8-20-96

DATA EVALUATION RECORD
PLANT RESIDUE TEST - SINGLE APPLICATION

1. **CHEMICAL:** PIRATE™ **PC Code No.:** 129093
4-bromo-2-(4-chlorophenyl)-1-(ethoxymethyl)-5-(trifluoromethyl)-1H-pyrrole-3-carbonitrile

2. **TEST MATERIAL:** AC 303,630 3SC **Purity:** 30.4%

3. **CITATION**

Author: J.P. Sullivan, L. Henry and A.G. Grant
Title: Evaluation of Potential Ecotoxicological Effects of AC 303,360 on Birds: Residues of AC 303,360 and Their Dissipation After Single Applications in Cotton Fields.
Study Completion Date: 1 December 1994
Laboratory: ABC Laboratories, Inc., Columbia, MO
Sponsor: American Cyanamid Company, Princeton NJ
Laboratory Report ID: 954-94-150
MRID No.: 434928-14
DP Barcode: ~~D211863~~ D210808

4. **REVIEWED BY:** John Eisemann, Wildlife Biologist, EEB, EFED

Signature: John D. Eisemann **Date:** 7/30/96

5. **APPROVED BY:** Ann Stavola, Head of Section (5), EEB, EFED

Signature: Ann Stavola **Date:** 8/20/96

6. **OBJECTIVES:**

To determine the residues of the test substance on or in components of a cotton field and its environs that could cause exposure to birds immediately after single application and up to 28 days post-application.

7. **CONCLUSIONS:**

With the exception of the residue data reported for the insects in cotton and adjacent habitats the residue data reported in this report were collected using acceptable methods and as a result they are considered acceptable to EEB.

Insect collection methods bias the data towards live insects. Insect residue information in this report should not be used to estimate average concentrations in the entire population.

EFGWB provided verification of target application rates, sample method validation, half-life determination in/on soil and cotton leaves. Additionally, EFGWB examined dislodgeable residue calculations. Contradictory to the registrant's claim of 0.1% dislodgeable residues form cotton leaf surfaces, it was determined that "*most, if not essentially all, PIRATE remaining*

of foliage at all sampling times is easily removed with the diluent surfactant solution'. **A copy of the EFGWB review is attached to this review.**

Residues reported on cotton leaf tissue five hours after application at rates of 0.2 lb a.i./acre and 0.4 lb a.i./acre, were 127% and 183% of predicted levels by methods proposed by Fletcher (1994). Residue levels dropped below the predicted post-spray concentrations between one and three days application.

Residues found in insects at an application rate of 0.4 lbs a.i./acre exceed the LOEC for Mallard and Northern bobwhite reproduction by a factor of 3.8 through day 1 post-sampling and declined below the LOEC by day seven.

8. **ADEQUACY OF THE STUDY:**

A. Classification: Supplemental

B. Rationale: This is not a required study.

C. Repairability: No additional information needs to be submitted.

9. **METHODS:**

Test Substance Characterization: AC 303,630 3SC is a 30.4% solution of AC 303,360 (A.I.) formulated as a suspension concentrate.

Site Selection and Description: The study was conducted in a gradually sloping (<1.0%) cotton field in Cal, GA. The site consisted of two adjacent untreated plots and two treated plots which were approximately 45 meters apart. The untreated and treated plots were separated by 305 m. Residues were measured in one treated and one untreated plot after an early season application simulating the control of *Thrips*. Residues in the other two plots were measured after late season spraying simulating the control of *Heliothis*. Untreated plots measured 11.6 m x 36.6 m and contained 12 rows of cotton. Treated plots were divide into three subplots measuring 34.7 m x 33.5 m and 34.7 x 20.4 m for *Thrips* and *Heliothis*, respectively. Both sets of untreated plots contained 36 rows of cotton.

The test site soil is characterized as a loamy sand. Untreated soils were determined to contain 83% sand, 12% silt, 5% clay have a pH of 5.9 and organic matter content of 1.5%. Treated soils were determined to contain 86% sand, 9% silt, 5% clay have a pH of 6.4 and organic matter content of 1.1%.

Site History: Untreated plots were fallow 1991, 1992, and 1993. Peanuts were grown on the treated plots in 1991 and 1992. They were left fallow in 1993. PROWL (1991 and 1992), BRAVO (1992), and POAST (1992) were applied to the treated fields. No pesticides were applied in 1993.

Test Substance Application and equipment: A Cross PTO boom sprayer equipped with 13 TeeJet 4X cone nozzles, spaced at 44.5 cm intervals, was used to apply the pesticide. The application procedure was calibrated to deliver 7.1 gal/A.

The test substance was applied (June 22, 1993) at a rate of 0.2 lb/A. A late season application (July 22, 1993) was made at a rate of 0.4 lbs/A. Samples of the test solution were collected in duplicate immediately prior to and after the application.

Climatic Conditions: Weather data were collected from a National Weather Service site 3.5 miles northeast of the test site. Climatic conditions at the test site on the application days were as follows: June 22, 1993 - air temperature 92° F, relative humidity 45%, sky partly cloudy, wind 0-2 mph from the northeast; July 22, 1993 - air temperature 99° F, relative humidity 54%, sky partly cloudy, wind calm. Daily climatic conditions were recorded.

Agronomic practices: The test site was disked to a depth of six to eight inches on May 19, 1993. The rows were cultivated weekly to a depth of three to four inches, for three weeks, prior to pesticide application. Stoneville 435' cotton was planted on May 20, 1993 at a rate of 10 lbs/A with 38 inches between rows and 4 inches between plants. Maintenance pesticides were applied as follows:

Pesticide	Rate Applied	Date(s)
ZORIAL	0.5 lb a.i./A	May 20 and 21, 1993
PROWL	1.0 lb a.i./A	May 20, 1993
COTORAN	1.5 lb a.i./A	May 21, 1993
POAST PLUS	0.3 lb a.i./A	July 12,1993

The test substance was applied to simulate *Thrips* control when the cotton had four to six true leaves (June 22, 1993) and on the second set of treated plots to simulate *Heliothis* control when the cotton was in the pin-head square stage (July 22, 1993).

Overhead sprinkler irrigation was used 7 times between May 27, 1993 and July 31, 1993 and averaged 1.34 inches per watering with a range of 0.46 - 2.6 inches. No irrigation was used within 9 days of an application.

Sample Collections: Samples were collected on days -1, 0, 1, 3, 7, 14, and 28 in relation to the test substance application dates. No samples were collected in a 3 m buffer strip along the outer area of the treated subplots. Additionally, a separate area was established for the collection of insects within the plots.

All samples were placed in a freezer (16°F to -22°F) within three hours after collection, except for the leaf disk samples. Samples were kept frozen until shipment to American Cyanamid, where they were stored frozen (-20°C) until analysis.

Earthworms: Two methods were employed to collect earthworms: sorting through soil from a 30 cm x 30 cm x 3 cm area was dug from the field and night collections. No earthworms could be found in any plot. Earthworm collections were abandoned.

Cotton Leaves: Individual leaves were collected by snipping the leaf from the petiole. Only one leaf was collected per plant each sampling period during the early season application. Two leaves, one in the upper and lower canopy, were collected per plant during the late season collections. Collections totaled 150 g per period.

Cotton Leaf Disks for Dislodgeable Residues: Thirty-five leaf disks, 2.54 cm in diameter, were collected from each plot. Each disk was deposited directly into a glass jar. Immediately after leaving the field the leaf was washed three times with 100 ml, 2% dioctylsulfosuccinate salt in aqueous solution. The resulting 300 ml of wash solution was combined and frozen.

Avian Food Items: Avian food item samples were collected within 15 m of both treated and untreated cotton in adjacent fallow areas. Collections were made by snipping flower heads, seed heads and fruits off the plant and depositing them directly into a plastic bag.

Insects: Insects were collected using two methods: sweep net and pit fall traps and stored in plastic bags. Sweep net samples were collected by sweeping above the vegetation in treated, untreated and adjacent habitats for 10 minutes. The pit fall traps consisted of 8.9 c x 11.4 cm plastic cups set in the ground so the lip of the cup and the soils surface were even. Ten pit fall traps were used in each plot and in adjacent habitat. Due to small sample sizes the insects collected from both methods were pooled for analysis.

Soil: Soil was scooped into plastic bag from and area 10.2 cm in depth and 5.5 cm in diameter. Six samples were collected from each plot on days -1, 0, 1, 7, 14, and 28.

Air Concentrations: Air concentrations were measured in each subplot of the treated plot during early applications at a distance of 30.5 cm above the ground. During the late season application measurements were taken at the same distance to the ground but below the canopy of the cotton. Measurements were taken, for one hour, during two time periods: during application and between 3 and 4 hours post application. Samples were collected using a Nalgene open-faced filter holder with 47 mm diameter Gelman Type A/E glass filter paper.

Spray Droplet Size: Water sensitive cards were used to measure spray droplet size at a distance of 30.5 cm above the ground.

GLP and Quality Assurance: Guidelines as specified by the U.S. EPA's Good Laboratory Practice Standards 40 CFR Part 160 were followed in addition to established SOP's of the test laboratory.

10. DEVIATIONS:

1. No earthworm samples were collected after the initial sample collection efforts made on the day before the early season application. No earthworms could be found during the sampling.

2. The Eberbach Reciprocating Shaker was calibrated on June 21, 1993 and August 4, 1993 at 240 - 243 strokes per minute. The protocol specifies the reciprocating shaker operate at 250 strokes per minute.

3. The lot number of the analytical standard used to analyze the tank mixes from the second application was AC 6937-118-1.

4. The surface washed for dislodgeable residues was 354.70 cm^2. A different diameter leaf punch was used to make the collections and the investigator made a calculation mistake.

11. STATISTICAL ANALYSIS:

Other than calculation of sample means, no statistical analysis was conducted. Samples which were found to contain residues below the method detection limit were given a value of one half the method detection limit.

12. **RESULTS:**

AC 303,630 residues in untreated plots were below the reported detection limits for all sample types collected during all collection periods. Concurrent analysis of fortified cotton leaf samples, cotton leaf wash samples, avian feed samples, soil samples, insect samples and air samples showed recoveries of 100.6%, 90.6%,103.3%, 102.6% 109.3% and 84.3%, respectively.

Tank mix analysis indicated the formulation contained 28.26 - 29.10% a.i. and was homogenous and stable. Based upon the rate applied and the number of gallons used, an average of 97.8 ± 2.82% of the theoretical application concentration was applied to the cotton crop.

Early season spray droplets averaged 266 ± 58 um. Late season spray droplets averaged 292 ± 36 um and 192 ± 36 um in the upper and lower canopies, respectively.

Early Season Application Residues from Treated Plots
Application Rate = 0.2 lb ai/Acre

Collection Interval (Days)	Cotton Leaf (ppm)	Leaf Wash (ng/cm²)	Avian Feed (ppm)	Soil (ppb)	Composite Insect-Cotton Habitat (ppm)	Composite Insect - Adjacent Habitat (ppm)
-1	<0.5	<15	<MDL	<10.0	NC	NC
0.2	34.4	398	<MDL	95.0	0.963	<0.5
1	26.6	359	<MDL	NC	0.963	<0.5
3	8.93	95.1	<MDL	NC	0.724	<0.5
7	1.14	<15	<MDL	44.5	0.724	<0.5
14	<0.5	<15	<MDL	86.7	<0.5	<0.5
28	<0.5	<15	<MDL	76.5	<0.5	<0.5

Note: NC - As per the protocol, no collection conducted.
: n=3 in all groups

Late Season Application Residues from Treated Plots
Application Rate = 0.4 lb ai/Acre

Collection Interval (Days)	Cotton Leaf (ppm)		Leaf Wash (ng/cm²)		Avian Feed (ppm)	Soil (ppb)	Composite Insect-Cotton Habitat (ppm)	Composite Insect - Adjacent Habitat (ppm)
	Upper Canopy	Lower Canopy	Below Canopy	Upper Canopy				
-1	<0.5	<0.5	<15	<15	<MDL	<10.0	NC	<0.5
0.2	98.9	39.8	779	451	<MDL	158	5.710	<0.5
1	61.5	20.6	866	303	<MDL	NC	5.710	<0.5
3	43.5	33.9	825	317	<MDL	NC	1.290	<0.5
7	27.2	26.0	186	97.1	<MDL	94.0	1.290	<0.5
14	6.9	6.63	<15	27.5	<MDL	170.0	<0.5	<0.5
28	3.3	4.3	<15	<15	<MDL	104.0	<0.5	<0.5

NOTE: NC- As per the protocol, no collection conducted.
: Four avian feed samples had detectable AI residues.
: N=3 in all groups except n=2 for the Leaf Wash in the Upper Canopy at 0.2 Days
: Residues were detected in the Avian Feed samples 0.576 ppm, 0.712 ppm, 0.524 ppm, and 0.768 ppm at Day 0.1, 3, 14 and 14, respectively.

Air concentrations during and post-application

Collection Period	Early Season Application (ug/cm²)	Late Season Application (ug/cm²)	
		Below Canopy	Upper Canopy
0 - 1 Hour	2.35	1.87	4.10
3 - 4 Hour	<1.00	2.51	<1.00

NOTE: n=3 in all groups
: Early season application rate = 0.2 lb a.i./acre
: Late season application rate= 0.4 lb a.i./ acre

REVIEWER'S RESULTS AND COMMENTS:

The results of EEB statistical analysis were the same as those reported. Only descriptive statistics were calculated. No statistical tests were preformed.

The methods used to collect insects in this study will bias the data towards those individuals able to fly or walk after exposure and underestimate the average residues found in

exposed insects. An assumption of pit fall and sweep net trapping methods is that the individuals are mobile and their mobility leads to their capture. These sampling methods exclude immobilized or dead insects and therefore do not provide an accurate estimate of the average residues in the insect population. One assumes higher residues will be found in immobilized or dead individuals than in unaffected individuals. Using only live insects underestimates the active ingredient residues in the prey population and the risk to foraging wildlife.

It should be noted that residues found in insects at an application rate of 0.4 lbs a.i./acre exceed the LOEC for mallard and northern bobwhite reproduction by a factor of 3.8 through day 1 post-sampling. The residues at day seven declined to a level slightly below the LOEC for mallard and northern bobwhite reproduction.

Residues reported on cotton leaf tissue after the early season application, 0.2 lb a.i./acre, averaged 34.4 ppm (30.2 to 40.3 ppm) five hours after spraying. The residues detected were 138% and 127% of the predicted concentration of 25 ppm and 27 ppm as determined by methods developed by Hoerger and Kenaga (1972) and Fletcher et al. (1994), respectively. Residue levels dropped below the predicted post-spray concentrations between one and three days after application.

Residues reported on cotton leaf tissue after the late season application, 0.4 lb a.i./acre, from the upper canopy, averaged 98.9 ppm (98.3 to 100.0 ppm) five hours after spraying. The residues detected were 197% and 183% of the predicted concentration of 50 ppm and 54 ppm as determined by methods developed by Hoerger and Kenaga (1972) and Fletcher et al. (1994), respectively. Residue levels dropped below the predicted post-spray concentrations between one and three days after application.

Residues reported on cotton leaf tissue after the late season application, 0.4 lb a.i./acre, from the lower canopy, averaged 39.8 ppm (37.2 to 41.6 ppm) five hours after spraying. The residues detected were below the predicted concentration as determined by methods developed by Hoerger and Kenaga (1972) and Fletcher et al. (1994), respectively.

DP Barcode: ~~D211863~~ D210705

MRID No.: 434928-14

REFERENCES:

Hoerger, F. and E.E. Kenaga. 1972. Pesticide residues on plants: Correlation of representative data as a basis for estimation of their magnitude in the environment. *In* F. Coulston and F. Korte, (eds.), Environmental Quality and Safety: Chemistry, Toxicology, and Technology. Georg Thiem Publishers, Stuttgart, West Germany, pp. 9-28.

Fletcher,J.S., J.E. Nellessen and T.G. Pfleeger. 1994. Literature review and evaluation of the EPA food-chain (Kenaga) nomogram, an instrument for estimating pesticide residues of plants. Environ. Toxico. and Chem. 13(9):1383-1391.

PC Code: 129093 (PIRATE) DP Barcode D220955 7/15/96

MEMORANDUM

SUBJECT: Residues of PIRATE in/on Cotton Fields
(AC 303,630 3SC Formulation, approximately 30% a.i.)

TO: Tony Maciorowski, Chief, EEB (7507C)
Ann Stavola, Section Head, EEB (7507C)
John Eisemann, Wildlife Biologist (7507C)

FROM: Alex T. Clem, Environmental Scientist, EFGWB (7507C)

THRU: Akiva D. Abramovitch, Ph.D; Head Sec. 3, EFGWB (7507C)

Henry Jacoby, Chief, EFGWB

EFGWB is formally responding to your 14 Nov 95 memo (copy attached) asking for an evaluation of specific areas of American Cyanamide's study on the potential ecological effects of PIRATE on birds (MRID 434928-14, D211863, Guideline 70-1d). We have been working closely with John Eisemann on various aspects.

Our responses to the "bulleted" items in your memo are as follows:

* **Verification of target application rates based on actual soil and leaf residues**

In our experience, the percentages of target we give below are typical of many field studies. It is noteworthy that, within experimental variation, soil concentrations remained essentially constant in both treatments (*Thrips* and *Heliothis*) throughout the periods of study. Soil samples were described as taken "between the rows" (pp. 55-56). This imprecise phrase does not make clear if areas beneath the cotton canopy were sampled, or if it is intended to imply that samples were taken only from the middle of the 38 inch rows. That fact that there were no discernable effects associated with rainfall or irrigation, in spite of the ease with which the pesticide is dislodged (see below), points to soil samples being collected in row middles, and not under the leaf canopy where they would be dislodged most probably.

Our calculations based on the soil analysis for parent at the target rate of 0.2 lb/acre (*Thrips* treatment in the early cotton growth stage on 22 June 93) shows that *the mean rate was about 60 ± 20% of target*. (Actual mean concentration over all 12 samples taken throughout the 28 day period of study (four sampling dates, each sample a composite of six cores) was 76 ± 28 ppb. The reported "sensitivity" was 10 ppb. The theoretical target concentration was approximately 130 ppb. Soil cores were 5.5 cm in diameter and 10.2 cm in depth in a loamy sand with bulk density of 1.69 g/cm^3.)

Similarly, for the targeted rate of 0.4 lb/acre (*Heliothis* treatment in separate plot in the

later pinhead square stage of cotton on 22 July 93) *the mean rate was about 50 ± 25% of target*. Actual mean concentration over all 12 samples was 132 ± 63 ppb with a target of approximately 260 ppb.

Dislodgeable leaf data and total leaf data yield an estimate of about 30 to 40% of the target rate for each of the various treatments. We did not attempt a statistical analysis for these data, preferring instead the soil results for which there were more samples and fewer complications. Soil and leaf results are thus seen to overlap.

* **Sampling method validation**

Our only comment is that the sampling methodologies seem "standard". (Note: It is our opinion that present sampling standards are inadequate to measure spatial variability. In general, also the case for this study, registrants do not conduct replicated field experiments and too few subsamples of insufficient size (area, volume, mass, etc.) are analyzed. As a result, systematic statistical effects or bias may often be responsible for apparent anomalies such as low day 0 recoveries of applied pesticide.)

* **Half-lives for PIRATE in/on 1) soil, 2) cotton leaves, and 3) for that which is dislodgeable (with dilute surfactant in water) from leaves.**

Meaningful "half-lives" cannot be determined from the study. That the DT50s given below for foliage are subject to interpretation is discussed in the Summary and Conclusions section. With this in mind, results are as follows:

1) *Soil half-lives* were indeterminately long in the 28 day study periods (concentrations did not change). We know from previous lab and field experiments that PIRATE is very persistent in soil with half-lives measured in units of years. Therefore, we would not expect appreciable degradation in 28 days.

2) *Total amounts of PIRATE on cotton leaves in both treatments decreased by over 50% by day 3 (DT50 more than 1 day but less than 3 days)*. Average maximum concentration for the *Thrips* treatment (0.2 lb/acre) was about 30 ppm; by day 14 PIRATE was below the reporting limit of 0.5 ppm. Average maximum concentration for the *Heliothis* treatment (0.4 lb/acre) was about 100 ppm on upper leaves and about 40 ppm on lower; on the last day of sampling (day 28) the average concentration was about 4 ppm for all leaves.

3) To remove "dislodgeable" pesticide a dilute foliar wash solution containing a surfactant salt (sodium dioctylsulfosuccinate) in water was used. *The DT50 for dislodgeable PIRATE in the Thrips treatment was more than 1 day but less than 3 days*. Average maximum concentration was about 400 ng/cm^2. (The units of ng/cm^2 are based on the sum of surface areas on top and underside of a leaf. For

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the 400 ng/cm^2 figure just given, this means that 800 ng would be on a leaf disk punched out with a punch which cuts out an area of one square centimeter. To convert the ng/cm^2 numbers as they appear in the data tables and as they are given here to ppm, a *roughly estimated* multiplicative factor of 0.1 can be used. For example, the above 400 ng/cm^2 surface rate multiplied by 0.1 gives a 40 ppm equivalent.) By the next sampling date (day 7) PIRATE was below the reporting limit of 15 ng/cm^2. *The DT50 for dislodgeable compound in the Heliothis treatment was more than 3 days but less than 7 days for both upper and lower leaf canopy samples.* Average maximum concentration was about 900 ng/cm^2 for upper leaves, and about 300 to 400 ng/cm^2 for lower leaves. By day 14, about 20 to 50 ng/cm^2 were measured on some samples in both canopies; other samples were below the reporting limit. By the last sampling date (day 28) all concentrations were below the reporting limit of 15 ng/cm^2. By comparing all data, it is clear that *most, if not essentially all, PIRATE remaining on foliage at all sampling times is easily removed with the dilute surfactant solution.*

Summary and Conclusions

* *A very large portion (even all, within experimental limits) of pesticide remaining on leaves was dislodgeable from all samples.* By implication, therefore, PIRATE is "bioavailable" on cotton foliage.

* *The route(s) and rates of apparent disappearance of pesticide from the leaves cannot not be reliably assigned.* There were no obvious effects associated with rainfall or irrigation. The fact that pesticide lost from leaves did not show up in the soil samples probably indicates a systematic bias in collection of the soil samples (see Verification above). Absence of any chemical analysis for degradates confounds interpretation. It is possible that some photodegradation may have occurred on leaf surfaces. No doubt that formation of new leaves and leaf blade expansion accounts for much of the apparent drop in concentration, but this process would not normally be expected to have such a dramatic effect in the later *Heliothis* treatment. A systematic bias on the part of the leaf sample collector(s) or crop health (stress) could be at play. *Thus the DT50s given above are still open to interpretation.*

* The average day 0 recoveries of about 60% of the target application rate for the *Thrips* treatment and about 50% for the *Heliothis* treatment is typical and acceptable.

* Sampling methods were "standard" and acceptable at this time.