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UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
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OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

MEMORANDUM:

SUBJECT: PP#5F04456 - New Chemical - Pirate/Alert® on Cotton. Evaluation of Residue Data and Analytical Methods. MRID Nos. 434928-01 thru 05 and 434928-51 thru 61. CBTS No. 15094. DP Barcode: D211889.

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Background

The American Cyanamid Company has petitioned for permanent tolerances for residues of the insecticide/miticide Alert also known as Pirate (303,630) [4-bromo-2-(chlorophenyl)-1-(ethoxymethyl)-5-(trifluoromethyl)-1H-pyrrole-3-carbonitrile] as follows:

Cottonseed	0.5 ppm
Milk	0.01 ppm
Milk Fat	0.15 ppm
Meat	0.01 ppm
Meat By-Products	0.10 ppm

Under PP#3G4224 (see 1/26/94 review of G. Otakie) CBTS recommended in favor of a temporary tolerance for Pirate in/on cottonseed of 0.5 ppm, for a period of two years. Under PP#5G04507 (see 8/10/95 review of G. Kramer) CBTS recommended in favor of temporary tolerances for Pirate in/on oranges, orange oil, lemon oil and lemons of 0.50, 2.0, 2.0, and 0.50 ppm, respectively and in milk fat (reflecting 0.01 ppm in whole milk), fat, meat, and meat by-products (cattle, goats, horses and sheep) of 0.25, 0.20, 0.01, and 0.05 ppm, respectively. Other temporary tolerances for Alert/Pirate have been proposed under PP#5G04523 and PP#5G04548 in/on lettuce and cabbage, respectively. Per PP#3G4224 (see 5/16/95 review of G. Otakie) a satisfactory EPA method trial for Pirate in/on cottonseed has been completed.

Conclusions

1. Most product chemistry data requirements for the TGAI have been satisfied. Preliminary analysis data reflects pilot as well as large scale production. A revised CSF is required with certified limits for all impurities $\geq$0.1 w/w%. Pending submission of a revised CSF, adequate product chemistry data are available for the subject proposed permanent tolerances for Pirate in/on cottonseed and animal commodities. The petitioner should submit verification of an approved ANSI common name if one has been obtained.

2. CBTS notes that the maximum application rate has been reduced in the subject petition from 2.0 lbs ai/A/season in the temporary tolerance PP#3G4224 to 1.05 lbs ai/A/season. A revised label with a crop rotation statement not to plant to any food or feed crop within 60 days of the last application is required (see Conclusion 11).

3. a. The nature of the residue in cotton is adequately understood. Pending HED Metabolism Committee review, the residue of concern in cotton consists of the parent Pirate.

   b. Pirate will be referred to the HED Metabolism Committee once the toxicology data has been reviewed.

4. The nature of the residue in poultry is adequately understood and consists primarily of the parent in muscle and fat. In addition to the parent numerous Pirate metabolites have been identified. In eggs the parent and its N-dealkylation metabolite CL 303,268 [i.e. Pyrrole-3-carbonitrile, 4-bromo-2-(p-chlorophenyl)-5-(trifluoromethyl)-] are present at the highest level. In liver and kidney the parent, CL 303,268, CL 152,835/M-6 (i.e. Acetic acid, {[2-(p-chlorophenyl)-3-cyano-5-(trifluoromethyl)pyrrol-1-yl]methoxy}-) and CL 325,157/M-6A (i.e. Acetic acid, {[3-bromo-5-(p-chlorophenyl)-4-cyano-2-(trifluoromethyl)pyrrol-1-yl]methoxy}-) are present at the

highest levels. In light of the low dietary burden for poultry from the proposed use on cotton a decision on which moieties should be regulated **if any** is deferred pending HED Metabolism Committee review.

5. The nature of the residue in ruminants is adequately understood and consists primarily of the parent in muscle, fat and milk. In addition to the parent numerous Pirate metabolites have been identified. In the liver and kidney the metabolites CL 325,195 [i.e. 2-Pyrrolidine-3-carbonitrile, 2-(p-chlorophenyl)-5-hydroxy-4-oxo-5-(trifluoromethyl)-] and CL 322,250 [i.e. Pyrrole-2-carboxylic acid, 3-bromo-5-(p-chlorophenyl)-4-cyano-] were present at the highest level as well as the parent, other metabolites and conjugates. A decision on which moieties should be regulated is deferred pending HED Metabolism Committee review. The petitioner has proposed regulating only the parent for animal commodities.

6. a. Adequate analytical methods for Pirate in/on cottonseed are available to support the proposed permanent tolerance pending completion of HED Metabolism Committee review.

   b. Pending review by FDA, multiresidue data for the parent Pirate appear adequate. Additional multiresidue data for metabolites may be needed pending HED Metabolism Committee review.

7. Pending completion of a successful EPA Method Validation, submission of a revised method including a procedure for analyzing Pirate in milk fat, adequate method radiovalidation data, and HED Metabolism Committee review and agreement to regulate only the parent adequate analytical methods for Pirate in animal commodities are available to support the proposed permanent tolerances.

8. a. CBTS concludes that Pirate is stable in cottonseed up to 23 months when stored frozen.

   b. Considering the stability of Pirate in cottonseed stored frozen up to 23 months, CBTS also concludes that storage stability data on cottonseed processed fractions stored frozen up to four months, as in the original processing study, are not required.

   c. The storage stability data indicate that Pirate is stable in milk up to 3 months when stored frozen and up to four days when stored in a refrigerator.

d. In the poultry metabolism study the metabolites CL 152,835/M-6 and CL 325,157/M-6A were not stable in liver stored frozen for 10 months. This is not an issue at this time since tolerances on poultry commodities are not likely to be needed.

e. Additional data on the storage stability of the parent and any metabolites which need to be regulated are needed before a determination of the stability of parent and metabolite residues in muscle and fat samples can be made.

9. a. The cottonseed field trial data are adequate to support the proposed permanent tolerance for Pirate of 0.5 ppm in/on cottonseed harvested 21 days following the last of 5 foliar broadcast applications of Pirate, not to exceed a total seasonal maximum application rate of 1.05 lb ai/A.

b. Per Table II (September 1995) residue data on cotton gin byproducts (RAC) are required for Pirate. At least 3 field trials for each type of harvesting (stripper and picker) are needed, for a total of 6 field trials. These data may be provided on a conditional basis. Based on ^{14}C residue data CBTS anticipates a tolerance above 0.5 ppm for Pirate in/on cotton gin by-products will be required.

10. a. An additional cottonseed processing study is required to resolve questions concerning the diminution of the parent during processing. The dark color of the refined oil from the original processing study may indicate excessive temperatures during refining and accordingly for the new processing study a lower and consistent temperature during oil recovery is suggested (i.e. 165-175°F).

b. A final decision on the need for feed/food additive tolerances for Pirate is deferred pending the submission of an acceptable cottonseed processing study.

11. The confined crop rotation study indicates that residues of Pirate and or metabolites CL 312094 and CL 325195 at 0.01-0.02 ppm are possible in rotated crops with a 30 day plant back interval. However, since the study was conducted at approximately 2X the current proposed use rate of 1.05 lb ai/A/season with application made to bare ground (a worst case), at a plant back interval of 60 days or later all residue components in all rotated crops should be less than 0.01 ppm. Accordingly, at the current proposed use rate of 1.05 lb ai/A/season field rotational crop studies are not required provided a revised label specifying a 60 day plant back interval for all rotated food/feed crops is submitted.

12. a. Clearly the highest Pirate residue levels from the ruminant feeding study occurred in fat tissue at approximately 25X residue levels in muscle tissue. Furthermore, the ^{14}C goat milk fat study verified that Pirate concentrates in milk fat as well.

b. The ruminant feeding study is tentatively acceptable pending HED Metabolism Committee review and adequate muscle and fat storage stability data as referenced under Conclusion 8. e.

13. a. Using the data from the seven day poultry metabolism study one would estimate the highest residue in poultry at 0.005 ppm which would likely not be detectable. CBTS concludes that residues of Pirate are not likely to be found in poultry commodities, based on the feeding levels of the metabolism studies and the resulting residues. Therefore, a poultry feeding study and tolerances on poultry commodities are tentatively not required for the proposed use on cotton, pending HED Metabolism Committee review.

b. Based on a best estimate of possible residues in cotton gin byproducts tolerances for Pirate in ruminant commodities of 0.05, 0.75, 0.02, 0.50 and 0.05 ppm, respectively for milk, milk fat, meat, fat and meat byproducts of cattle, goats, hogs, horses, and sheep are required. A final determination of the secondary residues in meat, milk, poultry and eggs must be deferred pending HED Metabolism Committee review.

c. A revised Section F is needed proposing the animal commodity tolerances listed above.

d. In the poultry metabolism study the metabolites CL 152,835/M-6 and CL 325,157/M-6A were not stable in liver stored frozen for 10 months. This is not an issue at this time since tolerances on poultry are not likely to be needed.

14. An International Residue Limit Status sheet is included in this review as Attachment 3. Since no Codex, Canadian, or Mexican limits/tolerances have been established for Pirate, there are no compatibility problems at this time.

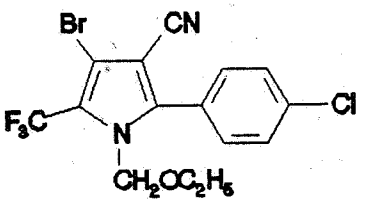
Recommendations

At this time CBTS recommends against establishing the proposed permanent tolerances of 0.50, 0.01, 0.15, 0.01, and 0.10 ppm for Pirate/Alert in/on cottonseed, milk, milk fat, meat and meat by-products, respectively for the reasons cited in Conclusion Nos. 1, 2, 3a, 4, 5, 6 a, b, 7, 8 e, 10 a, b, 11, 12 b, and 13 a, b, and c above.

Reviewer Aids/Detailed Considerations

Product Chemistry

PIRATE



Per CBTS's previous review of PP#3G4224 (see 1/26/94 memo of G. Otakie) adequate product chemistry data were available for a temporary tolerance for Pirate in/on cottonseed. Updated product chemistry and manufacturing process data have been reviewed by CBTS and product chemistry data gaps requiring resolution for a permanent tolerance identified. The results of the product chemistry review are included as **Attachment 1** and the Confidential Appendix is included as **Attachment 8.**

Most product chemistry data requirements for the TGAI have been satisfied. Preliminary analysis data reflects pilot as well large scale production. A revised CSF is required including certified limits for all impurities present at or above 0.1 w/w%. Accordingly, pending submission of a revised CSF, adequate product chemistry data are available for the subject proposed permanent tolerances for Pirate in/on cottonseed and ruminant commodities. Also, the petitioner should submit verification of an ANSI approved common name if one has been obtained.

Residue Chemistry

Proposed Use

Formulation

The proposed formulation for the subject temporary tolerance is Alert Insecticide-Miticide with 21.44% or 2 lbs ai/gal [4-bromo-2-(chlorophenyl)-1-(ethoxymethyl)-5-(trifluoromethyl)-1H-pyrrole-3-carbonitrile] and 78.56 % inert ingredients.

Labels

Two labels were included with the current submission. Pirate insecticide-Miticide 30.83 % ai (i.e. 3.0 lbs ai/gallon) and Alert Insecticide-Miticide 21.44% ai (i.e. 2.0 lbs ai/gallon). To assure a uniform spray solution, containers should be agitated prior to and during use. Pirate/Alert is to be applied with ground or aerial equipment with the minimum spray volumes of 5 gal/A and 2 gal/A, respectively. The labels include a restriction against using the product in any type of irrigation system. The maximum single application rate is 0.35 lbs ai/A (i.e. 15 fl oz/A) with the maximum application rate per crop year of 1.05 lb ai/A (i.e. 45 fl. oz.) with a 21 day PHI. Although a minimum interval between applications is not included the labels suggest proposed application intervals be determined as needed by scouting or at 5-7 day intervals. The label contains restrictions against grazing livestock on treated fields, a crop rotation statement not to plant small grain crops within 60 days of the last application and spray drift precautions. However, CBTS has concluded from the confined crop rotation study that the 60 day crop rotation restriction should apply to all food or feed crops.

CBTS notes that the maximum application rate has been reduced in the subject petition from 2.0 lbs ai/A/season in the temporary tolerance PP#3G4224 to 1.05 lbs ai/A/season.

Nature of the Residue in Plants

Cotton

Metabolism of pirate in cotton was discussed under PP#3G4224 (see 1/26/94 memo of G. Otakie). The parent was the major component identified and accounted for 63.7 and 57.7% of the TRR in cotton linters for 2-pyrrrole-^{14}C and phenyl-^{14}C, respectively. American Cyanamid (1993; MRID 42770234) submitted data depicting the metabolism of [2-pyrrole-^{14}C]-labeled and uniformly ring-labeled [phenyl-^{14}C]Pirate in cotton.

The TRR in foliage and cottonseeds following five applications of [^{14}C]Pirate are presented below:

Total radioactive residues in/on cotton foliage, seed meal, and linters following five applications of [2-pyrrole-^{14}C]- or [phenyl-ring-labeled-^{14}C]Pirate at a seasonal rate of ~2 lb ai/A (2x the current proposed rate).

Matrix/Sampling Interval	[^{14}C]Pirate equivalents (ppm)	
	[2-pyrrole-^{14}C]	[phenyl-^{14}C]
Foliage 1st Application	38.49	48.30
3rd Application	93.48	102.86
5th Application	122.13	131.79
Seed Meal	0.15	0.18
Linters	0.12	0.13
Total Cottonseed	0.27	0.31

Residues in foliage of [pyrrole-^{14}C]- and [phenyl-^{14}C] Pirate-treated plants were lowest after the first sampling interval and increased at each succeeding sampling interval to maximums of 122.13 and 131.79 ppm, respectively. Residues in foliage were not further characterized. Cottonseed meal and linters were analyzed for TRR separately. Apparent TRR in control samples of cottonseed and cotton foliage were <0.01 ppm.

The parent compound, Pirate, was the major radioactive component in cottonseeds from [2-pyrrole-^{14}C]Pirate-treated plants, accounting for 54.8% of the TRR (0.08 ppm) in meal and 63.7% of the TRR (0.08 ppm) in linters. In cottonseed from [phenyl-^{14}C]Pirate-treated plants, the parent compound accounted for 73.9% of the TRR (0.13 ppm) in meal and 57.7% of the TRR

(0.08 ppm) in linters. **Expressed on a whole seed basis, the parent compound accounted 59.3-67.7% of the TRR (0.16-0.21 ppm).** Expressed in the same manner, ^{14}C-residues in cottonseed oil and non-extractable residues accounted for <0.01-0.01 ppm (≤3.2% TRR) and 0.03 ppm (9.7-11.1% TRR), respectively.

An unidentified polar metabolite (designated Unknown-1) accounted for 4.7-7.1% of the TRR (0.01 ppm) in meal and 3.4-3.5% of the TRR (<0.01 ppm) in linters. Characterization of Unknown-1 indicated that it was not conjugated to glucose and it did not contain a functional hydroxyl group. Numerous minor radioactive peaks (≤0.01 ppm) also were observed in organosoluble extracts of meal and linters. The registrant made several attempts to identify these peaks using reference standards of N-dealkylation, dehalogenation, hydroxylation, and carboxylation analogs of the parent compound; however, none of these peaks corresponded to these reference standards. No raw data were provided for these analyses. Radiochromatograms were presented without any supporting raw data for radioanalysis. Therefore, the amount of radioactivity contained in each individual unknown peak could not be independently determined.

For purposes of the EUP and temporary tolerance petition, the qualitative nature of the residue was adequately understood and the residue to be regulated was the parent compound Pirate.

However, for establishment of a permanent tolerance the qualitative nature of the residue in cottonseed was not adequately understood. Raw data were requested showing the amount of radioactivity in individual unknown peaks detected in organosoluble extracts of cottonseed meal and linters. In addition, data must be submitted demonstrating the stability of ^{14}C-residues in samples stored for 545 days (~18 months). The petitioner indicated that the storage intervals for cottonseed prior to analysis ranged from 40 to 501 days. HPLC chromatograms showing that 40- and 501-day metabolite profiles are similar would fulfill this requirement.

In response the petitioner submitted the following study:

<u>Addendum to CL 303,630: Metabolism of Carbon-14 Labeled CL 303,630 in Cotton under Field Conditions: MRID No. 434928-53.</u>

The report includes raw data on the amount of radioactivity in HPLC chromatograms of organosoluble fractions of cottonseed meal and linters from both Pyrrole-^{14}C and Phenyl-^{14}C Pirate treatment. The data indicates that each of the fractions containing unidentified residues accounted for only a small portion of the TRR. The fractions with the largest portion of the TRR contained the parent. HPLC radiochromatograms showing the 88, 151 and 448-479-day metabolite profiles of the

organoextractable fractions for cottonseed meal and linters were also provided. The data indicates the metabolic profiles were similar from 88 to 479 days.

Conclusion - Nature of the Residue in Cotton

The nature of the residue in cotton is adequately understood. Pending HED Metabolism Committee review, the residue of concern in cotton consists of the parent Pirate.

Citrus

The nature of the residue in citrus was discussed in PP#5G04507 (see 8/10/95 memo of G. Kramer). In brief, Pirate/Alert was the major component of the residue identified accounting for 71-77% of the TRR in 7 day PHI orange samples. Other minor metabolites included CL 303,268 at 3% of the TRR, CL 322,250 at 1% of the TRR, and CL 325,195 at 2% of the TRR. A total of 74-78% of the TRR was identified in orange samples representing a 7 day PHI. Unidentified peaks, none of which exceeded 0.01 ppm, accounted for up to 20.2% of the TRR.

Nature of the Residue in Animals

Metabolism of Pirate In Poultry

Metabolism of pirate in poultry was discussed under PP#3G4224 (see 1/26/94 memo of G. Otakie). Based on a poultry diet consisting of 8% cottonseed meal plus soap stock (at 0.5 ppm proposed tolerance) and 92% grain (at 0 ppm), the maximum theoretical daily dietary intake of Pirate was proposed to be 0.04 ppm. In accordance with Table II (September 1995) there have been revisions in the livestock diet for cotton feedstuffs (see Secondary Residues).

Laying hens were orally dosed with [phenyl-^{14}C]- and [pyrrole-^{14}C]Pirate at 3.02-3.10 ppm and 14.42-15.04 ppm of Pirate in the diet. The distribution of TRR in eggs and tissues from both labeled samples was similar. In the high dose groups, the TRR in eggs increased from <0.01 to 0.42 ppm by day 7. Radioactive residues ranged from 0.02 ppm in muscle to 0.52 ppm in kidney and were highest in liver (1.25-1.31 ppm).

The data indicate that laying hens metabolize [phenyl-^{14}C]- and [pyrrole-^{14}C]Pirate in a similar manner yielding similar metabolites. The parent compound and eight metabolites were detected in eggs and tissues; two metabolites (AC 8508-78-BA/BB) were identified as a mixture. The total identified/characterized ^{14}C-residues accounted for 89% TRR (eggs), ≥87% TRR (liver), ≤89% TRR (kidney), 94% TRR (skin/fat), and ≥62% TRR (muscle). The petitioner identified the parent compound as the major metabolite from [phenyl-^{14}C]- and [pyrrole-^{14}C]Pirate samples in eggs (40-42% TRR, 0.16-0.17 ppm), skin/fat (79-84% TRR, 0.29-0.39 ppm), and muscle (25-31% TRR, <0.01-0.01 ppm).

In the liver and kidney, CL 303,268 was the major metabolite, which amounted to 7-17% TRR (0.09-0.21 ppm) and 17-25% TRR (0.09 ppm), respectively. Three other metabolites (CL 325,195, AC 8508-50-C, and AC 8508-33-B1) common to [phenyl-^{14}C]- and [pyrrole-^{14}C]-labeled samples of Pirate and two additional metabolites (AC 8508-78-BA/BB and CL 322,250) derived from [phenyl-^{14}C]Pirate were also identified. The chemical nature of Pirate, AC 8508-78-BA/BB, AC 8508-50-C, AC 8508-33-B-1, and CL 303,268 were confirmed by LC/MS or GC/MS. Components of unknown nature occurring at 3-10% TRR (0.01-0.05 ppm) included M-1, M-4 and M-10 and other unknowns, occurring each at <0.01 ppm were also characterized. Non-extractable residues in eggs and tissues accounted for 2-39% of the TRR (<0.01-0.17 ppm).

The metabolism of orally administered [phenyl-^{14}C]- and [pyrrole-^{14}C]Pirate in laying hens is similar to that of lactating goats. In both species, the main metabolic route for Pirate metabolism is N-dealkylation and ring hydroxylation.

Samples from the goat and hen metabolism studies were not analyzed using the proposed enforcement method. Radiovalidation of the method used for data collection and enforcement purposes using samples from the metabolism studies was requested.

CL 303,630: Metabolic Fate of [14C] CL 303,630 in Blood, Eggs and Tissues of Laying Hens; MRID Nos. 434928-54.

The final report on the metabolism of Pirate in poultry was included in the current submission. The worst case poultry diet was revised consisting of 20% cottonseed meal and soapstock at the proposed 0.5 ppm tolerance level . Therefore the maximum theoretical daily dietary intake of Pirate in the poultry diet was estimated at 0.11 ppm in the poultry diet (i.e. 0.5 ppm x 20%/.89DM =.11 ppm). Consequently, the 0, 3.02-3.10, and 14.42-15.04 ppm dosage rates would represent 0, 27 to 28X and 131 to 137X for the control low dose and high dose groups. In accordance with Table II (September 1995) there have been revisions in the livestock diet for cotton feedstuffs (see Secondary Residues).

To provide kidney and liver samples for metabolite identification, an additional 25 laying hens were divided into a control and two treatment groups (5 in the control and 10 in each dosage group) with the treatment groups dosed at 15.9 ppm phenyl ^{14}C or 16.9 ppm 2-pyrrole ^{14}C. Based on the diet above 15.9 and 16.9 ppm dosage group represent 145 and 154X, respectively. Tissue and egg samples were stored frozen for a maximum of 324 days before extraction and analysis. A summary of the total radioactive residue TRR for the poultry metabolism study and the highest ppm equivalents and % TRR is included in **Attachment 2.**

Based on the identity of metabolites, metabolism of Pirate takes place at the phenyl ring, and on the substituents of the pyrrole ring. Fragmentation of the two rings was not evident. Pirate undergoes extensive metabolism in the hen involving modification of the phenyl ring and the substituents of the pyrrole ring. The metabolic pathways of Pirate in the hen include N-dealkylation, dehalogenation, ring hydroxylation and oxidation of the N-alkyl group. components. The metabolic pattern of Pirate in the hen is qualitatively similar to that in the goat and rat (see **Attachment 3**).

Conclusion

The nature of the residue in poultry is adequately understood and consists primarily of the parent in muscle and fat. In addition to the parent numerous Pirate metabolites have been identified. In eggs the parent and its N-dealkylation metabolite CL 303,268 [i.e. Pyrrole-3-carbonitrile, 4-bromo-2-(p-chlorophenyl)-5-(trifluoromethyl)-] are present at the highest

level. In liver and kidney the parent, CL 303,268, CL 152,835/M-6 (i.e. Acetic acid, {[2-(p-chlorophenyl)-3-cyano-5-(trifluoromethyl)pyrrol-1-yl]methoxy}-) and CL 325,157/M-6A (i.e. Acetic acid, {[3-bromo-5-(p-chlorophenyl)-4-cyano-2-(trifluoromethyl)pyrrol-1-yl]methoxy}-) are present at the highest levels. In light of the low dietary burden for poultry from the proposed use on cotton a decision on which moieties should be regulated **if any** is deferred pending HED Metabolism Committee review.

Metabolism of Pirate in the Goat

Goats

Metabolism of pirate in the ruminant was discussed under PP#3G4224 (see 1/26/94 memo of G. Otakie). In brief, American Cyanamid (1993; MRID 42770235) submitted data depicting the metabolism of [^{14}C]Pirate in lactating goats. Four goats, two per test substance, were dosed orally once a day for seven consecutive days. One goat served as a control. The low and high doses represented a daily feeding level of 3.0 and 17.9 ppm for [phenyl-^{14}C]Pirate and 3.16 ppm and 16.4 ppm for [pyrrole-^{14}C]Pirate.

Based on a goat diet consisting of 20% cottonseeds, 15% cottonseed meal, and 10% cottonseed hulls plus soapstock (each at the 0.5 ppm proposed tolerance for cottonseed) and 55% alfalfa forage (at 0 ppm), the maximum theoretical daily dietary intake of Pirate is 0.23 ppm. Therefore, the low and high doses of [^{14}C]Pirate corresponded to a maximum daily dietary burden of ca. 13x and 75x, respectively.

Triplicate milk samples were analyzed directly by LSC and tissue subsamples were analyzed by LSC following combustion. The limit of detection for the radioassays was 0.01 ppm. Sample calculations and raw data were submitted. Both [phenyl-^{14}C]- and [pyrrole-^{14}C]-labeled samples showed similar distribution patterns of radioactive residues in tissues and milk. During the testing period, radioactive residues in milk samples from the low dose group remained at 0.02 ppm. In the high dose group, the TRR in milk samples increased during the test period from 0.03 ppm to 0.07 ppm. Radioactive residues were highest in liver (1.45-1.46 ppm); the ^{14}C-residues in other tissues ranged from 0.03-0.05 ppm in muscle to 0.62-0.94 ppm in kidney.

Milk and tissue organic fractions were analyzed by reversed-phase HPLC using a water:ACN (1:9; v:v) solvent system containing 1% glacial acetic acid (v:v). Reference compounds were visualized using a UV detector at 254 or 260 nm and radioactivity in the HPLC fractions were quantified by LSC. For confirmation

of metabolites resolved by HPLC, 1-D or 2-D TLC analyses were performed on silica gel plates using several solvent systems.

Approximately 79% of the administered radioactivity in Pirate was excreted by lactating goats in the urine and feces. The parent compound and eight metabolites were detected in milk and tissues; two metabolites (AC 8508-78-BA/BB) were identified as a mixture. The total identified/characterized ^{14}C-residues were: ≥83% TRR (milk), ≥65% TRR (liver), ≥81% TRR (kidney), ≥92% TRR (fat), and ≥45% TRR (muscle). Pirate per se was the major metabolite detected in milk (24.7-68.4% TRR), kidney (8.3-9.6% TRR), fat (46.9-78.0% TRR), and muscle (28.7-52.0% TRR). The major metabolite detected in liver was CL 325,195 (3.9-13.5% TRR, 0.06-0.20 ppm). Unknowns 1, 2, 4, 10, and 11, accounting for <0.1-19.2% of the TRR (<0.01-0.1 ppm), were also detected. Non-extractable residues in milk and tissue samples accounted for 4.4-68.0% of the TRR (<0.01-0.24 ppm). Non-extracted ^{14}C-residues from muscle samples (52.0-68.0% TRR, 0.02-0.03 ppm) were not further examined.

The data indicated that Pirate is metabolized in lactating goats by N-dealkylation, dehalogenation, ring hydroxylation, and/or oxidation of the terminal alkyl group. Pirate is N-dealkylated to form CL 303,268, which undergoes ring hydroxylation producing CL 325,195, AC 8944-45, and AC 8508-78-BB. The terminal alkyl group of Pirate is oxidized and dehalogenated to produce AC 8805-31-2-B. All the identified metabolites contain ^{14}C-labels from the [phenyl-^{14}C]- and [pyrrole-^{14}C]Pirate, indicating no fragmentation of the phenyl and pyrrole rings during the metabolism of Pirate in goats.

<u>CL 303,630: Metabolic Fate of Carbon-14 Labeled CL 303,630 in the Tissues and Milk of a Lactating Goat;</u> MRID No. 434928-55.

The final report on the metabolism of Pirate in the goat was included in the current submission. In addition to the low and high dose treatment group with two goats per group discussed above this report included goats treated in a second experiment, L-2461-A, to complete the identification/characterization of residues in the liver and kidney, and to provide tissue and milk for radiovalidation of the proposed analytical method (note: the method radiovalidation data for ruminants will be submitted in a separate report). A total of three goats were used with one goat orally dosed daily with 30 mg phenyl ^{14}C and one with 30 mg pyrrole-^{14}C both for a total of seven days and one control goat. The doses represented a daily feeding level of 17.3 and 24.8 ppm for the phenyl labelled and pyrrole label treatments, respectively.

The worst case goat diet was revised to consisting of 25% cottonseeds, 15% cottonseed meal, and 15% cottonseed hulls plus 5% soapstock (each at the 0.5 ppm proposed tolerance for cottonseed) and 40% alfalfa forage (at 0 ppm), with the maximum theoretical daily dietary intake of Pirate estimated at 0.34 ppm. Therefore, based on the above diet the low and high doses of [^{14}C]Pirate corresponded to a maximum daily dietary burden of 9X (low dose) 48 to 53X (high dose) in experiment L-2461, and 51 to 73X in experiment L-2461A. Meat and milk samples from Experiment L-2461A were stored frozen for a maximum of 63 days before extraction and analysis. A summary of the total radioactive residue TRR for the goat metabolism study and the highest ppm equivalents and % TRR is included in **Attachment 4.**

Pirate undergoes extensive metabolism in the goat involving modification of the phenyl ring and the substituents of the pyrrole ring. The metabolic pathways of Pirate metabolism include N-dealkylation, dehalogenation, and hydroxylation of both the phenyl and pyrrole ring, hydroxylation and oxidation of the N-alkyl group and conjugation to endogenous components. However, there is no fragmentation of the molecule. The metabolic pathway of Pirate in the lactating goat is similar to that in the laying hen and laboratory rats (see **Attachment 3**)

Conclusion

The nature of the residue in ruminants is adequately understood and consist primarily of the parent in muscle, fat and milk. In addition to the parent numerous Pirate metabolites have been identified. In the liver and kidney the metabolites CL 325,195 [i.e. 2-Pyrrolidine-3-carbonitrile, 2-(p-chlorophenyl)-5-hydroxy-4-oxo-5-(trifluoromethyl)-] and CL 322,250 [i.e. Pyrrole-2-carboxylic acid, 3-bromo-5-(p-chlorophenyl)-4-cyano-] were present at the highest level as well as the parent, other metabolites and conjugates. A decision on which moieties should be regulated is deferred pending HED Metabolism Committee review. The petitioner has proposed regulating only the parent for animal commodities. CBTS is not opposed to utilizing the parent as a marker compound in meat by-product tissues where it accounts for less than half the identified residue provided the HED Metabolism Committee has no objection.

Analytical Methodology - Plants

Per PP#3G4224 (see memo of 5/16/94) a satisfactory method trial has been conducted by EPA's Analytical Chemistry Laboratory for Method M 2216 for Pirate in/on cottonseed with some minor revisions required. In brief residues of Pirate are extracted from cottonseed with a methanol/water mixture, and cleaned up using C-18 solid phase extraction. Quantitation is done using

gas chromatography equipped with electron capture detector and a mega bore capillary column.

GC Analytical Method, GC/MS Confirmatory Method, Carbon-14 Extractability and FDA Multiresidue Method: MRID No. 434928-56

Revised Method

A copy of the revised analytical methodology M 2216.01 for Pirate residues in/on cottonseed was submitted. The revised method includes some minor revisions recommended in the EPA Method validation (see PP#3G4224 5/16/94 memo of G. Otakie). The method limit of quantitation is 0.05 ppm but method sensitivity is reported at 0.005 ppm.

Confirmatory Method

A copy of analytical Method M 2418, a GC/MS confirmatory method for Pirate residues in cottonseed was also submitted.

Extraction Procedure

Data were submitted on the extraction of Pirate residues (i.e. pyrrole label ^{14}C) in cottonseed samples from the cottonseed metabolism study using the extraction procedure in Method M 2216. The extraction procedure (i.e. methanol:water [85:15] and a 5-minute extraction time) yielded a total radioactive extractability of 40%. The report indicates that although the extraction efficiency using M 2216 is lower than reported in the metabolism report where multiple extractants (i.e. primarily hexane followed by methanol) were used, the Pirate residue level was similar in both cases. Specifically, the results from the metabolism study show that 57% (0.16 ppm) of the TRR in cottonseed was Pirate and in this study where 40% of the TRR was extracted with methanol the Pirate residue was 0.11 ppm.

We note the reported residue of pirate in the organosoluble fraction from extraction of cottonseed treated with pyrrole label ^{14}C Pirate was 0.08 ppm which is less than the 0.11 ppm reported in this extraction study. However, in light of possible sampling variation and the significantly higher solubility of Pirate in methanol than hexane (i.e. 7.09 vs. 0.89 g/110 ml) CBTS is satisfied that the methanol extraction scheme is adequate.

Conclusion

Pending completion of HED Metabolism Committee review adequate analytical methods for Pirate in cottonseed are available to support the proposed permanent tolerances.

Multiresidue Data

Multiresidue data for Pirate were submitted. Protocols A and B were not applicable to pirate. In Protocol C Pirate gave a good response and a good peak with the electron capture detector on three different GC columns. In Protocol D using pears as a nonfatty food representative the 5% OV-101 column gave the greatest sensitivity at 0.05 and 0.50 ppm. In Protocol E Pirate eluted well on Florisil in both the ethyl ether/petroleum ether system and the alternate hexane/acetonitrile/methylene chloride system and gave excellent recovery. The multiresidue data will be forwarded to FDA. Pending review by FDA the multiresidue data for the parent Pirate appear adequate. Additional multiresidue data for metabolites may be needed pending HED Metabolism Committee review.

Analytical Methodology - Animals

Analytical methods for Pirate on ruminant animal commodities were validated by Independent Method Validation and forwarded to ACB/BEAD for EPA Method Validation per a review of PP4F4346 (see 10/95 memo of G. Otakie). There are three different Pirate analytical methods proposed for milk, muscle/fat and liver/kidney.

M 2395

Parent residues are isolated from milk and purified using acetone precipitation, methylene chloride partition and solid phase extraction techniques. Residues are measured using gas chromatography (GC) with electron capture detection and residues are calculated as parent by direct comparison of sample peak height to that of an external standard. The validated sensitivity of the method is 10 ppb.

M 2398

Parent residues are extracted from muscle with methanol and from fat with acetonitrile. Residues are isolated by hexane partition and purified using solid phase extraction techniques. Residues are measured using GC with electron capture detection and calculated as parent by direct comparison of sample height to that of an external standard. The validated sensitivity of the method is 10 ppb.

M 2405

Parent residues are extracted from cattle liver and kidney tissues with acetonitrile. Residues are isolated by hexane partition and are purified using solid phase extraction

techniques. Residues are measured using GC with electron capture detection and calculated as parent by direct comparison of sample height to that of an external standard. The validated sensitivity of the method is 50 ppb.

Conclusion

Pending completion of a successful EPA Method Validation, submission of a revised method including a procedure for analyzing Pirate in milk fat, adequate method radiovalidation data (i.e. referenced in goat metabolism study to be submitted in a separate report), and HED Metabolism Committee review and agreement to regulate only the parent adequate analytical methods for Pirate in animal commodities are available to support the proposed permanent tolerances.

Residue Data

Storage Stability Data

Freezer Stability of Residues of CL 303,630 in Cottonseed and Cow's Milk; MRID No. 434928-58

Samples of control cottonseed were fortified with 0.25 ppm parent and stored frozen at -10 to -20 °C in cardboard containers. Duplicate fortified cottonseed samples and a control sample were removed and analyzed using Method 2216 at 0, 3, 6, 12, 18 and 23 month intervals. Parent recoveries in the cottonseed were 93, 96 and 99% at 12 months and 91, 91, and 96% at 23 months. The storage stability data indicate that Pirate is stable in cottonseed up to 23 months when stored frozen.

Samples of cow's milk were fortified with 0.05 ppm parent and stored frozen at $\pm$10 to $\pm$20 °C in glass bottles. Duplicate fortified cottonseed samples and a control sample were removed and analyzed using Method 2395 at 0, 1, 2 and 3 month intervals. Parent recoveries in the milk were at 78, 86, and 84% at 3 months. In another study parent fortified milk was stored in a refrigerator for four days and analyzed with parent recoveries of 72, 74, and 76% after four days. The storage stability data indicate that Pirate is stable in milk up to 3 months when stored frozen and up to four days when stored in a refrigerator.

Cottonseed samples from the field residue and processing studies were stored frozen for 1.6-9.8 months prior to analysis, and processed commodities were analyzed after approximately 4 months of frozen storage. Accordingly, the Pirate residues in these studies were stable for the storage interval utilized. Adequate storage stability data for Pirate in cottonseed is available. Considering the stability of the parent in cottonseed

stored frozen up to 23 months CBTS concludes that storage stability data on cottonseed processed fractions stored frozen up to four months as in the original processing study are not required.

In the poultry metabolism study the metabolites CL 152,835/M-6 and CL 325,157/M-6A were not stable in liver stored frozen for 10 months. This is not an issue at this time since tolerances on poultry commodities are not likely to be needed.

The goat metabolism study included a HPLC ^{14}C radioactivity profile of HCl **digest** of liver after 8 months frozen storage. The profile was similar but total dpm was approximately 20% lower after the 8 month interval. Meat samples from the cattle feeding study were stored frozen up to 90 days before analysis. Additional data on the storage stability of the parent and any metabolites (i.e. which are determined to need regulation if any) are needed before a final determination of the stability of parent and metabolite residues in meat and fat samples can be made.

Conclusion

CBTS concludes that Pirate is stable in cottonseed up to 23 months when stored frozen. Considering the stability of Pirate in cottonseed stored frozen up to 23 months CBTS also concludes that storage stability data on cottonseed processed fractions stored frozen up to four months as in the original processing study are not required. The storage stability data indicate that Pirate is stable in milk up to 3 months when stored frozen and up to four days when stored in a refrigerator. Additional data on the storage stability of the parent and any metabolites (i.e. which are determined to need regulation, if any) are needed before a determination of the stability of parent and metabolite residues in meat and fat samples can be made.

Magnitude of Residue - Plants

Cotton Field Trials

Cotton field trial data were discussed under PP#3G4224 (see 1/26/94 memo of G. Otakie). Applications were made using ground equipment at 10-20 gal/A, with the last application being made when 5 to 70% of the bolls were opened. Cottonseeds were ginned after harvest and stored frozen at -24 to -18 C. Samples analyzed at ACC were stored frozen at -20 C for 7.6-8.6 months prior to analysis. Samples from the 1991 and 1992 trials analyzed by ABCL were shipped overnight and stored frozen for 7.4-9.8 and 1.6-3.5 months, respectively, prior to analysis.

Cottonseeds were analyzed for residues of parent Pirate using GC/ECD method M-2216, which has a detection limit of 0.05 ppm. Apparent residues of Pirate in/on 26 control samples of cottonseeds were nondetectable (<0.05 ppm). Concurrent method recoveries from 30 control samples fortified with Pirate at 0.05-2.53 were 84-103%. Uncorrected residues of Pirate in/on treated cottonseeds are presented below:

Residues of Pirate in/on cottonseeds following the last of five foliar broadcast applications, made at 7-day intervals, of the 2 or 3 lb/gal EC formulations at 2 lb ai/A/season (2x).

Trial Year	PTI [a] (Days)	Test States	Number of Samples	Residues (ppm)
1991	**21 [b]**	**AR, CA, LA, MS, TX**	**6**	**<0.05-0.26**
	24	LA	1	0.13
	28	AR, CA, LA, MS	6	<0.05-0.24
	30	TX	1	<0.05
	35	CA, LA, MS	3	<0.05-0.15
	36	LA, TX	2	<0.05, 0.17
	38	MS	1	0.12
	39	AR	1	0.16
1992	0	LA	1	1.51
	7	LA	1	0.61
	14	LA	1	0.80
	21	**LA**	**1**	**0.32**
	28	LA	1	0.31

[a] Posttreatment interval.

[b] AR, CA, LA, MS, TX Pirate residues were 0.09, 0.26, 0.23, <0.05 to 0.24, and <0.05 ppm, respectively at a 21 day PHI. Data representing the proposed PHI of 21 days are bolded.

Geographic representation was adequate for the purposes of the EUP and the available data were determined to be adequate for the proposed temporary tolerance of 0.5 ppm in/on cottonseed.

For a permanent tolerance additional field trial data representing commercial application procedures are needed. A minimum of an additional 5 field trials (i.e. including two replicate samples per site) in geographically representative areas are required with at least one field trial conducted in either Georgia, North Carolina or South Carolina.

Cl 303,630: Residues of Cl 303,630 in Ginned Cottonseed and in Processed Cottonseed Commodities; MRID No. 434828-60.

American Cyanamid submitted additional residue field trial data from ten field trials conducted in 1992 in AR(1), CA(1), LA(2), MS(2), TX(1), AL(1), SC(1) and GA(1) depicting residues of Pirate in/on cottonseeds following the last of five foliar broadcast applications of the 3 lb/gal EC formulations (EPA file No. 241-EUP) each at 0.4 lb ai/A/application. The total seasonal application rate was 2 lb ai/A (approximately 2x the current maximum proposed seasonal application rate). Applications were made at 7-day intervals using ground equipment at 10-20 gal/A, with the last application being made when 5 to 70% of the bolls were opened.

Cottonseeds from the additional 1992 trials were harvested from 0-28 days after the final application. Cottonseeds were ginned after harvest and stored frozen. Residue samples were stored frozen for up to five months prior to analysis by ABC Laboratories.

Cottonseeds were analyzed for residues of parent Pirate using GC/ECD method M-2216, which has a detection limit of 0.05 ppm. Apparent residues of Pirate in/on the field trial control samples of cottonseeds were nondetectable (<0.05 ppm). Concurrent method recoveries for Pirate from fortified control samples ranged from 96-106%. Uncorrected residues of Pirate in/on treated cottonseeds are presented below:

Residues of Pirate in/on cottonseeds following the last of five foliar broadcast applications, made at 7-day intervals, of the 3 lb/gal EC formulations at 2 lb ai/A/season (2x).

Test State/Site	PHI Days	Percentage of open bolls at last application	Pirate Residue - ppm
AR, Shoffner	1	10-20	0.24
"	7	"	0.07
"	14	"	0.09
"	21	"	<0.05
"	28	"	0.06
CA, Tulare County	0.1	25-35	0.32
"	7	"	0.19
"	14	"	0.22
"	21	"	0.10
"	28	"	0.09
LA, Cheneyville)	0.1	0-late bloom	not collected
"	11	"	<0.05
"	14	"	<0.05
"	21	"	<0.05
"	28	"	<0.05
LA, Rosa	0.1	45-55	1.51
"	7	"	0.61
"	14	"	0.80
"	21	"	0.32
"	28	"	0.31
MS, Greenville	0.1	10-20	0.45
"	7	"	0.23
"	14	"	0.20
"	21	"	0.26
	28	"	0.13
MS, Wayside	0.1	0-late bloom	not collected

"	7	"	<0.05
"	14	"	<0.05
"	21	"	<0.05
"	28	"	<0.05
TX, Uvalde	0.1	20	0.10
"	7	"	0.06
"	14	"	0.05
"	21	"	<0.05
"	28	"	<0.05
Al, Grangeburg	0.1	late bloom/cut out	0.24
"	7	"	0.24
"	14	"	0.24
"	21	"	0.11
"	28	"	0.15
SC, Elko	0.1	60	0.97
"	7	"	0.32
"	14	"	0.27
"	21	"	0.23
"	28	"	0.27
GA, Montezuma	0.1	late bloom/cut out	not collected
"	7	"	0.07
"	14	"	<0.05
"	21	"	<0.05
"	28	"	<0.05

Residues of Pirate were ≤0.32 ppm and ≤0.31 ppm in/on cottonseed samples harvested 21 and 28 days, respectively following the last of five foliar broadcast applications of 3 lb/gal EC formulations each at 0.4 lbs ai/A or approximately 2x the proposed current maximum seasonal application rate of 1.05 lbs ai/A/Season. As would be expected the highest cottonseed residues occurred when the last application was made with the highest percentage of cotton bolls open (i.e. Rosa, LA with 45-55% open bolls).

Field Trial Data Conclusion

The cottonseed field trial data are adequate to support the proposed permanent tolerance for Pirate of 0.5 ppm in/on cottonseed harvested 21 days following the last of 5 foliar broadcast applications of Pirate not to exceed a total seasonal maximum application rate of 1.05 lb ai/A.

Cotton Gin Byproducts/Conclusion

Per the Table II update (September 1995) residue data on cotton gin byproducts (RAC) are required for Pirate. Cotton gin byproducts include the plant residues from ginning cotton, and consist of burrs, leaves, stems, lint, immature seeds, and sand or dirt. Cotton must be harvested by commercial equipment (stripper and mechanical picker) to provide an adequate representation of plant residue for the ginning process. At least 3 field trials for each type of harvesting (stripper and picker) are needed, for a total of 6 field trials. These data may be provided on a conditional basis.

Cottonseed Processing Data

Cottonseed processing data were discussed under PP#3G4224 (see 1/26/94 memo of G. Otakie). In brief, American Cyanamid submitted data (1993; MRID 42770238) depicting the concentration of residues of Pirate in cottonseed processed commodities. In a test conducted in LA in the 1992 season, cottonseeds were harvested 14 days following the last of five foliar applications of the 3 lb/gal EC formulation at 0.4 lb ai/A/application (2x). Applications were made at 7-day intervals using ground equipment at 16.5-18.8 gal/A. The last application was made when 45-55% of bolls were opened.

Treated and control samples were harvested, stored frozen at -24 to -19 C, and shipped in a freezer truck to the Engineering Biosciences Research Center of Texas A&M University (Bryan, TX) for processing. Cottonseeds were processed into hulls, meal, crude oil, refined oil, and soapstock using a simulated industrial procedure. The processed fractions were then packed in dry ice and shipped to the analytical laboratory (American Cyanamid, Princeton, NJ). Untreated control and treated samples were analyzed for residues of Pirate using Method M-2274, which has a detection limit of 0.05 ppm. Concurrent method recoveries were 85-102% from six control samples (one sample per commodity) fortified at 0.05 ppm. Apparent residues of Pirate were nondetectable (<0.05 ppm) in each untreated control sample. Uncorrected residues in treated cottonseed were 0.66 ppm. Uncorrected residues were 0.36 ppm in hulls, 0.1 ppm in refined and crude oils, and <0.05 ppm in meal and soapstock.

The submitted processing data were determined to be adequate for a temporary tolerance pending submission of data depicting the frozen storage stability of residues of Pirate in cottonseed processed commodities. The deficiency of the processing study for a permanent tolerance was noted under PP#3G4224 (see 1/26/94 memo of G. Otakie) in that there was a discrepancy in the mass balance for the parent (i.e. significantly higher Pirate residues in the RAC than in any of the processed fractions). A new mass balance providing the disposition of 90 to 100% of the Pirate in the cottonseed used for processing was required.

Addendum to: CL 303,630: Residues of CL 303,630 in Ginned Cottonseed and in Processed Commodities; MRID No. 434928-61.

The study includes the Standard Operating Procedure for small-scale processing of cottonseed at Texas A&M University, original processing data and a cotton material balance. Ginned cotton samples and cottonseed processed samples were stored frozen approximately three and four months, respectively from the time of sampling to analysis. A material balance for the cotton with the weights of the cotton, cottonseed, lint cotton, gin trash, delintered cottonseed, linters, linter motes, flaked cottonseed used for processing, collettes, and meal (presscake) was provided. Notes on miscella refining (untreated sample) indicated that refined oil was very **dark in color** and the oil was contaminated with tap water (treated sample). The temperature for oil recovery from the untreated processed sample was reported as 184°F and no temperature was reported for the treated processed sample.

Cl 303,630: Residues of Cl 303,630 in Ginned Cottonseed and in Processed Cottonseed Commodities; MRID No. 434828-60.

In the study ginned cottonseed (i.e. samples held in frozen storage for 15 months) from the original processing study were reanalyzed and found to contain an average pirate residue of 0.66 ppm which is the same level as was found in the original sample. Additionally, samples of linters and delinted seed samples were analyzed to provide data for a mass balance. Pirate residue levels in linters/lint and delinted seed were 5.66 and 0.49 ppm, respectively. A mass balance for pirate including this additional residue data accounted for 77% of the pirate with the linters accounting for 56% of the residue. Nevertheless, 0.49 ppm of pirate remained in the delinted seed with only 0.1 ppm found in refined and crude oils, and <0.05 ppm in the meal and soapstock. The pirate residue in hulls from the processing study was lower than that in the delinted seed at 0.36 ppm suggesting possibly higher residues in the kernels (from hulling and separation) processed into oil. It appears that there was a diminution of the pirate during the processing into oil from the small scale cottonseed processing study. A new cottonseed processing study is needed.

Conclusion

An additional cottonseed processing study is required to resolve questions concerning the diminution of the parent during processing. The dark color of the refined oil from the original processing study may indicate excessive temperatures during refining and accordingly for the new processing study a lower and consistent temperature during oil recovery is suggested (i.e. 165-175°F).

A final decision on the need for feed/food additive tolerances for Pirate is deferred pending the submission of an acceptable cotton seed processing study.

Crop Rotation

Confined Crop Rotation

CL303,630:Confined Rotational Crop Study with Carbon-14- CL 303,630; MRID No. 434928-51.

An outdoor confined crop rotation study was conducted at Madera, California, during 1991-1992 with either Pirate pyrrole ^{14}C or phenyl ^{14}C label applied to **bare** sandy soil for five successive weeks at an application rate of 0.40 lb ai/A/application or approximately 2X the current proposed seasonal application rate of 1.05 lb ai/A. Rotational crops of leaf lettuce (leafy vegetable), carrot (root crop), barley (small grain) and soybean (legume) were planted at 31, 60, 119, and 364 days after treatment number 5 (DAT-5). All crop samples were immediately frozen and stored frozen until analysis by American Cyanamid. Some lettuce samples were stored frozen up to 29 months before analysis. HPLC radiochromatograms for lettuce over a 29 month period were provided and indicate a similar metabolic profile over the storage interval.

The radioactive residue in rotational crops was moderately extractable ranging from 30.0-93.4% TRR in the combined extracts for all the rotational crops. Terminal residues in the rotational crops contained the parent (CL 303,630) at <0.01-0.13, and metabolites CL 325,195 at <0.01-0.01, and CL 312,094 at <0.01-0.03 ppm, in addition to other minor extractable polar and nonpolar metabolites. **Attachment 5** summarizes the total ^{14}C residue and characterization crop rotation data obtained. The metabolic profile was similar for both the pyrrole ^{14}C or phenyl ^{14}C label and the bond between the phenyl and pyrrole ring apparently remains intact.

The highest residues were 0.13 and 0.07 ppm of parent detected in immature and mature carrot root, respectively at a 31 day plant back interval. However at the 60 day plant back interval parent residues were <0.01 ppm in immature carrot. Parent residue of 0.01 ppm in lettuce at a 31 day plant back was reduced to <0.01 ppm at a 60 day plant back interval. Residue data on barley and soybean matrices from a 31 day plant back interval showed parent residues up to 0.02, 0.01, and 0.01 ppm in barley forage, barley straw and soybean forage, respectively; residues of the metabolite CL 312094 up to 0.01 ppm in soybean forge and metabolite CL 325195 of 0.02 and 0.01 ppm in barley straw and grain. Residue data from a 60 day plant back interval for barley and soybeans were not generated.

Conclusion

The confined crop rotation study indicates that residues of the parent and or metabolites CL 312094 and CL 325195 at 0.01-0.02 ppm are possible in rotated crops with a 30 day plant back interval. However, since the study was conducted at approximately 2X the current proposed use rate of 1.05 lb ai/A/season with application made to bare ground (a worst case), at a plant back interval of 60 days or later all residue components in all rotated crops should be less than 0.01 ppm. Accordingly, at the current proposed use rate of 1.05 lb ai/A/season field rotational crop studies are not required provided a revised label specifying a 60 day plant back interval for all rotated food/feed crops is submitted.

Magnitude of the Residue in Animals

Poultry Feeding Study

A poultry feeding study and tolerances on poultry commodities are tentatively not required for the proposed use on cotton, pending HED Metabolism Committee review (see Secondary Residues in Meat, Milk Poultry and Eggs).

Ruminant Feeding Study

CL 303,630: Determination of CL 303,630 Residues in Milk and Edible Tissues From Dairy Cattle After Oral Administration of CL 303,630 for 28 Consecutive Days; MRID No. 434928-59.

This study was discussed briefly in PP#5G04507 (see 8/10/95 memo of G. Kramer). In brief, fourteen female non-pregnant Holstein dairy cows were divided into four treatment groups of four cows with the extra two cows placed in the high dosage group for residue sampling 14 days after treatment was stopped. The four treatment groups were dosed for 28 days at 0, 0.66, 2.19, or 6.81 mg per kg feed (i.e. ppm) on a dry matter basis of Pirate with capsules using a balling gun. Whole milk was collected twice daily and composited into a daily sample. Milk samples with an average fat content of 3.5% were stored in a refrigerator for up to three days, shipped on dry ice and frozen at Cyanamid freezer storage for analysis at Cyanamid or shipment by Federal Express to ABC Laboratories. Phenyl and pyrrole ^{14}C milk samples from the goat metabolism study were used to determine whether pirate partitions into milk fat. Radioanalysis of the fractions obtained after centrifugation showed that the average of the distribution was 42% of the total activity in milk fat, 9% in skim milk and 48% in the cell and casein fractions.

All cows except two in the high dosage group (i.e. sacrificed 14 days after the 28th dose) were sacrificed within 20 hours of the 28th consecutive treatment. Tissue samples were put on dry ice until placed into freezer storage. Milk samples were analyzed by method M-2395.01 and tissue samples by M-2398 (see Analytical Methods Section for a summary of the method. Milk and tissue samples were analyzed within 90 days of sampling. See Storage Stability Data section of this review for a discussion of storage stability data.

The following table from PP#5G04507 (see 8/10/95 memo of G. Kramer) summarizes the maximum residues found:

Table 7- Maximum residues in cow tissues following 28 days of administration of Alert at dietary burdens of 0.66, 2.19 and 6.81 ppm.

Tissue	Maximum Residues (ppm) at Dietary Burden of:		
	0.66 ppm	2.19 ppm	6.81 ppm
Milk	<0.010	0.035	0.042
Liver	<0.050	<0.050	0.054
Kidney	<0.050	<0.050	<0.050
Muscle	<0.010	0.017	0.022
Fat	0.067	0.429	0.597

Although residue levels in the two cows sacrificed 14 days after treatment at the high dose were significantly lower in the fat (i.e. 0.010-0.053 ppm), the residue levels in milk, muscle, liver and kidney were similar to the high dose residue levels. Residues levels in the milk, fat and muscle from high dose group were lower than would be expected from a linear extrapolation of residue data from the mid dose group. All the residue data from the ruminant feeding study are summarized in **Attachment 6.**

Conclusion

Clearly the highest Pirate residue levels from the ruminant feeding study occurred in fat tissue at approximately 25X residue levels in muscle tissue. Furthermore, the ^{14}C goat milk fat study verified that Pirate concentrates in milk fat as well.
The ruminant feeding study is tentatively acceptable pending HED Metabolism Committee review and adequate muscle and fat storage stability data as referenced under Conclusion 8. e.

Secondary Residues in Meat, Milk, Poultry and Eggs

Poultry

A poultry feeding study was not included with the current submission. Based on a poultry diet consisting of 20% cottonseed meal (at the 0.5 ppm proposed tolerance) and 80% grain (at 0 ppm), the maximum daily dietary intake of Pirate by poultry is estimated to be (.20 x 0.5 ppm or .10/89% DM) 0.11 ppm. The dietary burden for the two high dose groups in the seven day poultry metabolism study ranged from 14.42 to 16.09 ppm Pirate. The highest single residue from the poultry metabolism study was 0.70 ppm M-6A. This residue occurred in the 2-pyrrole poultry group dosed at 15.9 ppm which is equivalent to 145X (15.9/0.11) the maximum dietary burden from the proposed use on cotton.

Using the data from the seven day poultry metabolism study one would estimate the highest residue in poultry at 0.70/145 or 0.005 ppm which would likely not be detectable. Therefore, CBTS concludes that residues of Pirate are not likely to be found in poultry commodities, based on the feeding levels of the metabolism studies and the resulting residues. Accordingly, a poultry feeding study and tolerances on poultry commodities are tentatively not required for the proposed use on cotton, pending HED Metabolism Committee review.

Ruminant

A ruminant feeding study was submitted with this petition. Based on a diet consisting of 25% undelinted seed, 15% cottonseed meal, 20% cottonseed hulls and 20% cotton gin byproducts (each at the 0.5 ppm proposed tolerance for cottonseed) and 25% alfalfa forage (at 0 ppm), the maximum daily dietary intake of Pirate by beef cattle could be estimated to be 0.42 ppm (.75/.89 DM [average DM] x 0.5 ppm). However, this diet would be based on the assumption that Pirate residues in cotton gin byproducts would be similar to residue levels in cottonseed.

Alternatively, based on the same diet consisting of 25% undelinted seed, 15% cottonseed meal, 20% cottonseed hulls and 20% cotton gin byproducts (each at the 0.5 ppm **except cotton gin byproducts estimated at 10.0 ppm**) and 25% alfalfa forage (at 0 ppm), the maximum daily dietary intake of Pirate by beef cattle could be estimated to be 2.5 ppm (.60/.89 DM [average DM] x 0.5 ppm and .20/.89 DM x 10.0 ppm or 0.3 ppm plus 2.2 ppm). This diet would be based on the assumption that Pirate residues in cotton gin byproducts would be significantly higher than residue levels in cottonseed, and accordingly should represent a worst case.

Worst Case Dietary Burden: The maximum dietary burden associated with this proposed tolerance in dairy cows results from diet comprised of cottonseed, meal, hulls, and cotton gin byproducts:

Feed Item	% Diet	proposed Tolerance*	% DM	ppm in Diet
undelinted seed	25	0.5 ppm	88	0.14
meal	20	0.5	89	0.11
hulls	15 (20 for beef cattle)	0.5	90	0.08
cotton gin by products	20	10.0 ppm-worst case estimate	90	2.22
alfalfa hay	20	0	n/a	0
Total	100			**2.55**

*Covered by RAC tolerance

Cotton Gin By-Products - Basis for Estimate

CBTS notes that residue data are not currently available on Pirate in cotton gin byproducts. High residues of Pirate were reported on cotton foliage from the cotton metabolism study. For example 48.3, 102.9 and 131.8 ppm TRR were reported at day zero after an application rate of 2 lb ai/A phenyl C14 (i.e. 2X the current proposed rate) in cotton foliage following the first, third and fifth treatments, respectively. Approximately 59-68% of the TRR in **cottonseed** was identified as parent. Pirate field dissipation residue data have also been submitted to EFED (re: MRID No. 434928-14) which indicates Pirate residues on cotton leaves (upper canopy) after a single late season application of 0.4 lb ai/A (0.4X) were 6.9 and 3.3 ppm, at 14 and 28 days after application, respectively. Assuming a linear residue dissipation Pirate residues 21 days after application (the proposed PHI) would be 5.1 ppm at 0.4X. Accordingly, in the absence of field trial data a **worst** case estimate of 10.0 ppm Pirate in/on cotton gin by-products appears reasonable.

Milk Fat/Milk Concentration Factor - Basis for Estimate

^{14}C radioanalysis data of whole milk, milk fat and skim milk samples from the goat metabolism study was included with the ruminant feeding study (i.e. page 196). Residue data was provided on both phenyl and pyrrole labeled Pirate. The phenyl labeled Pirate samples represented the highest concentration factor in milk fat with the total DPM in whole milk, fat and skim of 42336, 20343, and 5254, respectively for milk with approximately 5% milk fat by weight (remaining DPM assumed to be contained in casein and cell fraction by difference). 42,336 DPM in whole milk x 5% milk fat = 2117 DPM in milk fat if no concentration; accordingly, 20343/2117=10X concentration in milk fat. Since based on proposed permanent tolerances (i.e. 0.15 and 0.01 ppm for milk fat and milk, respectively) the petitioner has proposed a 15X concentration factor, in the absence of additional data CBTS will utilize a 15X concentration factor for Pirate in milk fat.

Basis for Tolerances on Ruminant Commodities

Per Table II (September 1995) EPA will be flexible regarding whether studies already begun which do not generate data on all the commodities and feedstuffs are adequate for tolerance setting and registration purposes. Consequently, in the absence of Pirate residue data on cotton gin byproducts CBTS will not object to the establishment of <u>conditional</u> tolerances on ruminant commodities based on residue levels from the mid dose cattle feeding study reflecting a dietary burden of <u>2.19 ppm</u> Pirate. Although, the 2.19 ppm feeding level is slightly lower than the worst case dietary burden estimate above of 2.5 ppm the rounding up of required tolerances based on **actual residue data** from the feeding study makes this a practical approach. The following table summarizes the required tolerances.

RESIDUES OF PIRATE IN RUMINANT COMMODITIES

MATRIX	PROPOSED PERMANENT TOLERANCE (ppm)	MAX. RESIDUE IN MID DOSE CATTLE FEEDING STUDY (i.e. 2.19 ppm Pirate dosage group)	MAX. RESIDUE EXPECTED AFTER ADJUSTMENT TO REALISTIC WORST CASE EXPOSURE [required tolerance] (ppm)
MILK	0.01	0.035	0.05
MILK FAT	0.15	Residue data on milk fat not collected. 15X concentration factor proposed by petitioner.	0.75
MEAT*	0.01	0.017	0.02
MEAT BY-PRODUCTS*	0.10	<0.050	0.05 since is LOQ
FAT*	Not currently proposed	0.429	0.50

*=CATTLE, GOATS, HOGS, HORSES AND SHEEP.

Conclusion

Using the data from the seven day poultry metabolism study one would estimate the highest residue in poultry at 0.005 ppm which would likely not be detectable. Therefore, CBTS concludes that residues of Pirate are not likely to be found in poultry commodities, based on the feeding levels of the metabolism studies and the resulting residues. Therefore, a poultry feeding study and tolerances on poultry commodities are tentatively not required for the proposed use on cotton, pending HED Metabolism Committee review.

Based on a best estimate of possible residues in cotton gin byproducts tolerances for Pirate in ruminant commodities of 0.05, 0.75, 0.02, 0.50 and 0.05 ppm, respectively for milk, milk fat, meat, fat and meat byproducts of cattle, goats, hogs, horses, and sheep are required. A final determination of the secondary residues in meat, milk, poultry and eggs must be deferred pending HED Metabolism Committee review. If other deficiencies in this petition are resolved, the ruminant tolerances could be made conditional on the submission of the required residue data on cotton gin byproducts.

Revised Section F

The proposed permanent tolerances for Pirate in/on animal commodities from the proposed use on cotton should tentatively be revised as follows:

Milk	0.05
Milk Fat	0.75
Meat of Cattle, Goats, Hogs, Horses, and Sheep	0.02
Fat of Cattle, Goats, Hogs, Horses and Sheep	0.50
Meat Byproducts of Cattle, Goats, Hogs, Horses and Sheep	0.05

Other Considerations

An International Residue Limit Status sheet is included in this review as **Attachment 7.** Since no Codex, Canadian, or Mexican limits/tolerances have been established for Pirate, there are no compatibility problems at this time.

Attachment 1- Review of Product Chemistry; New Chemical - Pirate/Alert.

Attachment 2- Pirate Residues in Poultry Metabolism Study (from pages 6 and 9 of MRID No. 434928-54).

Attachment 3- Pirate Animal and Fish Metabolism (page 144 from Summary of Data Submitted in Support of the Registration Application for AC 303,630 Insecticide-Miticide Technical; American Cyananmid Company; 12/9/94; no MRID No.).

Attachment 4- Pirate Residues in Goat Metabolism Study (from pages 6 and 7 of MRID No. 434928-55).

Attachment 5 - Pirate Residues in Confined Crop Rotation Study (from page 8 of MRID No. 434928-51)

Attachment 6 - Pirate Residues in Dairy Cattle Feeding Study (from page 21 of MRID No. 434928-59).

Attachment 7 - International Residue Limit Status Sheet.

Attachment 8- Confidential Appendix - Pirate/Alert.

7509C:CBTS:CM#2:Rm 800:305-6991:G.Otakie:11/30/95
edit:GO: 12/1/95, 12/7/95/, 12/12/85.

CBI
cc with all attachments including **Attachment 8 (Confidential Appendix)**: PM 19-Dennis Edwards, Reviewer-Otakie, PP#5F04456.

NON-CBI
cc **without Attachment 8 (Confidential Appendix)**: RF, Circu, Karin Whitby (RCAB), E. Haeberer.

RDI: Ehaeberer:1/23/96 RLoranger:1/31/96

ATTACHMENTS - PIRATE/COTTON - PP#5F04456

ATTACHMENT 1 - PRODUCT CHEMISTRY

REVIEW OF PRODUCT CHEMISTRY (SUBDIVISION D), GLN'S 61 TO 63

New Chemical - Pirate/Alert®

Chemical Name (IUPAC, ANSI, etc.) -
Pirate/Alert [4-bromo-2-(4-chlorophenyl)-1-(ethoxymethyl)-5-(trifluoromethyl)-1H-pyrrole-3-carbonitrile]
IUPAC Name - [4-bromo-2-(4-chlorophenyl)-1-(ethoxymethyl)-5-(trifluoromethyl)pyrrole-3-carbonitrile]

Chemical Number (CAS; PC Code) - 122453-73-0		
Registration No. - T:241-no file symbol		
Test Substance - T		
Type of Product (T, FI, MP, EP) - T		
CB Nos. - 15094		
DP Barcode - D211889		
Reviewer - G. Otakie		
Approvals		
Branch Senior Scientist	R. Loranger	R. Loranger
Branch Chief	M. Metzger	F. Ives for

Table 1: Manufacturing and Impurity Data for Pirate (T).			
GLN	MRID	Status[1]	Deficiency
61-1: Product Identity & Disclosure of Ingredients	42770201 and 434928-01	A	The petitioner should submit verification of an ANSI approved common name if one has been obtained.
61-2: Starting Materials & Manufacturing Process	42770201 and 434928-01	A	The petitioner is reminded that any changes in the maufacturing process including the use of an alternate manufacturing process must be submitted to EPA.
61-3: Discussion of Impurities	42770201 and 434928-02	A	This requirement is satisfied for the current manufactiuring process.
62-1: Preliminary Analysis	434928-03	A	Preliminary analysis data from large scale and pilot scale production were provided.
62-2: Certification of Limits	434928-03 - CSF dated 10/17/94	N	A revised CSF is required with certified limits for all imupities $\geq$ 0.1 w/w %.
62-3: Analytical Methods	434928-03	A	Methods for impurities were also provided.
[1] A = Acceptable per current submission. N = Unacceptable (see Deficiency).			

Table 2: Physical and Chemical Properties for Pirate/Alert (T)			
GLN	MRID	Status[1]	Result[2] or Deficiency
63-2: Color	42770202	A	light tan or light yellow
63-3: Physical State	"	A	powdered solid
63-4: Odor	"	A	characteristic of halides and ketones
63-5: Melting Point	"	A	melting point apparatus 100-101° C
63-6: Boiling Point	"	A	n/a; TGAI is a solid
63-7: Density, Bulk Density, or Specific Gravity	"	A	0.543 g/ml tapped bulk density 0.355 g/ml untapped bulk density
63-8: Solubility	"	A	Solvent — Solubility at 25°C deionized water 0.12 mg/ml water, pH 4 0.13 mg/l water, pH 7 0.14 mg/l water, pH 10 0.12 mg/l hexane 0.89 g/100 ml methanol 7.09 g/100 ml acetonitrile 68.4 g/100 ml toluene 75.4 g/100 ml acetone 114 g/100 ml dichloromethane 141 g/100 ml
63-9: Vapor Pressure	"	A	<1.0 x 10-7 mm hg at 25 ° C
63-10: Dissociation Constant	"	A	since there are no ionizable groups in the Pirate structure, no dissociation will occur (PAI)
63-11: Octanol/Water Partition Coefficient	"	A	Kow = 67,670 (log Kow = 4.83) at 25 ° C
63-12: pH	"	A	7.16; 1% aqueous slurry at 24 ° C
63-13: Stability	" and 434928-05	A	stable at 25 ° C for 24 months, 37 ° C for 12 months, and 45 ° C for 3 months.
3-14: Oxidizing or Reducing Action	"	A	unreactive to oxidizing or reducing agents; no reaction was observed when wexposed to tap water, 1% monoammonium phosphate, 0.01M aqueous potassium permanganate and zinc foil.
63-15: Flammability		N/A	TGAI is a solid
63-16: Explodability	"	A	not sensitive to an impact of 2 kg/cm at room temperature; one exotherm at 183 ° C with a heat release of -350 kJ/kg in differential thermal analysis; dust did not ignite at any concentration or ignition delay time teste; classified as Class 0 dust (impact, differential thermal analysis, and dust explositivity assays)
63-17: Storage Stability	" and 434928-05	A	stable for one year under outdoor storage conditions (GC and HPLC assays).

63-18: Viscosity		N/A	TGAI is a solid
63-19: Miscibility		N/A	TGAI is a solid
63-20: Corrosion Characteristics	"and 434928-05	A	no corrosion observed after 12 months storage in a polyethylene bag or a VELOSTAT (non-conductive plastic) bag inside a fiberpak

[1] A = Acceptable (also see 1/26/94 review of PP#3G4224); N = Unacceptable (see Deficiency); N/A = Not applicable.
[2] For example, "light tan" for 63-2; "100-101° C" for 63-5.

Attachment 8: Confidential Appendix

CHLORFENAPYR

Page _____ is not included in this copy.

Pages 41 through 45 are not included in this copy.

The material not included contains the following type of information:

_____ Identity of product inert ingredients.

_____ Identity of product impurities.

_____ Description of the product manufacturing process.

_____ Description of quality control procedures.

_____ Identity of the source of product ingredients.

_____ Sales or other commercial/financial information.

_____ A draft product label.

_____ The product confidential statement of formula.

_____ Information about a pending registration action.

X FIFRA registration data.

_____ The document is a duplicate of page(s) _____.

_____ The document is not responsive to the request.

The information not included is generally considered confidential by product registrants. If you have any questions, please contact the individual who prepared the response to your request.

RESIDUES OF PIRATE IN ANIMAL COMMODITIES

MATRIX	PROPOSED PERMANENT TOLERANCE (ppm)	MAX. RESIDUE IN MID DOSE CATTLE FEEDING STUDY (i.e. 2.19 ppm Pirate dosage group)	MAX. RESIDUE EXPECTED AFTER ADJUSTMENT TO REALISTIC WORST CASE EXPOSURE (i.e. COLUMN 3/1.6 or 3/1) [required tolerance] (ppm)
MILK	0.01	0.035	0.05
MILK FAT	0.15	Residue data on milk fat not collected. 15X concentration factor proposed by petitioner.	0.75
MEAT*	0.01	0.017	0.02
MEAT BY-PRODUCTS*	0.10	<0.050	0.05 since is LOQ
FAT*	Not currently proposed	0.429	0.50

*=CATTLE, GOATS, HOGS, HORSES AND SHEEP.

ATTACHMENT L

INTERNATIONAL RESIDUE LIMIT STATUS

J. Keves 11/13/95

CHEMICAL ALEPT / PIRATE *

CODEX NO. ____

CODEX STATUS:

☑ No Codex Proposal Step 6 or above

Residue(if Step 8): ____

Crop(s)	Limit (mg/kg)

PROPOSED U.S. TOLERANCES:

Petition No. 5F04456

RCB Reviewer OTAKIE

Residue: PARENT, ONLY

Crop(s)	Limit (mg/kg)
COTTONSEED	0.5
MILK	0.01
MILK FAT	0.15
MEAT	0.01
MEAT BY-PRODUCTS	0.10

CANADIAN LIMITS:

☑ No Canadian limit

Residue: ____

Crop(s)	Limit (mg/kg)

MEXICAN LIMITS:

☑ No Mexican limit (on cottonseed)

Residue: parent (presumably)

Crop(s)	Limit (mg/kg)

47

NOTES:

* 4-bromo-2-(chlorophenyl)-1-(ethoxymethyl)-5-(trifluoromethyl)-1H-pyrrole-3-carbonitrile

Form revised 1986

CHLORFENAPYR

Page _____ is not included in this copy.

Pages 48 through 51 are not included in this copy.

The material not included contains the following type of information:

_____ Identity of product inert ingredients.

_____ Identity of product impurities.

_____ Description of the product manufacturing process.

_____ Description of quality control procedures.

_____ Identity of the source of product ingredients.

_____ Sales or other commercial/financial information.

_____ A draft product label.

X The product confidential statement of formula.

_____ Information about a pending registration action.

_____ FIFRA registration data.

_____ The document is a duplicate of page(s) _____.

_____ The document is not responsive to the request.

The information not included is generally considered confidential by product registrants. If you have any questions, please contact the individual who prepared the response to your request.