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

OFFICE OF PREVENTION, PESTICIDES AND TOXIC SUBSTANCES

JUN 09 2009

MEMORANDUM

SUBJECT: Review of Studies Submitted for WideStrike Insect Resistant Cotton Seed (EPA Reg. No. 68467-3) as a Condition of a Section 3 Registration for Control of Lepidopteran Pests in Cotton; Decision # 370934, DP Barcode # 346371 and 361621, MRIDs 468037-01, 476673-01 and 476673-02; Decision # 397019, DP Barcode # 354503, MRID 474623-01

FROM: Shannon Borges, Biologist
Microbial Pesticides Branch (7511P)
Biopesticides and Pollution Prevention Division
6/9/09

THROUGH: Zigfridas Vaituzis, Ph.D., Senior Scientist
Microbial Pesticides Branch (7511P)
Biopesticides and Pollution Prevention Division
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TO: Denise Greenway, Regulatory Action Leader
Microbial Pesticides Branch (7511P)
Biopesticides and Pollution Prevention Division

WideStrike Insect Resistant Cotton Seed was conditionally registered in 2004. Among the conditions of registration was the requirement that the registrant, Mycogen Seeds/Dow Agrosciences, submit an avian chronic toxicity study and a Tier I nontarget insect study with a minute pirate bug (*Orius insidiosus*). Studies have been submitted to meet the condition of registration, and are reviewed in this memorandum. Summaries of each study are presented below, and Data Evaluation Records are attached.

The registrant submitted a broiler chicken study with cottonseed processed from Widestrike cotton to meet the requirement of submission of an avian chronic toxicity study. Supplemental data requested by the Agency showed that the Cry1Ac and Cry1F proteins in Widestrike cotton seed were not detected in the cottonseed meal used in the test diets. Since the effects of the active ingredients were not actually tested, the study was determined to be unacceptable. Due to the potential effects of gossypol in cotton seeds, the Agency originally stated that a broiler chicken study with corn containing these active ingredients would be acceptable. However, this test material is not available and unprocessed cotton seed cannot be used. Furthermore, since WideStrike was registered, the Agency has begun waiving this data requirement for Bt cotton products because of the effects of gossypol in the seeds. Because of these difficulties with this test, BPPD will no longer require the data to be submitted.

The registrant also made several unsuccessful attempts to conduct a Tier 1 nontarget insect study with *Orius*. A meeting was held in November 2007 in which this issue was discussed, and BPPD agreed to accept alternate information on *Orius* spp. A two-year field study was submitted, and information on *Orius* spp. and other Hemiptera were available, along with data on other nontarget insect species. This study was determined to be supplemental due to some problems related to experimental design; but information contained in this study on Orius and other species is adequate to show no adverse effects on the abundance of several nontarget insect species. Based on this information and other Tier I data submitted on nontarget insects, adverse effects to nontarget insects as a result of the use of WideStrike is not expected. This data requirement has been fulfilled and this condition of registration has been met.

Study Summaries

Study: Nutritional equivalency study of Cry1F/Cry1Ac cottonseed meal: poultry feeding study
MRID: 468037-01
Classification: Unacceptable
Summary: In a 42-day study, newly-hatched broiler chickens were fed ration containing cottonseed meal from seeds expressing Cry1F and Cry1Ac proteins. The study also included broilers fed ration containing cottonseed meal from a related non-transgenic cotton or meal from two commercial sources. At test end, there was no statistically significant difference in mortality or final body weight among any of the groups in the test. The group fed meal containing Cry1F/Cry1Ac had a significantly lower feed conversion ratio than one of the control groups fed commercial cottonseed meal. Supplemental poultry processed fraction data (MRID 476673-02) also showed no significant differences except with the amount of abdominal fat and post-chill leg yield (in which cases the Bt cotton did not differ significantly from the parent line). The presence of Cry1F/Cry1Ac in the prepared diets was described in supplemental data submitted in MRID 476673-01. Based on these supplemental data, no Cry1Ac or Cry1F protein was detectable in the cottonseed meal after processing. Therefore, since the active ingredients were not contained in the test diet, the study does not adequately test the chronic effects of Widestrike for avian wildlife.

Study: Lack of effect of WideStrike™ Insect-Protected Bt Cotton on *Orius* spp.
MRID: 474623-01
Classification: Supplemental
Summary: Field tests were conducted at Winnsboro, LA, and Maricopa, AZ during 2002 and 2003 to compare the nontarget effects of WideStrike cotton [a.i., *Bacillus thuringiensis* var. aizawai Cry1F (Synpro) and the genetic material (from the insert of plasmid pGMA281) necessary for its production in cotton, and *Bacillus thuringiensis* var. kurstaki Cry1Ac (Synpro) and the genetic material (from the insert of plasmid pMYC3006) necessary for its production in cotton] to those associated with conventional cotton pest management. The treatments included WideStrike sprayed with insecticides active against non-lepidopteran cotton pests only, a non-transgenic control cotton (PSC355) sprayed with insecticides active against non-lepidopteran cotton pests only, and PSC355 sprayed with insecticides active against non-lepidopteran and lepidopteran pests. Counts of beneficial arthropods and cotton insect pests were conducted during the growing season using sweep nets, pitfall traps, shake sheets, and whole plant

inspections. The abundance of nontarget arthropods was similar in the WideStrike plots and the PSC355 plots that were sprayed for non-lepidopteran pests only, and the abundance of nontarget arthropods tended to be greater in the WideStrike plots than in the PSC355 plots treated with insecticides active against lepidopterans. This study is not adequately designed to be used for purposes of drawing conclusions about community level or long-term environmental effects for non-target insects, but it does provide evidence that adverse effects resulting from use of Widestrike cotton are unlikely. The study is sufficient to fulfill the nontarget insect data required as a condition of registration for Widestrike cotton.

DATA EVALUATION RECORD

Contractor Primary Reviewers:	**E. Lewis, A. Armstrong, Oak Ridge National Laboratory**
EPA Secondary Reviewer:	**Shannon Borges**
EPA Peer Reviewer:	**Zigfridas Vaituzis**

STUDY TYPE:	Poultry Feeding Study (Nonguideline)
MRID NO:	46803701
DP BARCODE:	DP346371
DECISION NO:	370934
SUBMISSION NO:	799398
TEST MATERIAL:	Meal produced from WideStrike Insect Resistant Cotton Seed (a.i., *Bacillus thuringiensis* var. aizawai Cry1F (Synpro) and the genetic material (from the insert of plasmid pGMA281) necessary for its production in cotton and *Bacillus thuringiensis* var. kurstaki Cry1Ac (Synpro) and the genetic material (from the insert of plasmid pMYC3006) necessary for its production in cotton
STUDY NO:	2003-DOW-01-B
SPONSOR:	Dow AgroSciences, LLC, Indianapolis, IN 46268
TESTING FACILITY:	Solution BioSciences, Inc., Salisbury, MD
TITLE OF REPORT:	Nutritional Equivalency Study of Cry1F/Cry1AC Cottonseed Meal: Poultry Feeding Study
AUTHOR:	McNaughton, J.L.
STUDY COMPLETED:	December 11, 2003
CONFIDENTIALITY CLAIMS:	None
GOOD LABORATORY PRACTICE:	A signed and dated compliance statement was provided. The authors state that the study was conducted under the GLP standards of 40 CFR Part 160 and 21 CFR Part 58.
STUDY SUMMARY:	In a 42-day study, newly-hatched broiler chickens were fed ration containing cottonseed meal from seeds expressing Cry1F and Cry1Ac proteins. The study also included broilers fed ration containing cottonseed meal from a related non-transgenic cotton or meal from two commercial sources. At test end, there was no statistically significant difference in mortality or final body weight among any of the groups in the test. The group fed meal containing Cry1F/Cry1Ac had a significantly lower feed conversion ratio than one of the control groups fed commercial cottonseed meal. Supplemental poultry processed fraction data (MRID 476673-02) also showed no significant differences except with the amount of abdominal fat and post-chill leg yield (in which cases the Bt cotton did not differ significantly from the parent line). The presence of Cry1F/Cry1Ac in the prepared diets was described in supplemental data submitted in MRID 476673-01. Based on these supplemental data, no Cry1Ac or Cry1F protein was detectable in the cottonseed

meal after processing. Therefore, since the active ingredients were not contained in the test diet, the study does not adequately test the chronic effects of Widestrike for avian wildlife.

CLASSIFICATION: **Unacceptable**.due to lack of Cry1Ac and Cry1F in test material.

Test Material

The test material was cottonseed meal from cotton seeds expressing the Cry1F/Cry1Ac insecticidal crystal proteins. It was received at the testing facility on March 13, 2003, and placed in secure storage.

The untreated control was cottonseed meal from cotton seeds of a related non-transgenic cotton. The reference controls were cottonseed meal from two commercial cotton sources.

Test Methods

The study was conducted to compare the performance of broiler chickens fed ration containing cottonseed meal from seeds expressing Cry1F/Cry1Ac with that of broilers fed ration containing meal from a related non-transgenic cotton or cotton from two commercial sources. The experimental design is given in Table 1.

Commercial-type broiler chickens (*Gallus domesticus*, Cobb x Cobb) were obtained from Mountaire Hatchery, Princess Anne, MD, of the day of hatch (Day 0). The chicks were vent sexed, weighed individually, and 480 birds were randomly placed into 48 sanitized floor pens (five males and five females/pen). The pens were 3 x 4.5 feet and were separated by a wire partition. Each pen contained 2 inches of litter. The pens were maintained in a chicken house under commercial conditions. Ventilation was via a cross-house system and sidewall fans, and forced-air heat was provided. Lighting was continuous.

Table 1. Experimental design

Diet treatment	Test/control substance level	No. of blocks	Broilers/block	Broilers/treatment
Commercial control 1	10% cottonseed meal	12	10 (5M, 5F)	120
Commerical control 2	10% cottonseed meal	12	10 (5M, 5F)	120
Related non-transgenic control	10% cottonseed meal	12	10 (5M, 5F)	120
Test material (Cry1F/Cry1Ac)	10% cottonseed meal	12	10 (5M, 5F)	120

Data from p. 7, MRJD 46803701

Diets were formulated at the testing facility to contain 10% of the test or reference cottonseed meals. The basal diet was a commercial-type ration (Solution BioSciences Feeds, Salisbury, MD) that met the National Research Council Nutrient Recommendations, and consisted of starter diet until Day 21 and grower diet thereafter. A vitamin and mineral premix was added to the basal diet, but no medications were added. The cottonseed meals and the prepared diets were stated to be analyzed by the study sponsor for moisture, protein, total fat, ash, carbohydrates, calories, free gossypol, total gossypol, acid detergent fiber, neutral detergent fiber, crude fiber, calcium, copper, iron, magnesium, manganese, molybdenum, phosphorus, potassium, sodium, zinc, sulfur, aspartic acid, threonine, serine, glutamic acid, proline, glycine, alanine, cystine, valine, methionine, isoleucine, leucine, tyrosine, phenylalanine, histidine, lysine, arginine, and trypophan. All analyses were stated to be within specification. These data were were submitted in a separate report (MRID

476673-01). The prepared diets were added to the feed trough in each pen. The diet replacement interval was not specified in MRID 46803701.

Beginning on Day 0, the broilers were observed three times/day for mortality, behavior, and signs of toxicity. Individual body weight was recorded on Days 0, 21, 28, and 42, and body weight gain was calculated for Days 0-21, 0-28, and 0-42. Food consumption/pen was calculated for Days 0-21, 0-28, and 0-42 by measuring the amount of food added and removed from each pen. Feed conversion was calculated by dividing the total food consumption/pen by the total body weight gain/pen. At test end, four males and four females from each pen were processed, and weight and yield data were recorded.

Treatment means were compared using ANOVA. Means were further separated using the least significant difference. A significance level of 5% was used in all statistical tests.

Results Summary

Results for the grower phase (Days 0-28) are summarized in Table 2. The feed conversion ratio for the test material group was significantly lower than for the commercial control 1 group. The average body weight gain for the test material group was significantly higher than that of the commercial control 1 group. There was no significant difference in mortality among the groups.

Table 2. Performance averages for grower phase (Days 0-28)

Criteria	Commercial control 1	Commercial control 2	Related non-transgenic control	Test material (Cry1F/Cry1Ac)
Initial weight (g)	51.44 a	51.47 ab	51.81 ab	51.11 b
Final weight (g)	1008.77 b	1026.25 ab	1027.66 ab	1032.04 a
Feed conversion ratio (corrected)[1]	1.500 b	1.479 ab	1.480 ab	1.455 a
Average weight gain (g)	957.32 b	974.79 ab	975.86 ab	980.93 a
Mortality (%)	0.833 a	0.000 a	1.667 a	0.000 a

Data from p. 15, MRID 46803701

Means within a row without a common letter are significantly different ($p \leq 0.05$ by least significant difference)

[1]Corrected for mortality

Results at test end are summarized in Table 3. The feed conversion ratio for the test material group was significantly lower than for the commercial control 1 group. There was no significant difference in mortality or body weight gain among the groups.

Table 3. Performance averages for grower plus finisher phases (days 0-42)

Criteria	Commercial control 1	Commercial control 2	Related non-transgenic control	Test material (Cry1F/Cry1Ac)
Initial weight (g)	51.44 a	51.47 ab	51.81 ab	51.11 b
Final weight (g)	1904.21 a	1920.92 a	1924.30 a	1926.16 a
Feed conversion ratio (corrected)[1]	1.858 b	1.845 ab	1.846 ab	1.819 a
Average weight gain (g)	1852.766 a	1869.455 a	1872.496 a	1875.054 a
Mortality (%)	0.833 a	0.833 a	1.667 a	0.833 a

Data from p. 16, MRID 46803701

Means within a row without a common letter are significantly different ($p \leq 0.05$ by least significant difference)

[1]Corrected for mortality

Additional data were submitted on the processed fraction results (MRID 476673-02) and the compositional analysis of the cottonseed meal (MRID 476673-01).

Results from the analyses of processed fraction data show no significant differences between the group receiving cottonseed meal from the Widestrike transgenic line and the non-transgenic parent and commercial reference lines. The only exceptions were differences seen in abdominal fat yield and pre-chill leg yield weight. However, these differences existed between the commercial control 2 and all other treatments, and do not represent any real difference resulting from the test diet.

The compositional analysis showed that gossypol levels were low in all of the treatments after processing. Free gossypol (reported as percentage of fresh weight) was determined to be 0.048%, 0.112%, 0.051%, and 0.049% for commercial control 1, commercial control 2, the non-transgenic parent, and the test material, respectively. The analyses for the Cry1Ac and Cry1F proteins in the diets showed that these proteins were not detectable in the cottonseed meal processed from any of the cotton seeds used in this study in both the starter and grower diets.

Study Author's Conclusions

The study author concluded that there was no significant difference in mortality or final body weight among any of the groups in the test. The test material group had a significantly lower feed conversion ratio than one of the control groups.

EPA Reviewer's Conclusion

The study does show no effect to the parameters measured between diets made of cottonseed meal processed from Widestrike cotton seeds and those of the parent line and two commercial cotton varieties. However, the Cry1F/Cry1Ac test diet contained none of the active ingredients, so the study did not actually test their effects. This study is classified as unacceptable, and cannot be used to determine the hazard of these proteins to avian wildlife.

DATA EVALUATION RECORD

Contractor Primary Reviewers:	**E. Lewis, A. Armstrong, Oak Ridge National Laboratory**
EPA Secondary Reviewer:	**Shannon Borges**
EPA Peer Reviewer:	**Zigfridas Vaituzis**

STUDY TYPE: Nontarget Insect Testing (885.4340)

MRID NO: 47462301

DP BARCODE: DP354503

DECISION NO: 397019

SUBMISSION NO: 831853

TEST MATERIAL: WideStrike Insect Resistant Cotton Seed (a.i., *Bacillus thuringiensis* var. aizawai Cry1F (Synpro) and the genetic material (from the insert of plasmid pGMA281) necessary for its production in cotton and *Bacillus thuringiensis* var. kurstaki Cry1Ac (Synpro) and the genetic material (from the insert of plasmid pMYC3006) necessary for its production in cotton)

STUDY NO: 071100

SPONSOR: Dow AgroSciences, LLC, Indianapolis, IN 46268

TESTING FACILITY: Dow AgroSciences, LLC, Regulatory Laboratories – Indianapolis Lab, 9330 Zionsville, Road, Indianapolis, IN 46268

TITLE OF REPORT: Lack of Effect of WideStrike Insect-Protected *Bt* Cotton on *Orius* spp.

AUTHORS: Storer, N.P., and R.A. Herman

STUDY COMPLETED: June 25, 2008

CONFIDENTIALITY CLAIMS: None

GOOD LABORATORY PRACTICE: A signed and dated compliance statement was provided. The study was does not meet the definition of a GLP study as it appears in 40 CFR Part 160.

STUDY SUMMARY: Field tests were conducted at Winnsboro, LA, and Maricopa, AZ during 2002 and 2003 to compare the nontarget effects of WideStrike cotton [a.i., *Bacillus thuringiensis* var. aizawai Cry1F (Synpro) and the genetic material (from the insert of plasmid pGMA281) necessary for its production in cotton, and *Bacillus thuringiensis* var. kurstaki Cry1Ac (Synpro) and the genetic material (from the insert of plasmid pMYC3006) necessary for its production in cotton] to those associated with conventional cotton pest management. The treatments included WideStrike sprayed with insecticides active against non-lepidopteran cotton pests only, a non-transgenic control cotton (PSC355) sprayed with insecticides active against non-lepidopteran cotton pests only, and PSC355 sprayed with insecticides active against non-lepidopteran and lepidopteran pests. Counts of beneficial arthropods and cotton insect pests were conducted during the growing season using sweep nets, pitfall traps, shake sheets, and whole plant inspections. The abundance of nontarget arthropods was similar in the WideStrike plots and the PSC355 plots that were sprayed for non-lepidopteran pests only, and the abundance of nontarget arthropods tended to be greater in the WideStrike plots than in the PSC355 plots treated with insecticides active against lepidopterans. This study is not adequately designed to be regarded as a long-term or rigorous examination of field effects, but it does provide evidence that adverse effects resulting from use of Widestrike cotton are unlikely. The study is sufficient to fulfill the nontarget insect data required as a condition of registration for Widestrike cotton.

CLASSIFICATION: **Supplemental**

Test Material

WideStrike cotton (a.i., *Bacillus thuringiensis* var. aizawai Cry1F (Synpro) and the genetic material (from the insert of plasmid pGMA281) necessary for its production in cotton, and *Bacillus thuringiensis* var. kurstaki Cry1Ac (Synpro) and the genetic material (from the insert of plasmid pMYC3006) necessary for its production in cotton.

Background

As a condition for registration of WideStrike cotton (EPA Reg. No. 68467-3), the Agency requested that the registrant submit a maximum hazard dose laboratory study using a minute pirate bug (*Orius insidiosus*). The registrant states that attempts to develop methods for a feeding assay and transfer them to experienced laboratories have been unsuccessful. Each of four attempts over 18 months resulted in high negative control mortality (MRID 47437601). Furthermore, commercial insectaries have not been able to reliably supply quality insects. The registrant has therefore instead submitted a nontarget organism field study that included *Orius* and other arthropods.

Test Methods

Field tests were conducted at Winnsboro, LA, and Maricopa, AZ during 2002 and 2003 to compare the nontarget effects of WideStrike to those of conventional cotton pest management. The treatments included WideStrike and a non-transgenic control cotton (PSC355), each sprayed with insecticides active against cotton pests, but not sprayed with insecticides active against lepidopterans, and PSC355 sprayed with insecticides active against cotton pests, including lepidopterans (Table 1).

Table 1. Treatment descriptions	
Treatment	**Description**
WideStrike (NL)	WideStrike cultivar PHY440W, untreated for lepidopteran pests. Insecticides active against non-lepidopterans were used when required to protect plant health.
PSC355 (NL)	Conventional cotton cultivar (recurrent parent of PHY440W), untreated for lepidopteran pests. Insecticides active against non-lepidopterans were used when required to protect plant health.
PSC355 (S)	Conventional cotton cultivar (recurrent parent of PHY440W) treated for Lepidoptera and other pests as needed following local extension-recommended practices.

Data from p. 30, MRID 47462301

In 2002, the test plots at Winnsboro were 2667 ft^2 (16, 50-foot rows with a 40-inch row spacing). Plots were separated by four rows planted with a mixture of mustard and pigweed. At Maricopa, plots were 6400 ft^2 (24, 80-foot rows with a 40-inch row spacing). Plots were separated by four buffer rows of bare ground. In 2003, the plots were planted at the same location, but the sizes were expanded to one acre (64, 200-foot rows, 40-inch row spacing) at Winnsboro and 0.5 acre at Maricopa. The Winnsboro plots were separated by four rows of non-*Bt* corn. Plots at both locations were laid out in a randomized complete block, with two or three replicates of each treatment at each location each year. Sampling was conducted in the center of each plot to minimize edge effects and the effects of insect dispersal.

Counts of beneficial arthropods and cotton insect pests were conducted using several methods. At Winnsboro in 2002, sampling was by sweep nets and shake sheets, together with visual examination of leaves, squares, bolls, and white flowers. In 2003, yellow sticky traps and pitfall traps were added, and visual examination of whole plants (aerial structures) replaced examination of the separate tissues. At Maricopa in 2002, sweep nets and blue sticky traps were used; in 2003, pitfall traps were added. Additional details for the 2002 tests were previously submitted in MRID 45808419.

MRID 47462301 provides data on sampling dates only for the Winnsboro site in 2003. The insecticide treatments and Winsboro site sampling dates are given in Tables 2 and 3, respectively.

In 2003 at Winnsboro, sweep net sampling consisted of sweeping the canopy in 10 sets of 10 motions/plot and combining into one sample/plot. Shake sheet sampling consisted of collections at four sites/plot and combining into one sample/plot. Four sticky trap sites/plot were randomly established about one week pre-bloom. Yellow traps were set weekly by suspending them directly above the crop canopy for 24 hours. The height was adjusted as the plants grew. Eight pitfall traps consisting of a cup containing one inch of ethylene glycol buried to the rim were set in each plot prior to planting and sampled approximately weekly during the growing season. A cover over the trap deflected rainfall. The cups were removed after 24 hours. Plant sampling consisted of a minimum of three structures/plant on 20 adjacent plants/plot, combined into one sample/plot for each sampling date. Sampling dates are given in Table 3.

Table 2. Insecticide treatments (2003)			
Winnsboro			
Date	**Plots treated**	**Insecticide active ingredient**	**Rate (lb a.i./acre)**
7/03/03	All	Thiamethoxam	0.047
7/16/03	All	Dicrotophos	0.4
7/24/03	PSC355 (S)	Lambda-cyhalothrin Emamectin benzoate	0.03 0.0075
8/05/03	All	Thiamethoxam	0.047
8/05/03	PSC3551 (S)	Lambda-cyhalothrin Spinosad	0.03 0.06
8/22/03	PSC355 (S)	Lambda-cyhalothrin Spinosad	0.03 0.06
Maricopa			
7/31/03	All	Pyriproxyfen	0.86
8/8/03	All	Acetamiprid Acephate	0.1 0.75
8/22/03	All	Oxamyl	1.0
8/29/03	All PSC355 (S)	Buprofezin Cyfluthrin	0.35 0.05
9/2/03	All	Acephate	0.9
9/5/03	PSC355 (S)	Lambda-cyhalothrin	0.04
9/10/03	PSC355 (S)	Lambda-cyhalothrin	0.04
9/15/03	PSC355 (S)	Lambda-cyhalothrin	0.04
9/19/03	PSC355 (S)	Lambda-cyhalothrin	0.04
9/30/03	All	Acetamiprid	0.1

Data from p. 149, MRID 47462301

Table 3. Sampling dates for each sampling method, Winnsboro, LA, 2003				
Pitfall traps	Plants	**Shake sheets**	**Sweep nets**	**Sticky traps**
5/28/03				
6/3/03				
6/10/03				
6/17/03	6/19/03	6/19/03	6/19/03	
6/24/03	6/24/03	6/24/03	6/24/03	
7/1/03	6/30/03	6/30/03	6/30/03	7/2/03
7/8/03	7/7/03	7/7/03	7/7/03	7/7/03
7/15/03	7/14/03	7/15/03	7/15/03	7/15/03
7/22/03	7/21/03	7/21/03	7/21/03	7/21/03
7/31/03	7/29/03	7/29/03	7/29/03	7/29/03
8/5/03	8/4/03	8/4/03	8/4/03	8/5/03
8/12/03	8/11/03			
8/19/03	8/18/03			
8/26/03				
9/3/03				
9/9/03				

Data from p. 47, MRID 47462301

For the 2003 Winnsboro data, ANOVA was conducted on the number of each morphotype (a unique species or lifestage) collected. When the F-test for treatment effect was significant, separation of means was conducted using Fishers protected LSD (α=0.05) to determine whether there were differences between the PSC355 (S) and PSC355 (NL) treatments (spray effect), or between the WideStrike (NL) and PSC355 (NL) treatments (*Bt* effect).

For the 2003 Maricopa data, morphotype frequency and seasonal cumulative counts of the more abundant morphotypes were examined. Repeated measures ANOVAs were conducted using PC-SAS and or JMP 4.0, and morphotypes with a significant treatment effect were further examined using Tukey's HSD ($p \leq 0.05$) and/or orthogonal contrasts ($p \leq 0.10$). Redundancy analyses were conducted using CANOCO 4.0, and principle response curves were constructed for each sampling method.

Results Summary

Data for the 2002 tests were previously submitted in MRID 45808419 (unavailable to the reviewer). According to MRID 47462301, the WideStrike plots at Winnsboro in 2002 showed significantly higher seasonal survey counts than the PSC355 (NL) control for the combined predatory Heteroptera from *Geocoris, Orius, Nabis, Podisus* and family Reduviidae, as well as for lady beetle adults in leaf sampling. Boll and white flower samples contained significantly more *Orius* in the PSC355 (NL) control than in the WideStrike (NL) and PSC355 (S) control groups. The study author states that the latter effect may have been due to the reduction in prey (lepidopteran larvae) in the WideStrike and PSC355 (S) plots. There were no major nontarget effects of WideStrike on the arthropods examined in the sweep net or sticky trap samples.

At Maricopa in 2002, the number of green lacewing larvae was stated to be significantly higher in the WideStrike (NL) plots than in the PSC355 (NL) plots, and both treatments had more green lacewing larvae than the PSC355 (S) plots. There were no major nontarget effects of WideStrike on the arthropods in the sweep net samples or sticky traps. Specific data for the 2002 results at both locations were not provided in MRID 47462301.

The 2003 results for Winnsboro are provided in Tables 4-8. Compared to the Control (NL) plots, the WideStrike (NL) plots had significantly lower counts for "other Hemiptera" in pitfall traps (Table 3); however, the study author noted that pitfall traps are not a reliable method for sampling Hemiptera. Significantly more brown lacewings were caught in shake sheets from the WideStrike (NL) plots than from the Control (NL) plots (Table 5), but this was likely not treatment-related since the Control (S) plots contained the same number of brown lacewings as the WideStrike (NL) plots.

Table 4. Cumulative mean of target and nontarget arthropods/plot for pitfall traps Winnsboro (2003)					
Group	**Common name**	**PSC355 (S)**	**PSC355 (NL)**	**WideStrike (NL)**	**LSD[1]**
Neuroptera	Green lacewing	0.333	0.000	0.333	--[2]
	Lacewing (L)[3]	0.000	0.333	0.333	--
Coccinellidae	Lady beetle (A)[3]	1.333	2.333	0.000*	2.224
	Lady beele (L)	2.667	3.333	0.333	3.316
	Scymnus spp.	0.667	0.667	0.000	--
Predatory Hemiptera	Damsel bug	0.000	0.000	0.333	--
	Spiders	79.000	130.000	72.333	96.982
Herbivorous Hemiptera & Homoptera	Aphids	0.333	2.000	1.667	1.510
	Brown stink bug (A)	0.000	0.333	0.333	--
	Tarnished plant bug (A)	0.000	0.667	0.333	--
	Cotton fleahopper (A)	8.667	8.667	13.667	8.208
	Other Hemiptera	12.667	15.667	5.333*	4.027
	Other Homoptera	11.667	10.667	6.000	10.347
Hymenoptera	Parasitic hymenoptera	1.000	0.000	0.333	--
	Ants	7.000	9.333	8.333	4.418
	Other	4.667	4.667	7.667	4.301
Diptera	Diptera	34.000	25.333	22.667	15.787
Orthoptera	Grasshoppers	0.667	1.667	0.000	--
	Crickets	14.333	13.333	14.333	8.162
Other Coleoptera	Sap beetles	26.000	20.333	14.000	15.256
	Tiger beetles	34.000	44.667	45.333	21.563
	Carabids	54.333	47.333	57.000	22.064
	Scarabs	61.667	63.667	52.333	17.872
	Click beetles	1.333	2.667	3.667	3.208
	Weevils	0.667	1.000	0.000	--
	Rove beetles	1.667	0.000	0.333	--
	Other	4.333	4.000	5.333	6.155
Lepidoptera	Saltmarsh caterpillar	5.333*	12.667	1.000*	5.133
	Bollworm/tobacco budworm	3.000	1.000	1.333	2.190
	Other larvae	1.667	0.667	1.667	1.874
	Armyworm (A)	0.333	0.000	0.000	--
	Yellowstriped armyworm (A)	0.000	0.667	0.000	--
	Sum of all Lepidoptera	10.333	15.000	4.000*	5.878

Data from pp. 48-49, MRID 47462301

[1]Least significant difference, calculated only for those morphotypes averaging at least one individual/plot

[2]-- Statistical comparison not valid due to low numbers (<1/plot)

[3] A = adults, L = larvae

*Significantly different from Control (S), Control (NL) (Fisher's protected LSD, p≤0.05)

Table 5. Cumulative mean of target and nontarget arthropods/plot for whole plants Winnsboro (2003)					
Group	**Common name**	**PSC355 (S)**	**PSC355 (NL)**	**WideStrike (NL)**	**LSD[1]**
Neuroptera	Lacewing (L)[2]	2.000	2.667	3.667	2.546
	Lacewing (egg)	2.000	3.333	4.667	4.452
Coccinellidae	Lady beetle (A)[2]	8.000	7.667	11.333	6.269
	Lady beele (L)	3.000	4.000	4.667	5.641
	Scymnus spp.	0.000	0.000	0.333	--[3]
Predatory Hemiptera	Big-eyed bug (A)	0.000	1.000	0.000	--
	Minute pirate bug	1.000	0.333	0.000	--
	Assassin bug	0.000	0.333	0.000	--
	Sum of predatory Hemiptera	1.000	1.667	0.000	1.687
	Spiders	11.667	12.667	12.333	5.031
Herbivorous Hemiptera & Homoptera	Aphids	47.667	48.000	51.667	8.808
	Brown stink bug (A)	1.667	0.333	1.000	1.647
	Tarnished plant bug (A)	2.000	2.333	1.333	2.882
	Tarnished plant bug (N)	3.000	1.667	2.333	3.016
	Cotton fleahopper (A)	0.000	0.667	1.333	--
	Cotton fleahopper (N)	0.000	0.333	0.000	--
	Whiteflies	4.333	5.333	5.000	3.964
Thysanoptera	Thrips	1.333	1.333	1.000	1.677
Other Coleoptera	Carabids	0.000	0.667	0.333	--
	Click beetles	0.000	0.667	1.667	--
Lepidoptera	Saltmarsh caterpillar	1.000*	7.000	0.000*	3.710
	Loopers	0.667	0.333	0.000	--

Data from p. 50, MRID 47462301

[1] Least significant difference, calculated only for those morphotypes averaging at least one individual/plot

[2] A = adults, L = larvae, N = nymphs

[3] -- Statistical comparison not valid due to low numbers (<1/plot)

*Significantly different from Control (S), Control (NL) (Fisher's protected LSD, p≤0.05)

Table 6. Cumulative mean of target and nontarget arthropods/plot for shake sheets Winnsboro (2003)					
Group	**Common name**	**PSC355 (S)**	**PSC355 (NL)**	**WideStrike (NL)**	**LSD[1]**
Neuroptera	Green lacewing	0.667	2.000	0.667	2.013
	Lacewing (L)[2]	6.333	6.667	9.333	5.438
	Brown lacewing	2.000*	0.000	2.000*	1.810
Coccinellidae	Lady beetle (A)[2]	29.667	32.333	32.667	19.352
	Lady beele (L)	26.333	40.000	33.000	35.394
	Lady beetle (eggs)	1.333	0.000	0.000	--[3]
	Scymnus spp.	7.333	7.333	10.000	7.984
Predatory Hemiptera	Big-eyed bug (A)	11.667	11.333	7.333	5.857
	Big-eyed bug (N)	0.000	0.333	0.333	--
	Damsel bug	6.667	5.333	4.333	4.254
	Spined soldier bug (A)	1.333	1.333	0.000	--
	Spined soldier bug (N)	0.667	0.333	0.333	--
	Assassin bug	3.333	3.000	3.333	3.079
	Sum of predatory Hemiptera	23.667	21.667	15.667	10.190
	Spiders	93.000	103.333	109.000	45.038
Herbivorous Hemiptera & Homoptera	Aphids	1.000	1.667	1.667	0.777
	Brown stink bug (A)	9.667	6.000	5.333	5.886
	Brown stink bug (N)	0.667	0.000	1.333	--
	Green stink bug (A)	1.000	0.667	2.667	3.529
	Green stink bug (N)	1.667	0.333	0.667	--
	Tarnished plant bug (A)	6.333	5.667	6.667	5.030
	Tarnished plant bug (N)	11.333	7.000	8.000	8.719
	Cotton fleahopper (A)	1.000	1.333	2.333	2.500
	Other Hemiptera	3.333	2.333	2.000	3.302
	Other Homoptera	0.667	0.333	0.000	--
Hymenoptera	Ants	0.667	1.000	0.667	--
Diptera	Diptera	0.667	0.667	0.667	--
Orthoptera	Grasshoppers	0.333	1.000	1.000	--
	Crickets	0.667	1.000	0.000	--
Other Coleoptera	Carabids	3.333	3.000	1.667	4.042
	Click beetles	24.667*	11.333	14.333	13.047
	Sap beetles	1.667	0.000	4.000	6.162
	Other	6.667	3.000	4.333	4.684
Lepidoptera	Saltmarsh caterpillar	10.333*	6.333	0.667*	3.614
	Bollworm/tobacco budworm larvae	47.333	62.333	2.000*	22.127
	Looper larvae	6.667	7.333	0.000*	4.004
	Yellowstriped armyworm	1.000	1.000	0.000	--
	Other larvae	0.333	0.667	0.333	--
	Sum of all Lepidoptera	67.000	77.667	3.000*	24.396

Data from pp. 51-52, MRID 47462301

[1] Least significant difference, calculated only for those morphotypes averaging at least one individual/plot

[2] A = adults, L = larvae, N = nymphs

[3] -- Statistical comparison not valid due to low numbers (<1/plot)

*Significantly different from Control (S), Control (NL) (Fisher's protected LSD, p≤0.05)

Table 7. Cumulative mean of target and nontarget arthropods/plot for sweep nets Winnsboro (2003)					
Group	Common name	PSC355 (S)	PSC355 (NL)	WideStrike (NL)	LSD[1]
Neuroptera	Green lacewing	1.000	0.667	0.667	--[2]
	Lacewing (L)[3]	4.333	4.333	6.333	4.090
	Brown lacewing	1.333	1.000	1.333	2.023
Coccinellidae	Lady beetle (A)[3]	22.333	21.000	21.667	12.348
	Lady beele (L)	21.333	18.667	33.333	28.519
	Scymnus spp.	3.000	6.667	9.000	5.613
Predatory Hemiptera	Big-eyed bug (A)	7.333	5.000	9.000	6.724
	Big-eyed bug (N)	0.000	1.667	0.667	--
	Damsel bug	4.333	7.667	5.000	3.917
	Spined soldier bug (A)	0.333	0.667	0.000	--
	Assassin bug	2.000	2.000	1.333	2.376
	Sum of predatory Hemiptera	14.000	17.000	16.000	7.245
	Spiders	121.333	129.000	145.000	32.688
Herbivorous Hemiptera & Homoptera	Aphids	1.667	1.667	1.000	1.078
	Brown stink bug (A)	4.000*	0.000	1.000	2.045
	Southern green stink bug (A)	0.333	0.333	0.000	--
	Southern green stink bug (N)	0.000	0.000	0.333	--
	Tarnished plant bug (A)	13.333	8.000	14.333	7.862
	Tarnished plant bug (N)	7.333	5.667	8.000	11.946
	Cotton fleahopper (A)	6.667	10.000	11.333	9.755
	Leafhoppers	4.333	5.000	3.667	4.628
	Alfalfa hopper	2.000	2.333	2.667	3.867
	Other Hemiptera	2.333	1.000	4.333	6.442
	Other Homoptera	6.667	2.667	3.333	4.428
Hymenoptera	Parasitic hymenoptera	0.000	0.333	0.000	--
	Other	1.000	0.667	0.333	--
Diptera	Mosquito	3.333	2.333	4.000	4.110
	Other	13.000	16.333	13.333	11.073
Orthoptera	Grasshoppers	2.000*	0.000	1.000	1.822
Other Coleoptera	Carabids	3.333	7.333	9.333	9.094
	Scarabs	0.000	0.667	0.333	--
	Click beetles	7.667	8.667	6.333	7.363
	Weevils	0.333	1.000	0.667	--
	Other	2.667	4.667	2.667	4.300
Collembola	Springtails	0.667	0.000	0.000	--
Lepidoptera	Bollworm/tobacco budworm larvae	17.667	20.667	2.333*	8.825
	Saltmarsh caterpillar	3.000	3.333	0.667*	2.585
	Looper larvae	2.000	0.667	0.000	--
	Yellowstriped armyworm	0.333	1.333	0.000	--
	Other armyworms	0.667	0.000	0.000	--
	Sum of all Lepidoptera	27.333	27.333	3.333*	11.592

Data from pp. 53-54, MRID 47462301

[1] Least significant difference, calculated only for those morphotypes averaging at least one individual/plot

[2] -- Statistical comparison not valid due to low numbers (<1/plot)

[3] A = adults, L = larvae, N = nymphs

*Significantly different from Control (S), Control (NL) (Fisher's protected LSD, p≤0.05)

Selected results for the more abundant morphotypes identified at Maricopa in 2003 are summarized in Tables 9 –11. For the sweep net samples, the numbers of two coleopterans (*Conotelus* and Nitidulidae) were significantly lower in the WideStrike (NL) group than in the PSC355 (S) group. However, there was no significant difference between the WideStrike (NL) and PSC355 (NL) groups. The study author speculates that this was likely the result of elimination of key predators by the WideStrike (NL) and PSC355 (S) treatments. For the pitfall traps, the numbers of two coleopterans (*Conotelus* and Mycetophagidae) were significantly lower in the WideStrike (NL) group than in the PSC355 (S) group. Again, however, there was no significant difference between the WideStrike (NL) and PSC355 (NL) groups. For the sticky traps, the numbers of one coleopteran (*Chaetocnema repens*) and two dipterans (Achradocera and Sciaridae) were significantly lower in the WideStrike (NL) group than in the PSC355 (S) group, but not lower than in the PSC355 (NL) group. The study authors stated that principal response curve analysis indicated that there were no major effects on the arthropod community in the WideStrike (NL) plots compared to the other treatments; however, the resulting PRC plots were difficult to read and were not explained.

Table 8. Cumulative mean of target and nontarget arthropods/plot for sticky traps Winnsboro (2003)

Group	Common name	PSC355 (S)	PSC355 (NL)	WideStrike (NL)	LSD[1]
Neuroptera	Green lacewing	0.333	0.333	0.333	--[2]
	Lacewing (L)[3]	0.000	0.333	0.000	--
	Brown lacewing	0.333	0.000	0.667	--
Coccinellidae	Lady beetle (A)[3]	65.000	62.667	59.667	27.519
	Lady beele (L)	0.333	0.000	0.000	--
	Scymnus spp.	10.333	11.333	15.000	6.017
Predatory Hemiptera	Big-eyed bug (A)	1.667	2.000	2.000	3.125
	Minute pirate bug	15.000	14.000	13.333	7.503
	Damsel bug	0.333	0.333	0.000	--
	Assassin bug	1.667	0.000	0.000	--
	Sum of predatory Hemiptera	18.667	16.333	15.333	8.326
	Spiders	11.667	14.000	13.667	10.514
Herbivorous Hemiptera & Homoptera	Aphids	6.000	5.667	6.000	0.556
	Brown stink bug (A)	1.000	0.333	0.667	--
	Southern green stink bug (A)	0.333	0.000	0.000	--
	Tarnished plant bug (A)	9.000	9.000	7.000	6.012
	Tarnished plant bug (N)	1.000	0.000	0.333	--
	Cotton fleahopper (A)	21.667	22.000	28.667	9.505
	Leafhoppers	289.333*	237.667	214.333	51.103
	Other Hemiptera	4.333	4.333	4.333	3.803
	Other Homoptera	7.333	5.000	10.000	8.672
Thysanoptera	Thrips	6.000	5.667	6.000	0.556
Hymenoptera	Parasitic hymenoptera	1.333	0.667	0.667	--
	Ants	4.000	3.667	3.333	2.251
	Other	4.000	3.333	3.333	3.289
Diptera	Diptera	377.000	348.667	320.333	93.668
Other Coleoptera	Carabids	114.000	139.667	154.333	62.262
	Click beetles	4.000	2.667	4.000	2.607
	Sap beetles	2.333	3.000	3.000	3.298
	Weevils	7.000	4.000	5.333	5.542
	Other	42.333	33.000	36.333	18.580
Lepidoptera	Saltmarsh caterpillar	0.333	0.000	0.000	--
	Other larvae	3.333	13.000	3.667	15.932
	Sum of all Lepidoptera	3.667	13.000	3.667	15.892

Data from pp. 55-56, MRID 47462301

[1] Least significant difference, calculated only for those morphotypes averaging at least one individual/plot

[2] -- Statistical comparison not valid due to low numbers (<1/plot)

[3] A = adults, L = larvae, N = nymphs

*Significantly different from Control (S), Control (NL) (Fisher's protected LSD, p≤0.05)

Table 9. Mean cumulative seasonal counts for sweep net samples Maricopa (2003)			
Order/identification	**PSC355 (NL)**	**PSC355 (S)**	**WideStrike (NL)**
Coleoptera/Anthicus	0.00 ± 0.00	0.33 ± 0.33	1.00 ± 0.00
Coleoptera/*Chaetocnema repens*	34.67 ± 8.45 a	15.00 ± 1.00 b	29.33 ± 2.96 a
Coleoptera/*Corticoria elongato*	1.33 ± 0.33	0.33 ± 0.33	2.33 ± 0.33
Coleoptera /*Conotelus mexiconus*	65.67 ± 14.52 a	108.33 ± 8.69 b	62.00 ± 8.08 a
Coleoptera/Nitidulidae larvae	5.33 ± 1.45 a	106.00 ± 28.75 b	22.33 ± 11.62 a
Coleoptera/*Oxyporus*	0.33 ± 0.33	2.67 ± 0.88	1.00 ± 0.58
Hemiptera/Lygus	204.67 ± 3.84 a	157.67 ± 9.53 **b**	185.00 ± 2.52 ab
Hemiptera/*Spanagonicus*	49.33 ± 5.04 a	71.67 ± 21.23 ab	108.00 ± 7.64 b
Hemiptera/Rhopalidae	4.33 ± 0.67	8.00 ± 0.00	5.33 ± 1.67
Lepidoptera/*Pectinophoro gossypiella*	0.67 ± 0.33	0.00 ± 0.00	0.00 ± 0.00
Neuroptera/Chrysopa	64.33 ± 10.11 a	39.67 ±6.06 **b**	38.67 ± 2.03 b
Neuroptera/Chrysopa	29.33 ± 7.31 a	16.67 ± 1.76 b	37.67 ± 2.60 a
Lepidoptera/*Spodoptera exigua*	11.67 ± 3.18 a	13.33 ± 2.03 a	0.00 ± 0.00 **b**
Lepidoptera/Pyralidae	2.33 ± 1.20	5.33 ± 0.88	1.33 ± 0.33
Hymenoptera/Eulophidae	2.33 ± 0.33	0.67 ± 0.33	0.67 ± 0.33
Diptera/unknown	0.00 ± 0.00	1.00 ± 0.58	2.33 ± 0.33
Homoptera/Cicadellidae	8.00 ± 0.58	4.33 ± 0.67	5.00 ± 1.53

Data from pp. 68-70, 72 MRID 47462301
Means within the same row sharing the same letter not significantly different (p>0.10)

Table 10. Mean cumulative seasonal counts for pitfall samples Maricopa (2003)			
Order/identificatin	**PSC355 (NL)**	**PSC355 (S)**	**WideStrike (NL)**
Coleoptera /*Conotelus mexiconus*	1.00 ± 0.00 a	7.00 ± 0.58 c	3.67 ± 1.20 b
Diptera/Phoridae	32.67 ± 3.76 a	25.33 ± 2.91 ab	19.00 ± 2.08 b
Coleoptera/*Systena blanda*	4.00 ± 0.58 a	2.00 ± 0.58 a	7.00 ± 0.58 b
Coleoptera/Coccinellidae or Corylophidae	3.33 ± 1.20 a	1.00 ± 1.00 b	2.00 ± 1.00 ab
Coleoptera/Mycetophagidae	18.00 ± 4.04 ab	23.33 ± 1.86 a	11.33 ± 3.48 b
Hemiptera/*Spanogonicus*	2.33 ± 0.67 a	2.00 ± 1.00 a	6.67 ± 1.20 b
Homoptera/*Bemisio tobaci*	22.67 ± 1.45 **ab**	16.00 ± 1.00 a	38.00 ± 9.07 b
Hymenoptera/Ceraphronidae	1.67 ± 0.67 ab	0.67 ± 0.33 **a**	4.00 ± 1.00 b
Hymenoptera/Tiphiidae	0.00 ± 0.00 a	1.33 ± 0.33 b	1.00 ± 0.00 b
Lepidoptera/Noctuidae	1.33 ± 0.33 a	0.67 ± 0.33 ab	0.33 ± 0.33 **b**
Neuroptera/*Chrysopa*	1.00 ± 0.58 **ab**	0.67 ± 0.67 a	1.67 ± 0.67 b
Lepidoptera/*Spodoptera exigua*	2.33 ± 1.86 ab	4.00 ± 1.00 a	0.00 ± 0.00 b
Araneae/	0.00 ± 0.00 a	1.67 ± 0.67 b	1.67 ± 0.33 b

Data from pp. 68-70, 73 MRID 47462301
Means within the same row sharing the same letter not significantly different (p>0.10)

Table 11. Mean cumulative seasonal counts for sticky trap samples Maricopa (2003)				
Trap color	Order/Identification	PSC355 (NL)	PSC355 (S)	WideStrike (NL)
Blue	Araneae/immature	1.67 ± 0.33	0.33 ± 0.33	0.33 ± 0.33
Yellow	Araneae/immatur	2.33 ± 0.33 a	0.00 ± 0.00 b	0.67 ± 0.67 ab
Blue	Coleoptera/*Chaetocnema repens*	0.33 ± 0.33	1.33 ± 0.33	0.67 ± 0.67
Yellow	Coleoptera/*Chaetocnema repens*	0.00 ± 0.00 a	1.67 ± 0.33 b	0.00 ± 0.00a
Blue	Diptera/Camilidae	3.33 ± 0.33 a	4.67 ± 1.45 a	8.33 ± 1.67 b
Yellow	Diptera/Camilidae	0.67 ± 0.67	0.33 ± 0.33	1.67 ± 0.88
Blue	Diptera/Psectrosciara	15.67 ± 2.91 a	11.00 ± 2.08 ab	8.33 ± 3.84 b
Blue	Hymenoptera/*Telenomus telenominae*	0.33 ± 0.33 ab	0.00 ± 0.00 a	1.00 ± 0.00 b
Yellow	Hymenoptera/*Telenomus telenominae*	0.67 ± 0.67	0.33 ± 0.33	0.67 ± 0.33
Blue	Thysanoptera/Thripidae	2897 ± 431	3172 ± 196	2867 ± 507
Yellow	Diptera/Ephydridae	6.67 ± 0.88	11.33 ± 2.33	8.33 ± 2.03
Blue	Diptera/Otitidae	4.33 ± 1.67	5.33 ± 2.33	6.00 ± 3.51
Blue	Diptera/*Asyndetus*	29.33 ± 4.26 a	20.00 ± 3.06 ab	13.67 ± 1.67 b
Yellow	Diptera/*Asyndetus*	30.00 ± 3.79 a	17.67 ± 4.70 b	13.67 ± 0.67 b
Blue	Diptera/*Achradocera*	1.00 ± 0.58	0.33 ± 0.33	0.00 ± 0.00
Yellow	Diptera/*Achradocera*	0.67 ± 0.67 a	1.67 ± 0.67 b	0.33 ± 0.33 a
Blue	Diptera/Ceratopogonidae	1.67 ± 1.20	2.33 ± 0.33	1.33 ± 0.67
Yellow	Diptera/Ceratopogonidae	2.33 ± 0.67 a	0.67 ± 0.33 a	0.33 ± 0.33 a
Blue	Lepidoptera/micro-Lepidoptera	1.00 ± 0.00 a	0.00 ± 0.00 b	0.33 ± 0.33 ab
Yellow	Lepidoptera/micro-Lepidoptera	0.67 ± 0.33 ab	0.33 ± 0.33 a	1.33 ± 0.33 b
Blue	Diptera/Sciaridae	0.00 ± 0.00	0.33 ± 0.33	1.33 ± 0.67
Yellow	Diptera/Sciaridae	0.33 ± 0.33 a	2.00 ± 0.00 b	0.67 ± 0.33 a
Blue	Diptera/Sciaridae	0.67 ± 0.67	0.00 ± 0.00	0.67 ± 0.67
Yellow	Diptera/Sciaridae	1.33 ± 0.33 a	0.00 ± 0.00 b	1.00 ± 0.58 ab
Blue	Diptera/unknown midge	1.67 ± 1.20	0.00 ± 0.00	0.33 ± 0.33

Data from pp. 68-70, 74 MRID 47462301

Means within the same row sharing the same letter not significantly different ($p>0.10$)

Study Author's Conclusion

The study author concluded that abundance of nontarget arthropods (including *Orius* and other beneficial species) in WideStrike cotton was similar to that in non-*Bt* cotton that was similarly managed for pests but received no insecticidal sprays to control Lepidoptera. Therfore, WideStrike will not have an adverse impact on nontarget arthropods compared to non-*Bt* cotton.

EPA Reviewer's Conclusion

This study does not have a robust experimental design and should be considered preliminary for the purposes of drawing conclusions about community level or long-term environmental effects. However, this study does provide some information on the impact of Widestrike cotton to nontarget insects in the field. It also provides some information about *Orius* species and other hemiptera, and is sufficient to fulfill the requirement of a Tier I nontarget insect study with *Orius* as condition of registration.

The information in MRID 474623-01 contains the results of the second year of this two-year study. Data collected from these sites in the first year of this study (2002) were submitted in MRID 458084-19, reviewed by the Agency (and classified as supplemental), and also presented to a FIFRA Science Advisory Panel in June 2004. The SAP found several problems in the experimental design, particularly that the plot sizes were too small to ensure independence of samples and that the large number of taxa studied complicates the analyses and reduces the resolution of true differences such that no definitive conclusions can be made of most taxa.

Similar problems were also noted in the 2005 BRAD for Widestrike cotton, but the data were considered useful as preliminary findings. The SAP did not recommend any further field testing.

This problems outlined by the SAP were addressed in the 2003 season by increasing the size of the experimental plots, which does alleviate some of the uncertainty caused by non-independence. Additional collection methods were also used. However, most of the SAP's comments also apply to the 2003 data set, and these problems are significant if the study is ever to be considered a rigorous field study of effects to nontarget insects.

Since the independence between the plots was questionable in the first year, the study essentially presents only one year of data. Furthermore, samples collected at each of the two sites were processed and analyzed in different ways, essentially dividing efforts at the two sites into two different studies. The study also gathered information on a broad spectrum of taxa in both years, and the data analyses included insects identified down to species in some cases and only genus in others. This reduces the number of species-level comparisons that can be made and lessens the detection of changes in community composition. The results also show very low numbers for several species, and some species were not detected in the Widestrke plots, whereas they were detected in the control plots.

Despite its issues with experimental design, the study does provide evidence that adverse effects resulting from use of Widestrike cotton are unlikely to several nontarget species for which sufficient data were collected. The study is classified as supplemental and is sufficient to fulfill the nontarget insect data required as a condition of registration.