toxicological Considerations for Protein Components of Biological Pesticide Products," Regulatory Toxicology and Pharmacology 15, 3-9 (1992)]. Therefore, since no effects were shown to be caused by the plant-pesticide, even at relatively high dose levels, the CryIA(b) delta-endotoxin protein is not considered toxic.

Adequate information was submitted to show that the CryIA(b) test material derived from

microbial cultures were biochemically and, functionally similar to the proteins produced by the plant-pesticide ingredients in corn. Production of microbially produced protein was chosen in order to obtain sufficient material for testing. In addition, the *in vitro* digestibility studies indicate the proteins would be rapidly degraded following ingestion.

The genetic material necessary for the production of the plant-pesticide active and inert ingredients are the nucleic acids (DNA) which comprise (1) genetic material encoding these proteins and (2) their regulatory regions. "Regulatory regions" are the genetic material that control the expression of the genetic material encoding the proteins, such as promoters, terminators, and enhancers. DNA is common to all forms of plant and animal life and the Agency knows of no instance where these nucleic acids have been associated with toxic effects related to their consumption as a component of food. These ubiquitous nucleic acids as they appear in the subject active ingredient have been adequately characterized by the applicant. Therefore, no mammalian toxicity is anticipated from dietary exposure to the genetic material necessary for the production of the subject active and inert plant pesticidal ingredients.

BPPD has considered available information on the variability of the sensitivities of major identifiable subgroups of consumers including infants and children and the neurological differences between infants and children and adults and the neurological differences between infants and children and adults and effects of *in utero* exposure to the plant-pesticides. Since CryIA(b) is a protein, allergenic sensitivities were considered. Current scientific knowledge suggests that common food allergens tend to be resistant to degradation by heat, acid, and proteases, are glycosylated and present at high concentrations in the food. Data has been submitted which demonstrates that the CryIA(b) delta-endotoxin is rapidly degraded by gastric fluid *in vitro* and is non-glycosylated. Studies submitted to EPA done in laboratory animals have not indicated any potential for allergic reactions to *B. thuringiensis* or its components, including the delta-endotoxin in the crystal protein. Despite decades of widespread use of *Bacillus thuringiensis* as a pesticide (it has been registered since 1961), there have been no confirmed reports of immediate or delayed allergic reactions to the delta-endotoxin itself despite significant oral, dermal and inhalation exposure to the microbial product. Several reports under FIFRA § 6(a)2 have been made for various *Bacillus thuringiensis* products claiming allergic reactions. However, the Agency determined these reactions were not due to *Bacillus thuringiensis* itself or any of the Cry toxins. Thus, the potential for the CryIA(b) protein to be a food allergen is minimal.

BPPD has considered available information on the cumulative effects of such residues and other substances that have a common mechanism of toxicity. These considerations included the cumulative effects on infants and children of such residues and other substances with a common mechanism of toxicity. Consideration of a common mode of toxicity is not appropriate given that there is no indication of mammalian toxicity of the plant-pesticides and no information that indicates that toxic effects would be cumulative with any other compounds.

BPPD has considered available information on the aggregate exposure levels of consumers

8827

(and major identifiable subgroups of consumers) to the pesticide chemical residue and to other related substances. These considerations include dietary exposure under the tolerance exemption and all other tolerances or exemptions in effect for the plant-pesticide chemical residue, and exposure from non-occupational sources. Exposure via the skin or inhalation is not likely since the plant-pesticides are contained within plant cells which essentially eliminates these exposure routes or reduces these exposure routes to negligible. Oral exposure, at very low levels, may occur from ingestion of processed corn products and drinking water. However a lack of mammalian toxicity and the digestibility of the plant-pesticides has been demonstrated. The use sites for CryIA(b) delta endotoxin are all agricultural for control of lepidopteran insects. Therefore, exposure via residential or lawn use to infants and children is not expected.

BPPD has considered available information on whether the plant-pesticides may have an effect in humans that is similar to an effect produced by a naturally occurring estrogen or other endocrine effects. The active ingredient is a protein plant-pesticide derived from the microorganism *Bacillus thuringiensis*. No known metabolite that acts as an "endocrine disrupter" is produced by this microorganism. Therefore, no adverse effects to the endocrine system is known or expected.

3. Tolerance Exemption Conclusions

The Agency has concluded that establishment of a tolerance is not necessary to protect the public health and established an exemption from tolerance requirements for the active ingredient in this product on 8/2/96 as set forth below. This exemption remains in effect pursuant to FFDCA section 408(j)(3).

a. 40 CFR 180.1173

Bacillus thuringiensis CryIA(b) delta-endotoxin and the genetic material necessary for its production all plants.

Bacillus thuringiensis CryIA(b) delta-endotoxin and the genetic material necessary for its production in all plants are exempt from the requirement of a tolerance when used as plant-pesticides in all plant raw agricultural commodities. "Genetic material necessary for its production" means the genetic material which comprise (1) genetic material encoding the CryIA(b) delta-endotoxin and (2) its regulatory regions. "Regulatory regions" are the genetic material that control the expression of the genetic material encoding the CryIA(b) delta-endotoxin, such as promoters, terminators, and enhances.

B. Gene Flow Potential

9827

1. Potential for Outcrossing and Weediness

Although corn is thought to have descended from a wild weedy species, corn today cannot exist in the wild as a weed because the female inflorescence, or the ear, restricts seed dispersal. Corn is an open pollinating (cross-fertilizing) species, probably descended from teosinte, which is more weedy, has more tillers, and does not have ears, as such.

2. Potential for Outcrossing with Wild Corn Species

a. *Teosinte*

Like corn, teosinte also has 10 chromosomes, is wind (open) pollinated, and tends to outcross, but is a highly variable species genetically compatible and interfertile with corn. Corn and compatible species of teosinte freely hybridize when in proximity to each other. In Mexico and Guatemala, teosinte exists as a weed around the margins of corn fields. A frequency of one F1 hybrid (corn x teosinte) for every 500 corn plants, or 2-5% of the teosinte population, has been reported. The F1 hybrid is robust, fertile, and capable of backcrossing to corn. However, except for special plantings, teosinte is not present in the U.S. Its natural distribution is limited to Mexico, and Guatemala.

b. *Tripsacum*

Tripsacum/corn hybrids have not been observed in the field, but have been accomplished in the laboratory using special techniques under highly controlled conditions. The risk of *Tripsacum*/corn hybrids in the field is considered minimal. Tripsacum/teosinte hybrids have not been able to be produced. Tripsacum species are perennials and seem more closely related to the genus *Manisurus* than either corn or teosinte. *Tripsacum*/corn offspring, when they occur, display various levels of sterility. Of the 16 species of *Tripsacum* described, one is native to the southern tip of Florida, 12 are native to Mexico and Guatemala, and 3 are native to South America.

3. Potential for Outcrossing with Cultivated Zea Varieties

Corn pollen has been shown to travel up to 2 miles under favorable wind conditions. All corns will interpollinate except for certain popcorn varieties. Corn pollen germinates almost immediately after pollination and completes fertilization within 24 hours. Thus corn pollen is highly promiscuous and certification standards for distances between different corn genotypes have been established to maintain desired levels of purity in the production of hybrid corn (a minimum of 660 feet).

4. Weediness of Corn

Transformation causes no change in a corn plant's inability to exist as a weed. Likewise, the

ability to outcross with teosinte and tripsacum (under carefully controlled conditions) will not be changed. Since both teosinte and tripsacum are included in botanical gardens in the U.S., the possibility exists (although unlikely) that exchange of genes could occur between corn and its wild relatives. However, no such case has been known or reported in the U.S.

Gene exchange between cultivated corn and transformed corn would be similar to what naturally occurs at the present time within cultivated corn. Plant architecture and reproductive capacity of the intercrossed plants will be similar to normal corn, and the chance that a weedy type of corn will result from outcrossing with cultivated corn is extremely remote.

C. Environmental Fate

1. Laboratory Degradation Study

B.t.k. CryIA(b) protein bioactivity, added to the soil as a component of corn line #754-10-1 tissue decreased with an estimated half life of 1.6 days and an estimated DT_{90} of 15 days. CryIA(b) protein bioactivity of corn line #754-10-1 tissue incubated without soil decreased with an estimated half life of 25.6 days, and a DT_{90} of 40.7 days. The bioactivity of purified CryIA(b) protein in soil decreased with an estimated half life of 8.3 days and a DT_{90} of 32.5 days.

2. Field Data

1994 field data regarding MON 810 demonstrated expression levels of 0.18-0.39 ug/g in grain, 7.93 -10.34 ug/g in the leaf, 3.65-4.65 ug/g in the whole plant, and 0.09 ug/g in the pollen. MON 810 does not express detectable levels of the marker gene products and the CryIA(b) protein is more truncated then in MON 801.

MON 810 was shown to be stable in expression between 1994 and 1995. 1995 U.S. field data showed 5.2-10.6 ug/g in the leaf, 2.3-4.5 ug/g in forage, and 0.4-0.9 ug/g in the grain. 1995 French field data showed 7.6-9.4 ug/g in the leaf, 4.1-5.6 ug/g in forage, and 0.4-0.7 ug/g in the kernel.

D. Ecological Effects

1. Background

Acceptable studies have been submitted which demonstrate that *E. coli*-derived, purified B.t.k. CryIA(b) toxin has minimal adverse impact on the honey bee, and other non-target insects (parasitic hymenopteran, green lacewing, and lady bird beetles), and soil organisms (earthworm). Quail and catfish studies were generated using CryIA(b) containing kernels. Additional data are needed to more fully characterize the risk to Collembola and aquatic invertebrates. However, the overall risk to populations of these organisms is anticipated by

11827

the Agency to be minimal during the duration of this conditional registration.

MON 810 and MON 801 were each transformed with the same plasmid construct (PV-ZMCT01). The MON 810 progeny express a slightly truncated version of CryIA(b) compared to MON 801, but the active site is still retained. The MON 810 progeny do not express in detectable levels the marker gene products found in MON 801 progeny.

The level of CryIA(b) produced in corn line MON 801 progeny has decreased with breeding over time. On 5/29/96, BPPD registered *Bacillus thuringiensis* delta-endotoxin as produced by the *cryIA(b)* gene and the genetic material necessary for its production (PV-ZMCT01) in corn. Although this new active ingredient is not limited to a particular corn line, the registration was originally limited to corn line MON 801.

On 7/16/96, BPPD amended this registration to allow plantings of corn line MON 810. However; additional studies of quail, catfish, and Daphnia were required for the full commercial registration of MON 810. These studies were listed as data gaps because although some of the data in the nontarget organism database supporting the registration were generated using *E. coli* produced Bt protein, the test substance for the quail and catfish studies already reviewed was MON 801 seed. Further, MON 810 expresses detectable levels of CryIA(b) in pollen and therefore may pose some degree of exposure to Daphnia, whereas MON 801 does not.

2. New Information

According to Monsanto "{t}he fish and quail studies were performed with MON 801 grain which expressed the CryIA(b) protein in the range of 0.2 - 0.9 ug/g fresh wt."

In response to the Agency's inquiry as to why there was such great variation for the MON 801 expression, Monsanto states the following in their 7/22/96 facsimile/email message: "The levels of CryI(b) protein in leaves, grain and whole plants of MON 801 have decreased during breeding. The reason for the decrease is not known. The DNA insert in line MON 801 is stable, as demonstrated through Southern blot analysis. The decrease in expression appears to be related to the repeated cycles of inbreeding required to convert the inbred parents. Since the breeding started in 1992, the expression has not increased in any of the approximately 150 hybrids tested to date. There has been no published evidence of transgene expression increasing during breeding." No such decrease has been observed with MON 810 and is therefore not anticipated.

3. MON 801 Data Applicability to MON 810 Progeny

Given the dosing and expression level information now available to the Agency, the MON 801 quail and catfish data are applicable to the MON 810 line progeny since the levels of CryIA(b) are similar.

12727

4. Impacts on Non-Target Organisms

a. Impacts on Non-Target Insect - Honey Bee (Larvae)

B.t.k. HD-1 protein at 20 ppm is practically non-toxic to larval honey bees. An LC_{50} was not possible to calculate since this was a single dose test. Therefore, the NOEL is greater than 20 ppm.

b. Impacts on Non-Target Insect - Honey Bee (Adult)

There were no statistically significant differences among the various treatment and control groups due to the sizable mortality that occurred in all treatments. B.t.k. HD-1 protein at 20 ppm resulted in a mean mortality of 16.2%. Because mortality was observed at the single dose tested, a NOEL could not be determined from this study, but it was less than 20 ppm. 20 ppm was determined to be significantly higher than exposure conditions in the environment.

c. Impacts on Non-Target Insect - Parasitic Hymenopteran

B.t.k. HD-1 protein at 20 ppm is practically non-toxic to *Brachymeria intermedia*. Since this is a single dose study, an LC_{50} cannot be calculated. The NOEL is greater than 20ppm.

d. Impacts on Non-target Insect - Green Lacewing Larvae

B.t.k. HD-1 protein at 16.7 ppm is practically non-toxic to green lacewing larvae after 7 days. The NOEL is greater than 16.7 ppm.

e. Impacts on Nontarget Insect - Lady Beetles

B.t.k. HD-1 protein at 20 ppm is practically non-toxic to lady beetles such as *Hippodamia convergens*. The NOEL is greater than 20 ppm.

f. Impacts on Birds - Northern Bobwhite Quail

No treatment related mortality or differences in food consumption, body weight or behavior occurred in birds fed 50,000 or 100,000 ppm transgenic corn meal derived from Monsanto's MON 80187 corn line (which contains CryIA(b) protein) relative to birds fed corn meal made from parental corn lines which did not express *Bt* toxin.

Although this study utilized Monsanto's MON 801 Bt corn for testing, the test material was considered sufficiently similar to the MON 810 corn grain to bridge the data because of the similarity in CryIA(b) levels.

g. Impacts on Earthworm

The 14-Day LC_{50} value for earthworms exposed to CryIA(b) insecticidal protein derived from *E. coli* in an artificial soil substrate was determined to be greater than 200 mg/kg (ppm), which was the single concentration tested. There were no statistically significant effects at the single dose tested. Therefore, the NOEL is greater than 200 ppm. Although this study was graded supplemental, *Bt* toxins expressed in the corn plant are not expected to generate a toxic effect in the earthworm; therefore, no additional follow-up of this study is required.

h. Impacts on Collembola

Impacts on non-target soil organisms are of interest because of the residual B.t.k. protein that exists in the corn plant at physiological maturity and the potential for incorporation into the soil. Monsanto has submitted a study assessing impacts on Collembola spp., which has been rated as a "supplemental" study due to the form of the test material. The Agency asked for a *Collembola* study using lyophilized leaf extract as the test material subsequent to the initial registration application, but, to date, the registrant has only cited one using purified CryIA(b) toxin derived from *E. coli* as the test substance. Therefore, Monsanto must fulfill this unfulfilled data requirement and submit or cite the required Collembola study.

In the study submitted by Monsanto, purified B.t.k. insecticidal proteins derived from *E. coli* (200 ppm), including CryIA(b) toxin, had no observable toxicological effect on two species of Collembola: *Folsomia candida* and *Xenylla grisea.* The applicant has been informed via Agency letter that this study does not adequately addresses the Agency's non-target soil organism questions because it was conducted with purified *E. coli*-produced B.t.k. protein and not lyophilized leaf extract, as the Agency requested. The rationale for the required study is that there is another study on file that demonstrates toxicity to Collembola, using lyophilized leaf extract as the test material, while control leaf extract did not.

i. Impacts on Channel Catfish

The study "Evaluation of the European Corn Borer Resistant Corn Line MON 801 as a Feed Ingredient for Catfish" was reviewed to determine potential impacts on channel catfish from Monsanto's MON 810 corn lines. Feed per fish, feed conversion ratios, final weight, percentage weight gain and survival were not significantly different between fish fed the control MON 800 diet when compared to those fed the diet containing transgenic corn from the test line MON 801. Body composition data exhibited no significant differences in percentage moisture, fat, or ash, with a higher protein content in the test fish on a dry weight basis. This difference in protein content disappears when one expresses the results on a wet weight basis. Data in this study are consistent with historical controls for catfish grown at the Delta Research and Extension Center.

Although this study utilized Monsanto's *MON 801 Bt* corn for testing, the test material was considered sufficiently similar to the MON 810 corn grain to bridge the results for the data requirement since the levels of CryIA(b) in the MON 801 grain tested were similar to MON

810 levels.

j. Impacts on Aquatic Invertebrates

Due to the potential exposure of aquatic invertebrates to corn pollen containing the B.t. CryIA(b) toxin, this requirement will need to be addressed by the applicant by conducting a *Daphnia magna* study; or by providing adequate rationale for waiver.

k. Impacts on Mammals

Both the scientific literature and the acute oral mouse study results indicate that no toxicity is expected in mammals. Therefore, no further testing on mammals in indicated.

5. Impacts on Endangered Species

A Biological Opinion was issued on December 18, 1986, concerning the possible effect of foliar spray of *Bacillus thuringiensis* subsp. *kurstaki* (*Bt*) on threatened and endangered species. Based on the difference in exposure scenarios between foliar *Bt* spray and *Bt* delta endotoxin expressed in corn plants, EPA believes that the Biological Opinion is not applicable and that reinitiation of consultation is not required.

The primary route of exposure to foliar *Bt* sprays is through either direct application to the crop or as a result of drift from spray or aerial applications thus exposure potential to *Bt* toxin incorporated within the plant is expected to be less than that of foliar sprays.

In comparison, the primary route of exposure to *Bt* delta endotoxin in corn is through ingestion of corn tissue. There are no reports of threatened or endangered insect species feeding on corn plants, thus such species are not expected to be exposed to corn tissue containing the CryIA(b) delta-endotoxin. Corn is widely grown, and above ground feeding damage is easily observed on corn plants due to its morphology and the way it is grown. Consequently, the identities of insects that feed on corn are well established.

Another possible route of exposure is from corn pollen containing the delta-endotoxin that can drift from corn fields. As discussed previously in this section, the applicant has submitted adequate data for representative species to substantiate that the delta-endotoxin is practically nontoxic. The applicant did not submit or cite sufficient toxicity data for Collembola and failed to submit a toxicity study for *Daphnia magna*. There are no threatened or endangered soil invertebrates that are closely related to Collembola. Similarly, there are no threatened or endangered arthropods that are closely related to the *Daphnia magna*.

In addition, EPA does not expect that any threatened or endangered species will be affected by exposure to the delta-endotoxin via weediness or outcrossing to wild relatives or by competition with such entities. Hybrid corn, such as the corn that contains the pesticide at

issue here, cannot exist in the wild nor are there wild relatives that can interbreed with corn in the United States. See section B above for a more detailed discussion of the potential for weediness and outcrossing to wild relatives.

Because EPA expects that threatened or endangered Lepidopteran insects and other species will not be exposed to the *Bt* delta endotoxin, and because the most probable exposure scenario does not appear to affect listed species, EPA believes that this action will have no effect on any threatened or endangered species.

E. Resistance Management

Monsanto has cited Northrup King's resistance management plan to support Monsanto's application for registration of the *Bt* corn product. With this resistance management plan there are two major resistance concerns for the primary target pest, ECB, and a secondary pest, corn earworm (CEW)[also known as the bollworm, sorghum headworm, tomato fruitworm, and soybean pod borer (*Helicoverpa zea* (Boddie))] . Monsanto's *Bt* corn is similar to NK's previously registered *Bt* corn product because they both express the CryIA(b) delta-endotoxin in kernels and silks. CEW prefer to feed upon silks and kernels, although they also feed in the whorl. The CEW can also migrate long distances, can move from corn to cotton and other crops and weed plants, and will move from the South to the North on prevailing winds as the season progresses. Therefore, the Agency believes that there will be similar selection pressure for resistant CEW by Monsanto's *Bt* corn as there is with NK's *Bt* corn. Additional mitigation measures over that required for Ciba and Mycogen *Bt* corn products are recommended to limit the selection pressure for CEW resistance especially in the southern U.S. where cotton production states and cotton growing areas are or could be in close proximity to Monsanto's *Bt* corn. These measures will be discussed more fully below.

1. General Conclusions

Northrup King's plan cited by Monsanto has addressed all of the general elements of a resistance management program, although some only superficially. These elements include pest biology, *Bt* dose deployment, refugia, monitoring for European corn borer (ECB) resistance, susceptible nontarget Lepidoptera pests, cross-resistance, IPM fit, grower education and communication, and development of alternative pesticides with different modes of action.

Based on an analysis of all of the available information submitted by Northrup King and cited by Monsanto, the Agency concludes that the submitted resistance management plan has the necessary workable elements, but additional data must be collected to provide a solid basis for a long-term resistance management plan. Research should be performed, data collected, and modifications to the plan should be implemented within approximately two to four years following the date of this registration.

2. European Corn Borer

The original requirement by the Agency was for an Agency-approved resistance management plan to be implemented within 5 years after initiation of commercial use. Market projections indicate that such market-driven unstructured refugia for ECB should exist during this initial commercialization period. However, *Bt* corn products have already been registered and grown commercially in 1996. As long as all commercial seed corn production is not immediately switched on a local or national level to corn producing CryI delta-endotoxins, unstructured or market-driven refugia should be adequate for approximately 2 to 4 more years because there will be natural refugia between fields (spatial mosaics). In the long term, because of larval mobility and availability of alternate hosts, within-field seed mixes and low usage of *Bt* corn products are not likely to be an effective refugia. Therefore, the Agency has required evaluation of structured refugia (where there are blocks of *Bt* plant-pesticide corn and conventional corn or other host plants to be evaluated as a potential effective refugia) for all of the Bt corn products registered to date and the Agency will include this requirement for this registration.

One of the most critical parts of the resistance management strategy is the high dose strategy coupled with an effective refugia. Evidence provided in the submitted resistance management plan for the high dose expression strategy in ECB is limited and is not necessarily representative of the selective conditions operating in the field. A high dose expression strategy will need to be validated in the field to determine whether the CryIA(b) delta-endotoxin will be produced uniformly at a high enough dose in the field to kill all susceptible individuals including heterozygotes. Without field validation, it is impossible to predict at this time the absolute success of the high dose expression strategy for ECB.

Possible high dose control exists for the first generation ECB on whorl stage corn, but not for later generation(s) on more mature corn plants. More data are needed on toxin expression in various parts of the plant at different stages of plant development. Lower expression as the plant matures and becomes senescent would not necessarily provide season-long high dose control for ECB. Lack of a high dose would mean that ECB would be exposed to sublethal doses of the CryIA(b) delta-endotoxin. This would increase the likelihood for selection of resistance. Impacts of season long-expression need to be addressed in research to refine the resistance management plan for this product.

3. Corn Earworm

The situation is different for corn earworm/cotton bollworm (CEW) resistance management for Ciba Seed and Mycogen *Bt* corn products than for Northrup King and Monsanto *Bt* corn products because of the expression of *Bt* CryIA(b) delta-endotoxin proteins in the kernels and silks of the latter two products. CEW prefer to feed upon silks and kernels, although they also will feed in the whorl early in the season. Therefore, the Agency believes that there will be substantial additional selection pressure for resistant CEW than there is for the *Bt* corn products registered to Ciba Seeds and Mycogen which do not significantly express the CryIA(b) delta-endotoxin proteins in the kernel and silk.

The Northrup King/Monsanto submitted resistance management plan should adequately manage lepidopteran resistance including CEW to the CryI delta-endotoxins produced in field corn in the first 2 to 4 years following commercialization except for cotton production states and cotton growing areas in border states where the selection pressure for the development of CEW resistance will be considerably higher. In these southern areas several factors are important: CEW can have from 3 to 6 generations per year versus 1 to 2 generation in the northern states; CEW can overwinter in cotton producing areas; there is a CEW generation which feeds predominately on corn and the next CEW generation often moves to cotton and other hosts such as vegetable crops; CEW moths can migrate long distances, therefore, each succeeding generation of moths may remain locally or individuals may be carried on wind currents to the northern states; and *Bt* foliar sprays and the *Bt* cotton products are used on cotton and several other crops for CEW control. There is limited, if any use of *Bt* foliar products to control ECB.

The Agency is concerned about the ramifications of CEW/bollworm resistance developing in insect populations that feed on both corn and cotton where corn and cotton acreage is in close proximity. CEW resistance could negatively affect the utility of *Bt* cotton and *Bt* foliar sprays on vegetables and other crops. The result might lead to a loss of the use of *Bt* products which are safer than conventional chemical insecticide alternatives. Although the risk of loss of *Bt* and increased use of chemical insecticides cannot be quantified, the Agency believes this risk is real. There also could be negative impacts on organic farmers from the loss of *Bt*.

Both Monsanto and Northrup King proposed use of their *Bt* corn products in the South contending that there were benefits to be gained by the use of their products in the South. Monsanto proposed to limit the use of their *Bt* corn product to 10% of the field corn grown in the South with no more 10% of the corn being Monsanto's *Bt* corn product in any southern county with 1000 acres or more of cotton. In addition, EPA has been contacted by Texas corn growers regarding the benefits of using these *Bt* corn products to control CEW and thereby reduce infection by secondary fungi which produce mycotoxins in field corn grown for human consumption in the panhandle area of Texas. Records indicate there is very little cotton production in the Texas panhandle.

EPA has considered the risks and benefits of allowing limited acreage of the *Bt* corn products which express in the kernels and silks. EPA has taken into consideration the public comments received, the comments from the potential users, and discussions with technical experts outside of the Agency.

Industry, academia, and government experts have not come to an agreement on the appropriate size and structure for an optimal refugia. A "*Bt* corn consortium" met twice in 1995 (composed of USDA NC-205 (corn stalk borer entomologists), other academicians, and industry) and agreed that a minimum of 5 to 20% refugia was needed, but many opposed stating any figure until more is known. Silk and kernel expression in Monsanto's *Bt* corn will likely increase the selection for CEW resistance especially in cotton-growing areas.

However, based on input from a senior staff entomologist from BEAD, BPPD has determined that the pending full commercial conditional registration for *Bt* corn (which limits use of this corn in Southern cotton growing areas to 100,000 acres and no more than 5% of the corn planted in any county that has than 1000 acres of cotton) would not be likely to significantly impact the potential for corn earworm resistance developing. Further, an additional 100,000 acres in the Southern cotton growing areas under the same limitations also would not be likely to significantly impact the potential for resistance.

EPA has determined that the benefits outweigh the risks of resistance development as long as no more than 100,000 acres of Monsanto's *Bt* corn product is used and no more than 5% of the corn produced in any county with 1000 acres or more of cotton is Monsanto's *Bt* corn product. These limitations also allow for use of *Bt* corn products which are of most benefit (kernel and silk expression to control CEW) to corn producers and the public; it will limit the risk by restricting the amount of this product to areas where the CEW overwinters and cotton dominates over corn, and will allow registrants and researchers to conduct in field testing to determine the best resistance management plan for the long term.

V. TERMS AND CONDITIONS OF THE REGISTRATION

The following listing gives the terms and conditions of the amendment agreed to by Monsanto. The expiration date for the registration was amended to allow the registrant to complete the studies related to ecological effects and resistance management.

1. This registration will automatically expire on midnight April 1, 2001. EPA will reevaluate the effectiveness of Monsanto's resistance management plan before April 1, 2001, and decide whether to convert the registration to a non-expiring registration.

2. This registration is for field corn only.

3. Monsanto will:

   a. unless demonstrated to EPA's satisfaction that alternative resistance management practices are equally or more effective than a structured refugia, develop and submit to EPA a draft plan for "structured" refugia by 8/9/98 and a final plan by 1/31/99;

   b. discuss the development and implementation of the plan and alternative resistance management practices with EPA throughout development and implementation; and

   c. implement an EPA approved "structured" refugia plan or an EPA approved alternative resistance management plan no later than April 1, 2001.

4. Monsanto will monitor for the development of resistance using baseline susceptibility data and/or a discriminating concentration assay when such an assay is available. Monsanto will proceed with efforts to develop a discriminating concentration assay. Monsanto will ensure that monitoring studies are conducted annually to determine the susceptibility of ECB and corn earworm (CEW) populations to the CryIA(b) protein. This resistance monitoring program will be developed to measure increased tolerance to *Bt* corn above the various regional baseline ranges.

Populations of ECB and CEW will be collected from representative distribution areas that contain Monsanto's Bt corn plant-pesticide and monitored/screened for resistance, with particular focus on those areas of highest distribution. The results of monitoring studies will be communicated to the Agency on an annual basis, by January 31 of the year following the population collections for a given growing season.

In addition, Monsanto will instruct its customers (growers and seed distributors) to contact Monsanto (e.g., via a toll-free customer service number) if incidents of unexpected levels of ECB and/or CEW damage occur. Monsanto will investigate and identify the cause for this damage by local field sampling of plant tissue from corn hybrids that contain Monsanto's Bt corn plant-pesticide and sampling of ECB & CEW populations, followed by appropriate *in vitro* and *in planta* assays. Upon Monsanto's confirmation by immunoassay that the plants contain CryIA(b) protein, bioassays will be conducted to determine whether the collected ECB population exhibits a resistant phenotype.

Until such time that a discriminating concentration assay is established and validated by Monsanto, Monsanto will utilize the following to define a confirmed instance of ECB and/or CEW resistance:

> Progeny from the sampled ECB or CEW population will exhibit both of the following characteristics in bioassays initiated with neonates:
>
> a. An LC50 in a standard CryIA(b) diet bioassay that exceeds the upper limit of the 95% confidence interval of the mean historical LC50 for susceptible ECB or CEW populations, as established by the ongoing baseline monitoring program. The source of CryIA(b) crystal protein standard for this bioassay will be *Bacillus thuringiensis* subsp. *kurstaki* strain HD1.
>
> b. > 30% survival and > 25% leaf area damaged in a 5-day bioassay using CryIA(b)-positive leaf tissue under controlled laboratory conditions.

Based upon continued experience and research, this working definition of confirmed resistance may warrant further refinement. In the event that Monsanto finds it appropriate to alter the criteria specified in the working definition, Monsanto must obtain Agency approval in establishing a more suitable definition.

5. Monsanto will report all instances of confirmed ECB & CEW resistance, as defined above, to the Agency within 30 days. Upon identification of a confirmed instance of ECB resistance Monsanto will take the following immediate mitigation measures:

a. notify customers and extension agents in the affected area,

b. recommend to customers and extension agents in the affected area the use of alternative control measures to reduce or control the local ECB population, and

c. recommend to customers and extension agents in the affected area that crop residues be incorporated into the soil following harvest, to minimize the possibility of overwintering of ECB.

Within 90 days of a confirmed instance of ECB and/or CEW resistance, as defined above, Monsanto will: (1) notify the Agency of the immediate mitigation measures that were implemented, and (2) submit to the Agency a proposed long-term resistance management action plan for the affected area, (3) work closely with the Agency in assuring that an appropriate long-term resistance management action plan for the affected area is implemented, and (4) implement an action plan that is approved by EPA and that consists of some or all the following elements, as warranted:

a. Informing customers and extension agents in the affected area of ECB and/or CEW resistance,

b. Increasing monitoring in the affected area, and ensuring that local ECB or CEW populations are sampled on an annual basis,

c. Recommending alternative measures to reduce or control ECB or CEW populations in the affected area,

d. Implementing a structured refuge strategy in the affected area based on the latest research results. The implementation of such a strategy will be coordinated by the Agency with other registrants.

e. If the above elements are not effective in mitigating resistance, Monsanto will voluntarily cease sale of all corn hybrids that contain Monsanto's Bt corn plant-pesticide in the county experiencing loss of product efficacy and the bordering counties until an effective local management plan approved by EPA has been implemented. During the voluntary suspension period, Monsanto may sell and distribute in these counties only by obtaining EPA approval to study resistance management in those counties. The implementation of such a strategy will be coordinated by the Agency with other registrants.

If EPA agrees that an effective resistance management plan has been implemented which mitigates resistance, Monsanto can resume sales in the affected county(ies).

6. Monsanto will maintain a (confidential) database to track sales (units and location) of its Bt corn on a county-by-county basis. Monsanto will provide annually, on a CBI basis, sales data for each state indicating the number of units of corn hybrids that contain Monsanto's Bt corn plant-pesticide that were sold. As part of the overall sales report, Monsanto will provide a listing of an estimate of the acreage planted with such states and counties with sales limitations detailed in item 10 of the pre-acceptance letter. This information will be provided by January 31 of the year following each growing season.

7. Monsanto will provide grower education. Monsanto will agree to include an active partnership with such parties as: university extension entomologists and agronomists, consultants, and corn grower groups. Monsanto will implement a grower education program (in part, as requested by Monsanto, through the Grower Agreement setting forth any resistance management requirements) directed at increasing grower awareness of resistance management, in order to promote responsible product use. As specific resistance management recommendations are developed (e.g., as a result of ongoing research or experience) these will be incorporated, as appropriate, into the various grower communication and educational media. Monsanto will inform the Agency as it develops, implements, and refines its communication strategies. In addition to grower communication vehicles, Monsanto will also develop a Grower Guide consistent with the terms and conditions of this registration, to be distributed to all customers, that will include current information regarding resistance management and integrated pest management.

8. Monsanto will develop a resistance management program that is acceptable to EPA and that includes the research and model development and testing specified in paragraph 8 a through f. Monsanto will confer with the EPA as Monsanto develops various aspects of its resistance management research program. Monsanto agrees, as a condition of this registration, to submit annually progress reports on or before January 31st each year on the following areas as a basis for developing a long-term resistance management strategy which include:

a. Monsanto must submit by January 31, 1997, available research data on CEW relative to resistance development and Monsanto's plans for producing resistance predictive models to cover regional management zones in the cotton belt based on *Helicoverpa zea* biology and cotton, corn, soybeans, and other host plants. These models must be field tested in the acreage allowed in item 10 of this letter and must be modified based on the field testing performed during the period of the conditional registration. EPA might modify the terms of the conditional registration based upon the field testing validation of the model and might require refugia in the future. EPA notes that there is some scientific work and even some models for H. zea on other crops in at least NC and TX

that could be used for reference. EPA wants to be in close communication with Monsanto as the model development and testing is ongoing. The requirement for development of resistance predictive models may be modified if Monsanto provides the results of research that demonstrates resistance to CEW would have no significant impact on the efficacy of foliar Bt products and other Bt crops. Actual usage data of Btk on crops to control specific pests as well as successes and failures and field validated research would be necessary to support such a waiver request.

b. ECB pest biology and behavior including adult movement and mating patterns, larval movement, survival on silks, kernels, and stalks, and overwintering survival and fecundity on non-corn hosts. A combination of a comprehensive literature review and research can fulfill this condition.

c. The feasibility of "structured" refuge options for ECB including both "block" refugia, "50-50 early/late season patchwork;" research needs to be done in both northern and southern areas on ECB as well as CEW.

d. Development of a discriminating concentration (diagnostic concentration) assay for field resistance (field screening) for ECB, CEW and other Lepidoptera pests of corn. Specific sampling locations will be established in each state to determine if increases in *Bt* toxin tolerance are occurring before crop failures develop. Increased tolerance levels need to be identified before field failure occurs. In monitoring for tunneling damage, the number of trivial tunnels may be less indicative of resistance development than the total extent of tunneling damage (e.g. length of tunnels). The extent of tunneling damage must be monitored as well as the number of tunnels.

e. Effects of corn producing the CryIA(b) delta endotoxin on pests other than ECB, including but not limited to CEW, fall armyworm, and the stalk borer complex.

f. The biology of ECB resistance including receptor-mediated resistance and its potential effect on population fitness, as well as the effects on insect susceptibility to other Cry proteins. Possible high dose control exists for the first generation ECB in whorl stage, but not for later generation(s) on more mature corn plants. More data are needed on toxin expression in various parts of the plant at different stages plant development in regard to ECB, CEW and other secondary pests of corn (i.e. stalk borer complex, fall armyworm, and S.W. corn borer).

9. Monsanto will ensure that in the combined states of Alabama, Arkansas, Georgia, Florida, Louisiana, North Carolina, Mississippi, South Carolina, Oklahoma (only the counties of

Bryan, Caddo, Canadian, Garvin, and Grady), Tennessee (only the counties of Carroll, Chester, Crockett, Fayette, Franklin, Gibson, Hardeman, Hardin, Haywood, Henderson, Lake, Lauderdale, Lawrence, Lincoln, McNairy, Madison, Obion, Rutherford, Shelby, and Tipton), Texas, Virginia (only the counties of Greenville, Isle of Wright, Northampton, Southampton, Sussex, Suffolk) and Missouri (only the counties of Butler, Dunkin, Mississippi, New Madrid, Pemiscot, Scott, Stoddard) that the combined sale of this plant-pesticide in all the above states will not exceed the amounts required to plant 100,000 acres per anum. Further, Monsanto will ensure that for the states and counties listed above that the amount sold will result in no more than 5% of the corn planted in any county with more than 1000 acres of cotton. Per item 6 of this letter Monsanto will report all sales of this product by Monsanto or its distributors annually to EPA no later than January 31st of the following year.

10. *Collembola* and *Daphnia magna* studies must be submitted by 5/14/97 for this active ingredient as already required in the seed increase registration.

11. Although the Agency is not requiring grower agreements as a term or condition of this registration, Monsanto has decided to comply with the terms or conditions of this registrations by use of a grower agreement. Distributors (supplemental registrants under 40 CFR 152.132), as agents of Monsanto, are bound by the terms and conditions of this registration.

VI. RATIONALE FOR RECOMMENDATION

Pursuant to FIFRA section 3(c)(7)(B), EPA may conditionally amend the registration of a pesticide to permit an additional use if two criteria are fulfilled: 1) the applicant has submitted satisfactory data pertaining to the proposed new use; and 2) amending the registration in the manner proposed by the applicant will not significantly increase the risk of any unreasonable adverse effect. BPPD believes that both these criteria have been fulfilled.

Additional Use Registration Under FIFRA 3(c)(7)(B)

The applicant has submitted or cited data to satisfy the first criterion for conditional registration under FIFRA 3(c)(7)(B). Monsanto has submitted or cited satisfactory data pertaining to the proposed additional use of the product in field corn, including the incremental risks that would result from approval of the applications. BPPD believes that the applicant has provided enough data to characterize the incremental risks associated with the development of resistance resulting from approval of their applications.

Although the data with respect to this particular new use is satisfactory, it is not sufficient to support an unconditional amendment under FIFRA 3(c)(5). Additional data is necessary to evaluate the risk posed by the development of resistance to Cry delta-endotoxins that is associated with generic use of these products. As discussed in more detail in section V above, the introduction of these products for any wide-scale use poses the risk that pests, such as the ECB and CEW, will develop resistance to many different *Bt* microbial pesticides that are used

on a wide variety of crops. BPPD believes that the applicants have submitted sufficient data to allow the Agency to determine that the applicants' plans to manage this risk will be workable for 2 to 4 years following initial commercial use of these products. Additional data, however, are necessary to determine how to effectively reduce the risks associated with resistance beyond that initial period. Consequently, BPPD recommends imposing the data requirements specified earlier in this Decision Document in section V.

BPPD also believes that the second criterion for a FIFRA 3(c)(7)(B) conditional registration has been fulfilled because it appears that the proposed additional use does not "significantly increase the risk of any unreasonable adverse effect." In essence, FIFRA requires a determination that the proposed additional use of these products differs from the current use only in ways that (1) would not modify the risk/benefit ratio so as to cause unreasonable adverse effects taking into account the economic, social, and environmental costs and benefits of the additional use as restricted by the terms and conditions of registration; and (2) would not result in pesticide chemical residues that result from use of the pesticide in or on food that cause a human dietary risk inconsistent with the standards of section 408 of FFDCA.

The proposed new use of this product poses the risk of the development of multiple- and cross-resistance in certain Lepidoptera pests on corn. As a result pests could develop resistance to certain microbial *Bt* pesticides that are applied to both field corn and other crops and reduce the utility of such products. Microbial *Bt* pesticides are critical for many organic farming programs and are identified by the Agency as a safer pest control method than many chemical pesticide alternatives. The Agency further recognizes that microbial *Bt* pesticides have low dietary, worker, and ecological risks when compared to the more hazardous alternatives that might replace the microbial *Bt* pesticides should resistance develop. The microbial pesticides also are important components in many IPM programs for a variety of crops and the loss of such pesticides could cause growers to substitute more harmful pest control agents.

This registration may provide substantial benefits to corn producers in the form of increased yields of field corn resulting from the control of damage caused by the ECB and possibly other corn pests.

The presence of a system to insure that the resistance management provisions of the registration apply to distributors (for example the use of Grower Agreements between the registrant and the growers) is an essential part of EPA's decision to grant this registration. Monsanto has decided to satisfy the terms or conditions of registration including insect resistance management, sales and distribution, and reporting requirements, by entering into binding agreements with growers. Absent such a system, EPA would not have granted the registration.

The risks are substantial and BPPD has concluded that the risks, if unchecked, would outweigh the benefits of the proposed new use. However, the terms and conditions of the amendment that are recommended in Section V of this Decision Document would mitigate the

risks from pesticide resistance sufficiently so that the risks of the proposed amendment would not significantly increase the risks of unreasonable adverse effects. The registration will expire automatically in April 1, 2001. At that time EPA can re-evaluate whether the registrant has an effective resistance management plan. In the interim, the registrant must conduct a grower education program directed at increasing grower awareness of resistance management; conduct monitoring to help detect the development of resistance in ECB and CEW to the CryIA(b) delta-endotoxin and stop selling the product in areas where the registrant has detected resistance to its own product; conduct research to determine how to develop an effective long-term resistance management plan; and implement an EPA-approved structured refugia system. Planting in southern cotton regions is prohibited beyond the 100,000 acre limit.

FIFRA section 2(bb) defines "unreasonable adverse effects on the environment to include "a human dietary risk from residues that result from a use of a pesticide in or on any food inconsistent with the standard under section 408 of the [FFDCA]." Under FFDCA section 408, a pesticide chemical residue is unsafe unless either a tolerance is in effect for the residue and the residue is within the tolerance limit or an exemption from the requirement of a tolerance is in effect for the residue. EPA may establish a tolerance or exemption only if it is safe. A tolerance or exemption is safe if "there is a reasonable certainty that no harm will result from aggregate exposure to the . . . residue, including all anticipated dietary exposures and all other exposures for which there is reliable information.." Section 408(b)(2)(C) also requires the Agency to ensure that there is a reasonable certainty that no harm will result to infants and children from aggregate exposure to the residue. This includes dietary exposures and exposures from drinking water and non-occupational sources, but does not include occupational exposure. Finally , section 408(b)(2)(D) specifies factors EPA is to consider in establishing an exemption.

Consistent with FIFRA and FFDCA section 408, EPA has reviewed the available scientific data and other relevant information in support of this registration and has concluded that human dietary risk that result from use of residues of this pesticide product in or on any food is consistent with the FFDCA section 408 standard.

VII. RECOMMENDATION

The submitted data in support of this registration under section 3(c)(7)(B) of the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) have been reviewed and determined to be adequate. Studies and information regarding resistance management are included in the terms, conditions, and limitations of this registration. Amending the existing registration will not significantly increase the risk of adverse effects to man or the environment, for example as a result of exposure to non-target organisms or from the potential for the development of resistance. Furthermore, the benefits of the new use pattern have been well established and the terms, conditions, and limitations imposed by this registration mitigate the risks posed from the potential pest resistance to *Bt*. Therefore, the potential benefits outweigh potential risks, e.g. from the development of resistance. Finally, human dietary exposure to residues

from use of the product in food is consistent with the standard of FFDCA section 408.

Based on the data submitted and cited by Monsanto and reviewed by OPP staff, Biopesticides and Pollution Prevention Division recommends that Monsanto's plant-pesticide product containing the active ingredient *Bacillus thuringiensis* CryIA(b) delta-endotoxin and the genetic material necessary for its production in field corn be CONDITIONALLY REGISTERED for food and feed use under 3(c)(7)(B) of FIFRA.

In addition, BPPD recommends that use of kernel & silk expressing *Bt* corn for an additional 100,000 acres in the Southern cotton growing areas be approved. Per your guidance, BPPD would, at the time we issue a registration for Monsanto's full commercial use of *Bt* corn, concurrently issue a pre-acceptance letter to Northrup King for an amendment to their full commercial use registration to allow use in Southern cotton growing areas of 100,000 acres with their *Bt* corn constituting no more than 5% of the corn planted in any county that has than 1000 acres of cotton.

CONCUR:

NONCONCUR:_____

DATE: 12/20/96

27827