R7

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

MAR 25 1991

OFFICE OF
PESTICIDES AND TOXIC
SUBSTANCES

MEMORANDUM

SUBJECT: PP#3F2788 - Pendimethalin (Prowl®) on/in Barley and Wheat.
Review of the November 29, 1990 Amendment.
(MRID No. 417139-01) [DEB Nos. 7595 and 7596]
(HED Project No. 1-0507)

FROM: Francis D. Griffith, Jr., Chemist
Chemistry Branch I - Tolerance Support
Health Effects Division (H7509C)

TO: Robert J. Taylor, PM 25
Fungicide-Herbicide Branch
Registration Division (H7505C)

and

Toxicology Branch II - Herbicide, Fungicide, and Antimicrobial Support
Health Effects Division (H7509C)

THRU: Richard D. Schmitt, Ph.D., Chief
Chemistry Branch I - Tolerance Support
Health Effects Division (H7509C)

American Cyanamid Company has submitted this amendment consisting of a cover letter, a new Section B (new directions for use of Prowl® on wheat and barley), Supplementary Section D (new ruminant metabolism study), and a revised Section F (new tolerance proposal) in response to deficiencies outlined in our reviews of September 23, 1983 and March 9, 1983 by R.B. Perfetti. In the interim, the Pendimethalin Registration Standard, issued on May 10, 1984, and the Pendimethalin Registration Standard Updated, dated March 19, 1990, identified these and additional residue chemistry deficiencies. In the cover letter, the petitioner requests reactivation of the petition contending that all major toxicology deficiencies are resolved. The deficiencies identified in the Registration Standard are repeated below in the body of this review in the order they appeared in the Pendimethalin Registration Standard, followed by the petitioner's response and then CBTS comments. Our conclusions and recommendation follow.

EXECUTIVE SUMMARY OF CHEMISTRY DEFICIENCIES

- Plant metabolism studies required.
- Poultry metabolism studies are necessary.
- Ruminant metabolism study characterization of residues is required.
- Confirmatory analytical method needed.
- Multiresidue method data are needed.
- Livestock feeding studies may be required.
- Wheat processing study is necessary.

CONCLUSIONS

1. CBTS Conclusion on **Directions for Use**

The petitioner has proposed an adequate set of directions for use of Prowl® Herbicide (EPA Registration No. 241-243) on barley and wheat fields to control various grasses and broadleaf weeds.

2. CBTS Conclusion on **Nature of the Residue - Plants**

CBTS reiterates that the nature of the pendimethalin residue in plants is not adequately understood. Additional plant metabolism studies are required in which pendimethalin, radiolabeled in the phenyl ring, is applied to plants at rates equal to, or greater than, the maximum application rate. Higher application rates are preferred to increase the level of radioactive material available for analysis and identification, provided there is no phytotoxicity. One plant metabolism study should be conducted on sweet corn with analysis of vegetative parts and grain from 1) plants treated preemergence and 2) plants treated postemergence. A second plant metabolism study is needed on a plant in which the edible portion grows in the soil. The deficiency remains unresolved and continues outstanding.

3. CBTS Conclusions on **Nature of the Residue - Livestock**

a. CBTS reiterates that poultry metabolism studies are required using laying hens dosed with ring-labeled ^{14}C-pendimethalin for at least 3 days at a level > 1 part per million (ppm), preferably 10 ppm, sufficient to have adequate radiolabeled material available for identification. The laying hens are to be sacrificed within 24 hours of the final dose. The distribution and characterization of at least 90+ percent of the radiolabeled residue need to be determined in eggs

(white and yolk), muscle, skin, and liver. This part of the deficiency is not resolved and continues outstanding.

b. The petitioner has conducted and reported on a lactating caprine ^{13}C- and ^{14}C-ring-labeled pendimethalin metabolism study. Measurable/detectable residues were found only in caprine liver ranging from 0.08 ppm to 0.17 ppm (n = 3) from a 6.5 ppm dose. Trace amounts were detected in kidney (about 0.02 ppm) and in milk (< 0.01 ppm). Fractionation of various liver extracts revealed numerous free, unbound ^{14}C-components in the 0.005 ppm to 0.025 ppm range that were not characterized. CBTS concludes that the petitioner has not adequately identified the nature of the residue in ruminants. CBTS suggests that the petitioner repeat the HPLC identification steps for all fractions above 0.005 ppm, characterizing major peaks and using detectors that can elucidate organic structures such as but not limited to MS, FTIR, FTUV, and NMR. The petitioner needs to confirm the presence of or absence of all metabolites identified in the rat metabolism study. Complete characterization of caprine radiolabeled residues is essential for CB to ascertain the need for a ruminant feeding study. If caprine metabolism differs significantly from that in rats, then a ^{14}C-ring labeled pendimethalin porcine metabolism study may also be necessary. The deficiency is not resolved and continues outstanding.

4. CBTS Conclusions on **Residue Analytical Methods**

a. CBTS reiterates that a validated confirmatory method (Mass Spec is suggested) for residues of pendimethalin, per se, and its metabolite (CL-202,347) is necessary. This part of the deficiency is not resolved and continues outstanding.

b. CBTS reiterates that additional multiresidue method (MRM) validation data are necessary for the Food and Drug Administration's (FDA) MRM's A through E. Chromatographic data are required for pendimethalin and its alcohol metabolite for protocol C. Representative samples of plant and animal tissues need to be analyzed by appropriate MRM protocols B, D, and E following the FDA decision tree for MRM testing. The protocols are found in FDA's PAM-I, Appendix II. This part of the deficiency is not resolved and continues outstanding.

c. CBTS reiterates that if radiolabeled validation of existing analytical methods for plants and animals indicates that a major portion of the total radioactive residue is not recovered and identified by these methods, then radiolabeled validation of any new proposed residue analytical method may be required.

This part of the deficiency is not resolved and continues outstanding.

5. CBTS Conclusion on **Storage Stability Data**

CBTS reiterates that data are needed reflecting the stability of pendimethalin and its 3,5-dinitro-benzene alcohol metabolite (CL-202,347) in or on representative plants [such as root and tuber vegetables, legume vegetables, cereal grains, and miscellaneous crops (e.g., cottonseed, peanuts, and sunflower/safflower seed)] and animal matrices stored at freezing temperatures for time intervals approximating those of the treated samples used to determine the magnitude of the residue. The sample storage conditions and intervals need to be supplied for all previously submitted residue data for wheat and barley commodities (raw and processed foods and feeds). Storage stability data are required for only those samples deemed to be useful for tolerance assessment. The purity of the reference standards used for fortification of samples and a complete description of the analytical method(s), including extraction procedures, and any method validation data generated need to be provided. The deficiency is not resolved and continues outstanding.

6. CBTS Conclusion on **Magnitude of the Residue - Crop Field Trials**

From 16 barley and wheat crop field trials no detectable residues (<0.05 ppm) were found in any barley or wheat grain samples. Likewise, no pendimethalin residues were detected (<0.05 ppm) in barley and wheat forage. Residues of parent only pendimethalin were detected in 5 barley and wheat straw samples ranging from 0.05 ppm to 0.22 ppm. It appears that residues of pendimethalin plus its alcohol metabolite will not exceed the proposed tolerances on barley and wheat grain, forage, and straw under the proposed conditions for use of Prowl® (see conclusion on Proposed Tolerances).

7. CBTS Conclusions on **Magnitude of the Residue - Meat/Milk/Poultry/Eggs**

a. CBTS reiterates that a conventional ruminant feeding study may be necessary, depending on the results of the ruminant metabolism study, in which lactating ruminants are dosed at 0.1, 0.3, and 1.0 ppm pendimethalin, *per se*, ≥ three animals per dose group in the total diet. The animals should be kept on the treated feed for 4 weeks. However if residues have not plateaued in milk by the end of 4 weeks, then the feeding period should continue until a plateau is reached. Milk should be collected twice daily and residues determined therein. Animals must be sacrificed within 24 hours of the final dose and residues determined in muscle, liver, kidney,

and fat. The deficiency is not resolved and continues outstanding.

b. CBTS reiterates that a conventional poultry feeding study may be necessary, depending on the results of the poultry metabolism study, in which laying hens are dosed at 0.1, 0.3, and 1.0 ppm pendimethalin, per se, in the total diet, ≥ 10 hens per dose group. The laying hens should be kept on the treated feed for 4 weeks. However, if residues have not plateaued in eggs by the end of 4 weeks, then the feeding period should continue until a plateau is reached. Eggs should be collected at least daily and residues determined therein. The laying hens need to be sacrificed within 24 hours of the final dose and residues determined in muscle, liver, skin, and fat. The deficiency is not resolved and continues outstanding.

c. CBTS reiterates that the nature of the pendimethalin residues in livestock is not adequately understood. If the feeding studies are necessary at this time, CBTS reiterates that we will request residue data for residues of pendimethalin, per se, and its metabolite CL-202,347 in the conventional feeding studies. CBTS points out that residue data may need to be presented for any additional metabolite(s) of toxicological concern if the requested metabolism studies so identify.

d. CBTS reiterates that the present ruminant metabolism study indicates that residues of pendimethalin may occur in meat and meat by-products. The petitioner should be advised that tolerances need to be proposed for these animal commodities if the requested pendimethalin metabolism and feeding studies indicate the transfer of residues. The deficiency is not resolved and continues outstanding.

8. **CBTS Conclusion on Magnitude of the Residue - Processed Food/Feed**

Upon further consideration, CBTS now concludes that the petitioner needs to conduct a pendimethalin wheat processing study using wheat bearing detectable residues, or, if no residues are detected, then use wheat treated at the highest practical application rate. The wheat is to be processed by standard commercial operations into wheat bran, flour, middlings, and shorts. Pendimethalin and its metabolite residue data are needed for each of these processed wheat commodities. If pendimethalin residues concentrate, then appropriate food and/or feed additive tolerance need to be proposed.

9. CBTS Conclusion on **Proposed Tolerances**

The petitioner's revised tolerances proposed for 0.1 ppm on barley and wheat grain and forage, and 0.3 ppm for barley and wheat straw appear to be adequate. However, judgment on these proposed tolerances is deferred until the nature of the residue is adequately understood.

RECOMMENDATION

CBTS cannot, at this time, recommend for the requested pendimethalin tolerance of 0.1 ppm on wheat grain, wheat forage, barley grain, and barley forage; and the 0.3 ppm tolerance on wheat and barley straw for the reasons cited in our Executive Summary of Deficiencies and detailed in our Conclusions 2 through 8 above.

For further consideration of this petition, the petitioner needs to be advised to resolve these deficiencies.

DETAILED CONSIDERATIONS

DIRECTION FOR USE

Petitioner's Response

The petitioner has presented a revised label for use of Prowl® Herbicide (EPA Registration No. 241-243) containing 4 lbs active ingredient/gallon, 42.3 percent of technical pendimethalin to control various grasses (e.g., barnyard grass, crabgrass, foxtail, Johnsongrass) and broadleaf weeds (e.g., carpetweed, lambsquarters, pigweed) in wheat and barley fields.

CBTS Comments

The petitioner proposes applying Prowl® Herbicide to barley or wheat fields by ground equipment in at least 10 gallons of water per acre or from aircraft in at least 5 gallons of water per acre. Prowl can be applied to barley or wheat fields as a preplant application up to 3 weeks prior to planting with mechanical incorporation to 1 to 1 1/2 inches. Prowl may also be used after barley and wheat seeding as a preemergence surface application with or without incorporation, and Prowl can be used as an early postemergence application when the barley and wheat are past the two to three leaf stage. The petitioner cautions that emerged weeds are not controlled by postemergence applications.

The rate of Prowl® application ranges from 1.5 pints (0.75 lb active ingredient [ai] pendimethalin) per acre in coarse sandy soils to 3 pints (1.5 lbs ai) per acre in fine clay soils. The preharvest interval (PHI) for wheat and barley is 120 days. The

petitioner cautions that Prowl is not to be used on peat or muck soils.

The petitioner has proposed an adequate set of directions for use of Prowl Herbicide on barley and wheat fields.

NATURE OF THE RESIDUE - PLANTS

Deficiencies

The following additional data are required:

1. Data involving the reasonably complete characterization of the extractable and unextractable radioactive residues found in plant tissues as the result of the application of radiolabeled pendimethalin in a manner simulating a treatment regime registered for use. Representative crops (potatoes, soybeans, corn, etc.) for which pendimethalin formations are registered should be used.

2. Data depicting the distribution and metabolism of [^{14}C]pendimethalin in or on mature plant parts from three dissimilar food crops (e.g., a root crop, oilseed crop, and a leafy vegetable). If metabolism is not similar in the three crops, additional studies using other crops may be required. A completely characterized test substance representative of technical pendimethalin (including impurities, if appropriate) used in commercial formulations must be applied at levels sufficiently high to permit characterization of ^{14}C-residues.

The identities and quantities of extractable and nonextractable residues must be determined. Confirmation of the identities of residues using a suitable confirmatory method such as MS or HPLC is also required. In addition, representative samples from the tests must be analyzed using a currently accepted or proposed enforcement analytical method in order to ascertain that this method will determine all possible metabolites of concern.

Petitioner's Response

The petitioner did not respond.

CBTS Comments

After a number of consultations with the petitioner, CBTS reiterates that the registrant should be informed that the plant metabolism data base for pendimethalin is not adequate. Although the available studies indicate that low levels of radioactivity are taken up from the soil into aerial parts of plants, these studies were conducted with pendimethalin radiolabeled in side chains as opposed to in the phenyl ring. In addition, most of

the studies were conducted using application rates lower than the maximum permitted on product labels.

Additional plant metabolism studies are required in which pendimethalin, radiolabeled in the phenyl ring, is applied to plants at rates equal to at least the maximum rates on product labels. Provided significant phytotoxicity does not occur, even higher application rates (2X-5X) are preferred to increase the level of radioactivity available for analysis and identification. One study should be conducted on sweet corn with analysis of vegetative parts and grain from 1) plants treated preemergence and 2) plants treated postemergence. A second plant metabolism study is needed on a plant in which the edible portion grows in the soil (e.g., potatoes or peanuts). The petitioner's report should include the percentage of the total radioactive residue (TRR) for each plant part and the report should include the ppm value for the TRR as well as each identified component of the TRR. The petitioner is expected to identify at least 90+ percent of the TRR. The petitioner is to confirm identities of all metabolites by a second technique.

Chemistry Branch I recently concluded that deficiencies in the knowledge of plant metabolism were not applicable for use on sugarcane (PP#2F2765, R. Cook, 11/26/90). CBTS emphasizes that this decision applies only to sugarcane and is based on the low total activity (<0.01 ppm) and long pre-harvest interval observed in that crop.

CBTS reiterates that the nature of the residue in plants is not adequately understood. This deficiency continues unresolved and remains outstanding.

NATURE OF RESIDUE - LIVESTOCK

Deficiencies

The following data are required:

1. Metabolism studies utilizing ruminants. Animals must be dosed with ring-labeled [^{14}C]pendimethalin for 3 days at a level (> 1.5 ppm) sufficient to make residue identification possible. Animals must be sacrificed within 24 hours of the final dose. The distribution and characterization of residues must be determined in milk, muscle, fat, kidney, and liver. If ruminant metabolism is found to differ significantly from that in rats, then swine metabolism data will also be required.

2. Metabolism studies utilizing poultry. Hens must be dosed with ring-labeled [^{14}C]pendimethalin for 3 days at a level (> 1 ppm) sufficient to effect residue identification. Birds must be sacrificed within 24 hours of the final dose. Residues must be characterized and quantified in eggs, muscle, fat, kidney, and liver.

3. Metabolism studies utilizing ruminants and poultry. Animals must be dosed orally with ring-labeled [^{14}C]pendimethalin for a minimum of 3 days at a level sufficient to make residue identification and quantification possible. Eggs and milk must be collected twice daily during the dosing period. Animals must be sacrificed within 24 hours of the final dose. The distribution and identity of residues must be determined in eggs, milk, muscle, fat, kidney (except poultry), liver, and poultry skin. Representative samples from both of the studies must be analyzed using a suitable confirmatory method such as MS or HPLC. In addition, representative samples from these studies must be analyzed using a currently accepted or proposed enforcement analytical method in order to ascertain that the method is capable of adequately recovering and identifying all residues of concern. If the ruminant and/or poultry metabolism differs significantly from the rat data, then swine metabolism data will also be required.

Petitioner's Response (See MRID No. 417139-01)

The petitioner has submitted a caprine metabolism study titled "Pendimethalin (AC 92.553): Disposition of Carbon-14 Labeled (AC 92.553) in Lactating Goats and Characterization of the Residue in Goat Liver" by J. Zulalian, T.M. Lee, and P. Miller, dated November 20, 1990 and coded Report Number CY 37.

CBTS Comments

The petitioner has not presented a poultry metabolism study. CBTS reiterates that the nature of the residue in poultry is not adequately understood and that a poultry metabolism study using ring-labeled ^{14}C-pendimethalin is necessary. The petitioner is reminded to identify at least 90+ percent of the TRR in any new poultry metabolism study. Current Branch policy, as outlined and explained in Attachment 3 to the "Overview of Residue Chemistry Guidelines," clearly states that CBTS now requires poultry metabolism studies whenever a pesticide is to be applied to a crop having a poultry feed commodity listed in Table II of the Residue Chemistry Guidelines. Wheat and/or barley can be 50 percent of poultry diets. Thus a poultry metabolism study is necessary. This part of the deficiency is not resolved and continues outstanding.

The petitioner presented the results of a two-part ruminant metabolism study. The first part of the study was designed to determine the disposition of ^{14}C-pendimethalin in various caprine tissue, and the second part of the study was an attempt to characterize the ^{14}C-residues in caprine liver from an intermediate dose of ^{14}C-pendimethalin.

In the disposition part of the pendimethalin caprine metabolism study, the petitioner purchased three female Nubian goats in Easton, Pennsylvania. Two goats were 3 years old, and

one goat was 4 years old. All three goats were in the early stages of lactation. These goats were acclimated for 9 days at American Cyanamid's Agricultural Research facilities in Princeton, New Jersey, prior to starting the tests. Each goat was housed in a separate stainless steel metabolism cage. The actual dosing period ran from February 2, 1988 to February 9, 1988. Each goat was fed 1.0 kg of Purina Goat Chow and 1.9 kg of alfalfa hay per day. Water was provided ad libitum. The petitioner presented no evidence to show these feeds were free of potentially interfering heavy metals, alfatoxins, or other pesticides. CBTS does not consider that this is a problem as health observations of each goat were recorded daily and all three goats showed excellent health during the study and no abnormalities were detected at autopsy. The control goat, No. 1, weighed 46.3 kg at start of dosing and 47.3 kg at end of the test. The goat dosed at 6.09 kg/day or 2.1 ppm feeding level weighed 52.0 kg at the start and 50.1 kg at the end of the study. The third goat, dosed at 18.27 mg/day or 6.3 ppm in the feed, weighed 57.0 kg at the start of the study and 53.1 kg at the end of the study.

The goats were dosed with ^{14}C-pendimethalin uniform ring-labeled of specific activity 1.7 mCi/m mol diluted with cold pendimethalin to specific activity of 1.0 μCi/mg. Two separate sets of doses were prepared. One dose was 6.09 mg ^{14}C-pendimethalin and the other was 18.3 mg. A 0.3 mL of aliquot was added to 1.88 grams of lactose in a size 12 gel cap. The ethanol was allowed to evaporate overnight, then the caps were sealed. Sufficient caps were prepared for 7 days of dosing with two caps being retained for storage stability analysis. The two goats were dosed daily for 7 days, orally with a balling gun immediately after the afternoon milking. The control goat was dosed with an ethanol-evaporated, lactose-filled size 12 gel cap using a balling gun.

Milk was collected twice daily; the volume was recorded, pooled, and refrigerated. Total collection of all urine and feces was made for each goat. A 10 ml blood sample was taken each day prior to treatment. Although the analysis of blood helps determine the distribution of ^{14}C-pendimethalin, the results are not germane to CBTS's conclusion on caprine metabolism.

All three goats were sacrificed 20 hours after the seventh and last dose. Leg and tenderloin muscle, omental fat, kidneys, and liver were collected, placed in plastic bags, identified, and frozen.

Tissues samples were ground with dry ice. Five grams of tissue (muscles, liver, and kidney) were mixed with an equal volume of water and homogenized using a polytron homogenizer. One gram of homogenate (0.5 g tissue) was weighed into a combusto-pack, then inserted into combusto-cones with 9 mL Carbosorb II and 1.2 mL of Permafluor V and combusted in a Tri-Carb Model 306 oxidizer.

2.5 mL of milk was mixed with 1.5 mL of water and 10 mL of Aquasol-2. 0.5 gram of fat was weighed directly into the combustion cones.

Tissues from the control goat were fortified with ring-labeled ^{14}C-pendimethalin at a level of 0.05 ppm. Control milk was fortified at 0.01 ppm ^{14}C-ring-labeled pendimethalin. Recoveries of ^{14}C-material in tissues ranged from 73 to 91 percent and in milk ranged from 79 to 81 percent.

All of the samples were combusted in a Tri-Carb Model 306 oxidizer. The radioactive memory, spillover, and recovery efficiency were checked with a radioactive calibration source. For these samples, the efficiencies were $\geq$ 94 percent with < 1 percent radioactive memory. Samples were counted on a Beckman LS-5801 or LS-9800 - LSS (Liquid Scintillation System). Samples were counted until 40,000 counts < 1 percent error at 95 percent confidence level or for 10 minutes, whichever came first.

No measurable/detectable radioactivity was detected in any milk samples from either dose at the 0.01 ppm level of detection. Of all the tissues, only liver had a measurable/ detectable level of radioactivity of 0.05 ppm from the 2.1 ppm dose and 0.17 ppm from the high dose of 6.3 ppm. Review of the raw data (dpm) indicated a trace level of pendimethalin (around 0.02 ppm) could be present in the goat kidney from the 6.3 ppm dose. Also in milk samples at days 6 and 7 there were indications of ^{14}C-pendimethalin equivalents being present between 0.006 ppm and 0.009 ppm.

In the characterization part of the pendimethalin caprine metabolism study, the petitioner followed the procedures used in the disposition phase. Three- to five- year-old lactating goats were purchased from the same source. These goats were acclimated for 7 days at the same test facilities before the dosing period of April 20-27, 1989. Each goat was fed a control diet of 1.3 kg of Purina Goat Chow (G) and 1.6 kg of alfalfa per day. The control goat weighed 48 kg at the start of dosing and at the end of dosing. For the two test goats receiving 18.3 mg/day or 6.5 ppm in the feed, goat No. 2 weighed 65 kg at the start and 67 kg at the end of dosing; goat No. 3 weighed 52 kg at the start and 50 kg at the end of the study.

Both goats were dosed at the same level of 18.3 mg (total) per capsule/day for 7 days. The dose was a mixture of ^{14}C-ring-labeled pendimethalin (99.6% radio purity via two-dimensional TLC and 94.0% chemical purity via GC) and ^{13}C-3 aromatic methyl group pendimethalin to give a final specific activity of 10.35 uCi/mg. The radiolabeled pendimethalin (^{13}C & ^{14}C) were dissolved in acetone, then individual doses were prepared by addition to 1.5 grams lactose in a size 12 gel cap. The acetone was allowed to evaporate overnight before the caps were sealed. Sufficient capsules were prepared for 7 days of dosing with two caps being retained for storage stability analysis. The two test goats were

dosed orally daily for 7 days with a balling gun immediately after the afternoon milking. The control goat was dosed with an acetone- evaporated lactose-filled size 12 gelatin cap at the same time the test goats were dosed.

Milk, urine, and total fecal samples from all three goats were collected as described in the disposition part of the study. No fractionation or characterization of ^{14}C-pendimethalin was attempted on these samples. All three goats were sacrificed 20 hours after the last dose with only the liver being removed for analysis. The liver samples were placed in individual plastic bags, identified, then frozen until analysis.

Liver samples were ground with dry ice as described in the disposition part of the study. Radio analysis was as described above. The results of the combustion followed by LSC were 0.077 ppm in one liver and 0.096 ppm in the other liver.

200 grams of goat No. 2 liver containing 0.077 ppm were refluxed in 1 liter of CH_2OH/H_2O (4:1) at 60 °C for 2 hours. The mixture was filtered through Whatman #1 filter paper. The aqueous extract (0.034 ppm) was partitioned with hexane then CH_2Cl_2. The hexane extract and CH_2Cl_2 extract were combined (0.025 ppm) for normal phase HPLC analysis. This is the free, unbound organosoluble fraction. The aqueous phase (0.009 ppm) was separated by reverse phase HPLC into a number of unidentified components.

The filter cake (or Marc 1) was refluxed again for 2 hours at 60 °C in a liter of CH_3 OH/H_2O (4:1) containing 1 percent HCl to free the bound residues. This extraction mixture (0.018 ppm) was also filtered and partitioned with hexane and then CH_2Cl_2. The extracts combined (0.013 ppm) for normal phase HPLC separation. The aqueous phase was centrifuged and counted with 0.003 ppm remaining in the aqueous phase and 0.002 ppm being in the precipitate.

The filter cake (or Marc 2) was divided into two equal portions then mixed with 300 mL 0.1N HCl and 5 grams pepsin, sealed with a plug of cheesecloth, and incubated overnight at 37 °C in a shaker water bath. The mixture was treated with a 1:1 mix of trichloroacetic acid (TCA):water. After 1 hour, the mixture was filtered through Celite. The TCA- insoluble precipitate was water washed. This bound, non-extractable fraction contained 0.017 ppm. The TCA-soluble filtrate was extracted, 3 x 50 mL ether then 3 x 50 mL hexane. No further residue was enzymatically released, that is, organosoluble as < 0.001 ppm was detected. The aqueous phase and the centrifuged pellet contained 0.009 ppm of bound, unextractable residues.

The HPLC was performed on an IBM LC model 9533 Ternary system. The reverse phase column was a Supelco RP C-18 (5 micron, 25 cm x 4.6 m id) connected to a Supelco guard column, 2 cm cartridge, LC-18. The reverse mobile phase was H_2O:CH_3OH:0.2 percent formic acid for gradient analysis. The normal phase

column is a Whatman Partisil 5, 25 cm x 9.6 mm id, connected to a Brownlee guard column (Silica Newguard, 7 micron, 15 x 3.2 mm). The normal mobile phase is 2-propanol:CH_3OH:hexane for gradient analysis. The petitioner presented copies of HPLC chromatograms showing elution of standards of pendimethalin and pendimethalin alcohol thorough normal phase and reverse phase column. The petitioner presented copies of three "chromatograms" showing radio counts of 1 mL/min fraction collections off of both HPLC column. No further identification was attempted. Correlation of elution standards to the peaks in the chromatograms was not possible. CBTS notes there are numerous radio peaks in the three chromatograms. Although the petitioner has fractionated the residue of 0.01 to 0.08 ppm as directed by Attachment 3 to the Overview of the Residue Chemistry Guidelines, CBTS concludes that this is not adequate. The petitioner should have used additional detection systems for HPLC that can elucidate organic structure. CBTS suggests that the petitioner repeat the identification HPLC steps for all fractions above 0.005 ppm and identify all major peaks using detectors such as but not limited to MS, FTIR, FTUV, and NMR. The petitioner needs to confirm the presence of or absence of all compounds identified in the rat metabolism study. The petitioner has not adequately characterized the ^{14}C-pendimethalin residues in caprine liver. This information is required for CBTS to ascertain whether or not a ruminant feeding study is necessary. If the characterization of residues from the ruminant metabolism study differs significantly from the rat metabolism study, then a ^{14}C-ring labeled pendimethalin porcine metabolism study maybe necessary.

In summary from a caprine metabolism study where lactating goats were dosed with ^{14}C-ring-labeled pendimethalin and ^{13}C-3 aromatic methyl group pendimethalin, measurable/detectable residues were found only in the caprine liver at 0.05 ppm from a 2.1 ppm dose and at 0.08 to 0.17 ppm (n = 3) from a 6.5 ppm dose. A trace amount of pendimethalin was detected in caprine kidney (about 0.02 ppm) and in caprine milk at a level less than 0.01 ppm from the 6.5 ppm dose. Fractionation of liver reveals a number of free unbound ^{14}C-components in the 0.005 to 0.025 ppm range which have not been characterized.

The petitioner has not adequately identified the nature of the residue in ruminants. The determination step needs to be repeated for HPLC using detectors that can elucidate organic structure. The deficiency is not resolved and continues outstanding.

RESIDUE ANALYTICAL METHOD

Deficiencies

The following additional method is required:

1. A validated confirmatory method (MS is recommended) for residues of pendimethalin per se and its metabolite (CL-202, 347).

2. Representative samples of plant and animal tissues containing residues of pendimethalin and its 3,5-dinitrobenzyl alcohol metabolite must be analyzed by multiresidue Protocols C and E from PAM-I, Appendix II.
3. If radiolabeled validation of existing analytical methodology for plants and animals (refer to "Qualitative Nature of the Residue in Plants" and "Qualitative Nature of the Residue in Animals" for additional details) indicates a major portion of the total radioactive residue is not recovered and identified by these methods, radiolabeled validation of new proposed analytical methodology will be required.

Petitioner's Response

The petitioner did not respond.

CBTS Comments

CBTS reiterates the above deficiencies. They continue unresolved and remain outstanding.

After reconsideration on the requirements for MRM testing CB now concludes that additional MRM validation data are necessary for FDA MRM's A thru E. Chromatographic data are required for pendimethalin and its alcohol metabolite for Protocol C. Representative samples of plant and animal tissues need to be analyzed by appropriate MRM Protocols B,D, and E following FDA's decision tree for MRM testing. These Protocols are from in FDA's PAM-I, Appendix II. This part of the deficiency is not resolved and continues outstanding.

STORAGE STABILITY DATA

Deficiencies

The following additional data are required:

1. Data reflecting the stability of pendimethalin and its 3,5-dinitrobenzyl alcohol metabolite (CL-202,347) in or on representative plants [such as root and tuber vegetables, legume vegetables, cereal grains, and miscellaneous crops (e.g., cottonseed, peanuts, and sunflower seed)] and animal samples stored at freezing temperatures for time intervals approximating those of the treated samples used to determine the magnitude of the residue.

2. The sample storage conditions and intervals must be supplied for all required and previously submitted residue data for plant commodities (raw and processed foods and

feeds). Storage stability data in support of previously submitted residue data are required for only those samples deemed to be useful for tolerance assessment. The purity of the reference standards used for fortification of samples and a complete description of the analytical methods (including extraction procedures) and method validation data, used to supply the data in MRID No. 405351-01, must be provided. For additional guidance on conducting storage stability studies, the registrant is referred to an August 1987 Position Document on the Effects of Storage Validity of Pesticide Residue Data available from NTIS under Order No. PB 88112362/AS.

Petitioner's Response

The petitioner did not respond.

CBTS Comments

CBTS reiterates the above deficiencies. They continue unresolved and remain outstanding.

MAGNITUDE OF THE RESIDUE - CROP FIELD TRIALS

The petitioner has previously submitted pendimethalin residue data on barley (5) and wheat (11) crop field trails for crop years prior to 1984 from Oregon (3), Washington (2), Minnesota (1), Montana (1), California (3), Arizona (1), North Dakota (1), and Canada (4). In 9 trials Prowl® was applied one time at 0.75 lb a.i. (0.5X) to 2 lbs a.i. (1.3X) pre- or post-emergence. In the last 2 trials reviewed by the Agency, Prowl® was applied pre-emergence at 1.0 to 2.0 lbs a.i. per acre followed by another early post-emergence application at 1.0 lb a.i. per acre plus either Gleam®, or dinoseb.

No detectable residues (<0.05ppm) of pendimethalin plus its alcohol metabolite were found in any barley or wheat grain sample. From 9 trials residues of pendimethalin plus its metabolite in wheat and barley forage were < 0.05 ppm after PHI's of 40 to 91 days. In 2 other post-emergence application trials residues of pendimethalin, per se, on wheat forage were 66.7 ppm 2 hours after application and were 0.91 ppm 30 hours after application. Pendimethalin residues in wheat and barley straw from pre-emergence application (as opposed to pre-plant application) ranged from <0.05 ppm to 0.22 ppm with 5 samples having residues above 0.05 ppm.

The proposed tolerances appear to be appropriate. Residues of pendimethalin plus its metabolite are not expected to exceed the proposed tolerances on barley and wheat grain, forage, and straw when Prowl® is used as proposed (see discussion under Proposed Tolerances).

MAGNITUDE OF THE RESIDUE - MEAT/MILK/POULTRY/EGGS

Deficiencies

The following data are required:

1. Lactating ruminants must be dosed with 0.1, 0.3, and 1.0 ppm pendimethalin *per se* (≥ three animals/dose group) in the total diet until residues plateau in milk or for 28 consecutive days if no residues are detected in milk. Milk samples must be obtained twice daily. Animals must be sacrificed within 24 hours of the final dose and residues in tissues (muscle, liver, kidney, and fat) determined.

2. Poultry must be dosed with 0.1, 0.3, and 1.0 ppm pendimethalin *per se* (≥ ten hens/dose group) in the total diet. Egg samples should be collected twice daily and analyzed for residues; dosing should continue until residues in eggs plateau or for 28 days if residues are nondetectable. Hens should be sacrificed within 24 hours of the final dose and residues determined in muscle, fat, kidney, liver, and other edible tissues.

3. Since the residues of concern in animal products have not been delineated, at the present time we require data reflecting residues of pendimethalin *per se* and its metabolite CL-202,347. Other residues may need to be sought if requested metabolism studies so indicate.

4. The available goat metabolism study (see Nature of the Residue in Animals) indicates that residues of pendimethalin may occur in meat and meat byproducts of food animals. Tolerances must be proposed for these food commodities if the above-required data so indicate.

Petitioner's Response

The petitioner did not respond.

CBTS Comments

After reconsideration CBTS reiterates the above deficiencies with modifications. They continue unresolved and remain outstanding.

CBTS points out that in the "Overview of the Residue Chemistry Guidelines," current Branch policy is that animals should be kept on the treated feed for 4 weeks. However, if residues have not plateaued in eggs or milk by the end of 4 weeks, then the feeding period should continue until a plateau is reached.

MAGNITUDE OF THE RESIDUE - PROCESSED FOOD/FEED

CBTS Comments

In our previous review by R.B. Perfetti on March 9, 1983, CBTS (aka RCB) concluded that "since no detectable residues are observed in grain, no milling studies or food-additive tolerance proposals for milling fractions are needed." Upon further consideration, CBTS now concludes that a pendimethalin wheat processing study is necessary. This study should be conducted to address considerations noted below.

Based on the requirements as stated in the Overview of the Residue Chemistry Guidelines, the petitioner needs to conduct additional wheat crop field trials treated with pendimethalin at the proposed use rate and/or highest practical application rate. If detectable residues are found in the raw agricultural commodity (rac) wheat, then a processing study is necessary; and if the data show a concentration of residues, then a Food/Feed Additive Tolerance (FAT) is required. Residue data are necessary for wheat bran, flour, shorts, and middlings.

If "exaggerated rate" data are available and there are detectable residues, then these samples should be used for a processing study. If residues concentrate on processing, then the concentration factor should be applied to the rac tolerance to arrive at a FAT.

If pendimethalin exaggerated rate data are available and there are no detectable residues in the rac wheat, then no FAT is required provided that:

1. the application rate is exaggerated by at least the theoretical concentration factor,

2. the crop field trial data are sufficiently representative of major wheat growing regions so that any reasonable potential for detectable residues has been realized, and

3. the exaggerated rate was not unrealistically high.

The level of exaggerated application acceptable will depend on the use.

If application of the highest practical exaggerated pendimethalin rate results in no detectable residues and the level of exaggeration is less than the theoretical concentration factor, then the wheat is to be processed. If no detectable residues are found in the processed wheat bran, flour, shorts, and middlings, then no FAT is required. If any of the processed commodities contain any pendimethalin and its metabolite residues, then a FAT is required. In cases where the raw wheat contains no detectable pendimethalin residue, the processing study will indicate only that the minimum concentration factor is the ratio of the concentration in the processed commodity to the

limit of detection (not quantification) in the rac. CBTS will evaluate all available data in determining what is the appropriate concentration factor. This will include, at a minimum, the metabolism studies and chromatographic support data for the rac. In some cases it may be possible to estimate residue levels from chromatograms where the response is below the limit of reliable quantitation but nonetheless indicative of a "true" residue.

PROPOSED TOLERANCE

Deficiencies

3a. The proposed 0.1 ppm tolerance level for wheat and barley grain is appropriate. Also, since no detectable residues of pendimethalin were observed in grain, no milling studies or food additive tolerance proposals are required. Any future uses of pendimethalin that result in detectable residues in grain may engender the need for both milling studies and tolerance proposals on milling fractions.

3b. Based on the limited data available, the 0.1 ppm tolerance proposed for wheat and barley straw and forage is not adequate. A more appropriate level would be 0.3 ppm. If the higher level in wheat and barley forage is not acceptable, a revised Section B incorporating a restriction prohibiting the feeding of forage to livestock could be submitted. Such a restriction for straw would not be practical, however, and therefore a revised Section F proposing a higher level in wheat and barley straw (and possibly forage) is needed.

Petitioner's Response

The petitioner has submitted the following revised Section F.

It is hereby proposed that 40 CFR 180.361 be amended by adding the following:

180.361: tolerances are established for combined residues of pendimethalin [N-([1-ethylpropyl)-3,4-dimethyl-2,6-dinitrobenzenamine] and its metabolite [4-([1-ethyl propyl]amino)-2-methyl-3,5-dinitrobenzyl alcohol] in or on wheat grain and forage and barley grain and forage at 0.1 part per million (ppm), and wheat straw and barley straw at 0.3 ppm.

CBTS Comments

The petitioner's proposed 0.1 ppm pendimethalin and its metabolite tolerance for wheat and barley grain and forage appears to be adequate. Likewise the petitioner has now proposed the suggested 0.3 ppm tolerance for barley and wheat straw. CBTS feels that it is prudent to defer judgment on these tolerances until the nature of the residue is understood.

cc: R.F.,Circ (7),Reviewer(FDG); PP#3F2788, 3F2844, and 3F3049, PIB/FOD (Furlow), R.D. Schmitt, Ph.D., Chief.

H-7509C:CBTS:Reviewer(FDG):CM#2:Rm814B:557-0826:JOB: 62831:I:WP5.0:C.Disk:KEVRIC:02/19/91:aw:wo:aw:ed:fdg:2/25/91.

RDI:SecHd:RSQuick:2/28/91:BrSrSci:RALoranger:2/28/91.