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OFFICE OF
PREVENTION, PESTICIDES
AND TOXIC SUBSTANCES

MEMORANDUM

Date: 30 January 2007

Subject: Penoxsulam. Section 3 Registration Application for Use of GF-443 SC in Aquatic Sites and Request for a Tolerance Exemption (PP#5F7012) on Fish and Shellfish. Summary of Analytical Chemistry and Residue Data.

DP Barcode:	D326985	Decision Number:	362871
PC Code:	119031	MRID Nos.:	46703504, 46703505, 46703506, 46703507, and 46703509
40 CFR §180.	605		
Chemical Class:	Sulfonamide Herbicide		

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Executive Summary

Penoxsulam (XDE-638) is a sulfonamide herbicide currently registered on rice for the selective control of grasses, broadleaf, and sedge weeds. The use on rice (PP#3F6542, DP Barcode D288152, 8/11/04, W. Cutchin) represented the first food/feed use of penoxsulam. The herbicide's mode of action at the cellular level involves the inhibition of acetolactate synthase (ALS).

Dow AgroSciences LLC has now submitted a Section 3 registration application for the end-use product GF-443 SC as an aquatic herbicide (PP#5F7012). Concurrently, the petitioner has requested an exemption from the requirement of a tolerance on fish and shellfish when penoxsulam is applied in aquatic areas.

Tolerances for residues of penoxsulam are listed in 40 CFR §180.605. Tolerances of 0.02 and 0.50 ppm have been established for rice grain and straw, respectively. The tolerance expression is in terms of the parent herbicide, penoxsulam [2-(2,2-difluoroethoxy)-N-(5,8-dimethoxy[1,2,4] triazolo[1,5-c]pyrimidin-2-yl)-6-(trifluoromethyl)benzenesulfonamide].

GF-443 SC is a suspension concentrate formulation containing 21.7% (2 lb ai/gal) penoxsulam that may be applied in-water, as a foliar application, or as an exposed sediment application for pre-emergence control of aquatic weeds. GF-443 SC is now proposed for use as an aquatic herbicide at a rate up to 150 ppb (penoxsulam) in the water of lakes, ponds, canals, and reservoirs. Typical application rates of penoxsulam will be 10-20 ppb in an initial application with additional 'bump' applications of 5-10 ppb to keep the water concentration at 5-10 ppb for 45-90 days. There is a season maximum of all applications of 150 ppb. Although typical multiple application rates are proposed at 5-20 ppb, a single in-water application is allowed at up to the maximum rate of 150 ppb.

The nature of the residue in rice is adequately understood. Based on the submitted rice metabolism study, penoxsulam primarily degrades to its 5-OH metabolite (5-OH XDE-638) and to at least two minor unknown metabolites in rice matrices. Little translocation of penoxsulam residues or its metabolites into the grain was observed.

The nature of the residue in rotational crops is also adequately understood. The reviewed confined rotational crop study showed that no quantifiable residues of penoxsulam or 5-OH XDE-638 are expected to be present in the raw agricultural commodities of small grains, leafy vegetables, and root crops planted 90 days following treatment with penoxsulam at 0.045 or 0.090 lb ai/A (1x or 2x the label rate for rice). Although the data indicate that residues of a metabolite Penoxsulam-BSTCA could be present at >0.01 ppm in the foliage of root crops planted 90 days following treatment at 0.090 lb ai/A (2x), the MARC has concluded that Penoxsulam – BSTCA is not a residue of concern in rotated crops.

The nature of the residue in animals is adequately understood. The available goat and poultry metabolism data indicate that penoxsulam is primarily excreted and is not significantly metabolized in either goats or poultry. When animals were dosed with penoxsulam radiolabeled on the benzene ring or on the triazine ring, i.e. either side of the sulfonanilide bridge, no significant differences were observed between the two labels, implying that the sulfonanilide bridge in penoxsulam is not cleaved during livestock metabolism.

The HED Metabolism Assessment Review Committee (MARC) has previously determined that for the tolerance expression and risk assessment, the residue of concern for penoxsulam in plants, rotational crops, and livestock (including poultry), following the rice use, is parent only. (MARC Decision Memo TXR No. 005740, DP Barcode D305542, W. Cutchin, 19 July 2004)

The available analytical methodology (LC/MS/MS method; GRM 01.25) is considered to be adequate for the rice tolerance enforcement. The method that was used to collect data in the analysis of freshwater clam and catfish samples from the field accumulation study is a modification of LC/MS/MS Method GRM 05.08. Adequate method validation data, including data from an independent laboratory, were submitted for Method GRM 05.08 applied to bovine

matrices and fish tissues. The validated limit of quantitation (LOQ) is 0.01 ppm, and the calculated limit of detection (LOD) was 0.003 ppm in tested bovine matrices and in fish tissue. Method GRM 05.08 is similar to Method GRM 01.25, using a reasonably similar extraction; therefore, including the ILV that was submitted, Method GRM 05.08 should also be appropriate for enforcement of tolerances for fish and shellfish.

The FDA multiresidue protocol data show that penoxsulam is not adequately recovered using any of the protocol methods. The multiresidue data have been forwarded to FDA for further evaluation.

No supporting storage stability data were submitted to validate the storage conditions and intervals of samples taken from the magnitude of the residue study in freshwater clams and catfish. Looking at the study dates, however, the samples could not have been stored for more than 1.6 months prior to residue analysis. Because samples were stored for a relatively short interval and because the Agency has also previously noted in the rice petition (PP#3F6542) that comparative analysis of goat milk and tissue extracts show that residue profiles are similar after 135 and 300 days of sample collection, no additional supporting storage stability data are required.

To support the current request, a study investigating the nature and potential for bioaccumulation of penoxsulam residues in bluegill sunfish was submitted. Bluegill sunfish were exposed for 28 consecutive days to the radiolabeled test substance, [phenyl-U-^{14}C]penoxsulam or [het-2-^{14}C]penoxsulam, under static conditions at concentrations of 0.150 mg ai/L or 1.50 mg ai/L (1x and 10x the maximum annual proposed application rate). Following exposure of the fish in this study at 0.150 mg/L, total radioactive residues (TRR) were below the minimum quantifiable limit (MQL; <7.10 to <7.45 ppb) (<0.007 ppm) in fish samples exposed to [phenyl-U-^{14}C]penoxsulam, and ranged from <MQL to 11.4 ppb (0.0114 ppm) in fish samples exposed to [het-2-^{14}C]penoxsulam. Calculated bioconcentration factors (TRR in tissue/TRR in water) were $\leq$ 0.10, indicating that there is little potential for the test substance or its metabolites to bioaccumulate.

Selected edible bluegill fish tissues from the 1.5 mg ai/L treatments were subjected to residue characterization/identification. In Day 7 fish samples exposed to [phenyl-U-^{14}C]penoxsulam, the following components were identified: penoxsulam (34% TRR, 0.039 ppm), 5-hydroxy penoxsulam (14% TRR, 0.016 ppm), penoxsulam sulfonamide (4% TRR, 0.005 ppm), and penoxsulam BSTCA (3% TRR, 0.003 ppm). In Day 28 fish samples exposed to [het-2-^{14}C]penoxsulam, only penoxsulam (23% TRR, 0.021 ppm) and 5-hydroxy penoxsulam (9% TRR, 0.009 ppm) were identified. [Note, however, that these total characterized residues were 174% of TRR in day 7 fish and 235% of TRR in day 28 fish. No explanation was provided for this discrepancy. HED review cannot classify this study as scientifically acceptable until a clarification of this discrepancy and/or confirmatory data are submitted. While the discrepancy leaves uncertain the proportion of the TRR represented by penoxsulam, the conclusions below are expected to be valid within the range of that uncertainty.]
A study investigating the magnitude and potential for bioaccumulation of penoxsulam residues in freshwater clams and catfish was also submitted (MRID 46703507). The test organisms were exposed for 28 consecutive days to penoxsulam under static aquatic conditions at concentrations of 0.150 mg ai/L and 1.50 mg ai/L. At 0.15 mg ai/L (1x the annual rate), residues of

penoxsulam, per se, ranged from below the MQL (<3.99 ppb) to 18.3 ppb (0.018 ppm) in clams, and from below the MQL to 4.16 ppb (0.004 ppm) in catfish tissues. Thus, residues ranged up to 0.02 ppm in clams and up to 0.004 ppm in fish at 1x application rate. At the 10x (1.5 mg ai/L) application rate residues in clams ranged up to 141 ppb and up to 56 ppb in catfish. The bioconcentration factors (concentration in tissue/concentration in water) in all samples were ≤0.15 indicating that penoxsulam has very low potential to bioconcentrate in edible tissues of freshwater clams and catfish. Because concurrent recoveries and raw data were not submitted with this study, HED has classified it as scientifically acceptable pending submission of this supporting data.

A previously reported study (MRID: 45831101, DP Barcode: D288160, L. Shanaman, 4/22/04) also showed the TRR residues in crayfish during 14 days of exposure to 494 ppb penoxsulam in water, followed by 7 days of depuration. (494 ppb is slightly greater than 10x the maximum estimated concentration in rice paddy water at 45 ppb.). Maximum TRR in crayfish tail muscle occurred on day 11 of the treatment and was 14.4 ppb (DP No. 288152). On that basis it was concluded that no tolerance associated with the rice use was needed for crayfish (or crustaceans). Since the 10x concentration in paddy water of 45 ppb (0.5 ppm) from the rice use is ~3x the proposed annual 150 ppb (0.15 ppm) aquatic application rate, assuming linearity, at 1x the proposed maximum aquatic application rate the TRR in crayfish would thus be estimated at about 14.4 ppb/3 = 4.8 ppb (0.005 ppm) and, if the linear extrapolation held the other way, at 10x the crayfish TRR would be around 48 ppb (0.05 ppm).

HED has reviewed the available data, and finds that it does not support the petitioner's request for tolerance exemptions on shellfish and finfish, resulting from the proposed aquatic uses. The studies show real residues of penoxsulam at both the 10X rate and the 1X rate. At 1x the application rate (0.15 mg ai/L) the maximum penoxsulam residues in catfish were 4.16 ppb (0.004 ppm) and in clams were up to 18.3 ppb (0.018 ppm). In bluegills at 1X, the maximum TRR were 11.4 ppb (0.0114 ppm). If these results are multiplied by 34% of the TRR to estimate penoxsulam, per se, the estimated concentration of parent only becomes about 3.9 ppb (0.004 ppm). Based upon the crustacean study using rice treatment rates, and assuming linearity, TRR in crayfish in water treated at 0.15 mg ai/L are estimated to be at about 4.8 ppb (0.005 ppm). At 10X (1.5 mg ai/L) the maximum application rate residues of penoxsulam were found in catfish up to 56 ppb (0.6 ppm) and in clams up to 141 ppb (0.14 ppm). Based upon the data as submitted, residues in bluegills at 10X were up to 39 ppb (0.04 ppm). As extrapolated from the rice field study, TRR in crayfish are expected to be up to up to 48 ppb (0.05 ppm).

Tolerances are therefore required for fish. Based upon these studies the tolerance expression for fish should be penoxsulam, per se. A tolerance of penoxsulam residues on mollusc is tentatively recommended at 0.02 ppm; and a tolerance on finfish and crustacean (or crayfish) is tentatively recommended at 0.01 ppm.

In general, from the bluefish study, 5-hydroxypenoxsulam is present in penoxsulam residues at about 40% of the parent penoxsulam. Thus, residues of concern in fish (finfish, mollusc and crustacean) for risk assessment based upon these studies should be penoxsulam plus 5-hydroxy penoxsulam.

Approval the proposed use of GF-443 SC as an aquatic herbicide will additionally require magnitude of the residue data on representative irrigated crops, and appropriate processing data

for those crops, because penoxsulam-treated water from lakes, ponds, canals, and reservoirs may unknowingly be used by second parties to irrigate food/feed crops.

The water from proposed aquatic uses does not contribute significantly to livestock or poultry commodities. Therefore livestock and poultry feeding studies have not yet been required. However studies have not yet been submitted to show residues in crops treated with that water If significant residues are found in those potentially irrigated crops, livestock and poultry feeding studies may become necessary.

Regulatory Recommendations and Residue Chemistry Deficiencies

If the petitioner wishes to pursue aquatic uses for penoxsulam, the residue chemistry data deficiencies listed below must be fulfilled.

860.1400 Water, Fish, and Irrigated Crops

Additional confirmatory data/information must be submitted to upgrade the submitted sunfish study (MRID 46703506) to an acceptable status. Information is required regarding the chromatographic system (i.e., instrumentation and detection) and the LOD/LOQ of the methodologies used for identification/characterization of the residues. In addition, raw data are required to support the reported characterization/identification of radioactive residues in the edible fish tissues. The petitioner must address the source of the discrepancy between recoveries of identified/characxterized residues and TRR. The petitioner should consult OPPTS 860.1000 regarding the types of raw data required for this type of study submission.

Concurrent recoveries and raw data must also be submitted to support the catfish and clam study (MRID 46703507).

Because penoxsulam-treated water from lakes, ponds, canals, and reservoirs may unknowingly be used by a second party for irrigation of food/feed crops, the proposed use of GF-443 SC as an aquatic herbicide will also require magnitude of the residue data on crops that may potentially be irrigated, and certain such crops also will need processing data as appropriate. Technically, crops that may potentially be irrigated can include any U. S. grown crop and so selected crops for testing might need to include a representative crop from all crop groups. ChemSAC has discussed this requirement for penoxsulam (ChemSAC Minutes for 10 January 2007). Because this requirement could be very onerous and because penoxsulam is intended for use only at concentrations less than 150 lbs ai/L, ChemSAC has suggested that the petitioner should initially propose studies at the maximum application rate to a very limited number of crops that are likely to be worst case for residues. Worst case overhead irrigation should be used and, because treatment would be unintentional, crops should be sampled with a zero day post treatment interval. A grass, a leafy vegetable, a cereal, and perhaps a root crop should be included. Either TRR should be measured, or based upon rice and confined rotational crop studies, analysis for penxulam and 5-OH penoxsulam should be adequate. However, HED also understands that other plant metabolism studies may currently exist and recommends that submission of these would allow for a more comprehensive understanding of the residues requiring analysis. If residues on this limited selection of worst case crops are found to be negligible then no further work will be needed. If measurable residues are found present, additional field trials, possibly

with processing studies as needed, may be required. In addition, livestock and/or poultry studies may become required.

860.1550 Proposed Tolerances

The petitioner is required to propose tolerances on fish and shellfish in a revised Section F. Based on the available residue data for freshwater clams and catfish treated at 0.15 mg ai/L, HED tentatively recommends a tolerances of 0.02 ppm for mollusc, and 0.01 ppm for both fish and for crustaceans after a direct aquatic use. A registration using such tolerances, will of course, be dependent upon submission of required supporting information for the relevant studies.

Background

EPA granted a conditional registration to Dow AgroSciences for penoxsulam use on water- or dry-seeded rice on 9/27/04. The petitioner is currently requesting an exemption from the requirements of tolerances on fish and shellfish when penoxsulam is applied in aquatic areas such as lakes, reservoirs, ponds, and canals to control hydrilla, water hyacinth, egeria, and various other aquatic weeds. The nomenclature of penoxsulam is summarized in Table 1, and the physicochemical properties are summarized in Table 2.

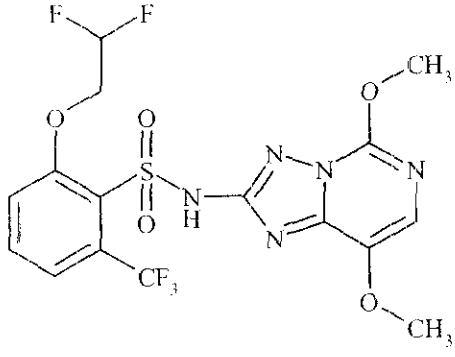
Table.1. Penoxsulam Nomenclature.	
Compound	
Common name	Penoxsulam
Company experimental name	XDE-638
IUPAC name	6-(2,2-difluoroethoxy)-N-(5,8-dimethoxy-s-triazolo[1,5-c]pyrimidin-2-yl)-α,α,α-trifluoro-o-toluenesulfonamide
CAS name	2-(2,2-difluoroethoxy)-N-(5,8-dimethoxy[1,2,4]triazolo[1,5-c] pyrimidin-2-yl)-6-(trifluoromethyl) benzenesulfonamide
CAS registry number	219714-96-2
End-use product (EP)	GF-443 SC

Table 2. Physicochemical Properties of Penoxsulam.		
Parameter	Value	Reference
Melting point/range	Not available	
pH	5.2	MRID 45830707
Density	1.61 g/mL at 20 °C	MRID 45830707
Water solubility at 19 °C	*Unbuffered* 4.91 mg/L pH 5 5.66 mg/L pH 7 408 mg/L pH 9 1460 mg/L	MRID 45830720
Solvent solubility at 19 °C	Xylene 0.017 g/L 1-Octanol 0.035 g/L Methanol 1.48 g/L Ethyl acetate 3.23 g/L Acetonitrile 15.3 g/L Acetone 20.3 g/L Dimethylsulfoxide 78.4 g/L	MRID 45830720
Vapor pressure	7.16×10^{-16} mm Hg at 25 °C	MRID 45830720
Dissociation constant, pK_a	5.1 (ambient)	MRID 45830720
Octanol/water partition coefficient, $Log(K_{OW})$	Unbuffered -0.354 pH 5 1.137 pH 7 -0.602 pH 9 -1.418	MRID 45830720
UV/visible absorption spectrum	Not available	

860.1200 Directions for Use

GF-443 SC is a suspension concentrate formulation containing 21.7% (2 lb ai/gal) penoxsulam.

The proposed label states

"Single In-Water Application to Treatment Zone

Apply GF-443 SC is applied as a single application at a rate of 5 – 150 ppb to the treatment zone. It may be necessary to re-treat the body of water if more established vegetation is present in the target area or heavy rainfall has diluted the treatment concentration. If re-treatment is necessary, refer to the Split of Multiple Applications section of the label. Note: The total concentration amount of all applications must not exceed 150 ppb per annual growth cycle."

"Split or Multiple In-Water Applications to Treatment Zone

To ensure efficacy over time and enhance selectivity, apply the minimum effective dose of 5 – 75 ppb to the treatment zone and, through the use of water analysis, add additional GF443 SC to maintain the lower concentration. Re-treat the water to maintain a sufficient concentration for satisfactory control of aquatic plants. Water analysis using ELISA is recommended to determine the actual concentration of GF-443 SC in the water over time. Note: The total concentration amount of all applications must not exceed 150 ppb per annual growth cycle."

For various types of application the label also specifies: For foliar applications apply at the rate of 2 – 5.6 fl oz per acre. For aerial application apply in a spray volume of 10 gallons per acre.

For boat or ground foliar applications, apply up to 100 gpa. For exposed sediment application, apply at the rate of 5.6 – 11.2 fl oz per acre in a total spray volume of 20 – 100 gpa.

Conclusions. Section G of the *Petition for Exemption from the Requirement for a Tolerance* states that : "Penoxsulam is proposed for use as an aquatic herbicide at a rate up to 150 ppb in the water of lakes, ponds, canals, and reservoirs. Typical application rates of penoxsulam will be 10-20 ppb in an initial application with additional 'bump' applications of 5-10 ppb to keep the water concentration at 5-10 ppb for 45-90 days. There is a season maximum of all applications of 150 ppb "

Although this statement in the petition is true, the maximum labeled single application use is 150 ppb (mg ai/L), the same as the maximum seasonal labeled use. At this maximum single application use rate, based upon the data submitted by the registrant to support an exemption, fish, mollusc and crustacean all have measurable residues and cannot be exempted from tolerances. A tolerance expressed as penoxsulam, per se, on mollusc is recommended at 0.02 ppm; and a tolerance, also expressed as penoxsulam, on finfish and on crustacean (or crayfish) is recommended at 0.01 ppm.

860.1300 Nature of the Residue - Plants

PP#3F6542: DP Barcode D288152, 8/11/04, W. Cutchin (45830712.der.wpd)
MARC Decision Memo TXR No. 0052740 (DP Barcode D305542, 7/19/04, W. Cutchin)

An acceptable rice metabolism study was reviewed in the rice petition (PP#3F6542). Based on the submitted rice metabolism study, penoxsulam primarily degrades to its 5-OH metabolite (5-OH XDE-638) and at least two minor unknown metabolites in rice matrices; little translocation of penoxsulam residues or its metabolites into the grain was observed.

Based upon this study the MARC determined that for the tolerance expression and risk assessment the residue of concern for penoxsulam in/on rice is parent only. The response to the rice petition (D288152), included a note that if uses on other crops are proposed, including uses on cereal grains, additional nature of the residue data will be needed. However, as an alternative to metabolism data on other cereal crops, the registrant might submit crop field trial data which include residue data for the metabolite 5-OH XDE-638 as well as parent.

860.1300 Nature of the Residue - Livestock

PP#3F6542: DP Barcode D288152, 8/11/04, W. Cutchin (45830713.der.wpd; 46267601.der.wpd)
MARC Decision Memo TXR No. 0052740 (DP Barcode D305542, 7/19/04, W. Cutchin)

Acceptable animal metabolism studies were reviewed in the rice petition (PP#3F6542). The available goat and poultry metabolism data indicate that penoxsulam is primarily excreted and not significantly metabolized in either goats or poultry. Because no significant differences were observed between the two labels, the sulfonanilide bridge in penoxsulam does not appear to be cleaved as a result of goat metabolism.

The MARC determined that for the tolerance expression and risk assessment, the residues of concern for penoxsulam in livestock (including poultry) is parent only.

860.1340 Residue Analytical Methods

Enforcement method PP#3F6542; DP Barcode D288152, 8/11/04, W. Cutchin (45830714.der.wpd)

In connection with the rice petition (PP#3F6542), Dow AgroSciences LLC has proposed an LC/MS/MS method, GRM 01.25, for the enforcement of tolerances for residues of penoxsulam in/on rice commodities. Using this method, samples of rice matrices are homogenized/extracted with acetonitrile/water; then shaken and centrifuged. An aliquot of the supernatant is diluted with water and cleaned up on a mixed-mode polymeric-anion exchange solid phase extraction (SPE) plate. Residues are eluted with ACN:formic acid (99.9:0.1, v:v), evaporated to dryness, and redissolved in mobile phase. Residues are quantitated by LC/MS/MS using a C8 column, a gradient mobile phase of ACN/methanol and water, each containing 0.1% acetic acid, and electrospray ionization in the positive ion mode. Residues are quantified using external standards. The validated limit of quantitation (LOQ) and calculated limit of detection (LOD) for penoxsulam were 0.01 and 0.002 ppm, respectively, in/on rice forage, straw, grain, hulls, bran, and polished rice. The LC/MS/MS method is adequate for enforcement of tolerances on plant commodities.

Method GRM 05.08, described below, may also serve as an acceptable tolerance enforcement method for residues in meat, milk and fish. An acceptable Interlaboratory Validation Study was done. The extraction step was reasonably similar to the extraction used in GRM 0.1.25 and in the Nature of the Residue in Fish Study, so no radiovalidation is needed. However, GRM 05.08 uses an isotopic internal standard. Hence, so long as the registrant agrees to supply EPA's Ft. Meade Standards Repository with penoxsulam standard and this isotopic internal standard for as long as the method exists, method GRM 05.08 can be acceptable as a tolerance enforcement method.

Data-collection method

46703504.der.doc (Includes MRID 46703505)

Dow AgroSciences has submitted Method GRM 05.08 for the determination of residues of penoxsulam in bovine milk, fat, kidney, liver, and muscle, and fish edible tissues. Using this method, residues in milk or ground tissue samples are extracted with acetonitrile/water for one hour at 60 °C. An aliquot of the extract is diluted with 0.1 N HCl for cleanup with an SPE Strata X (polymeric/hydrophobic) 96 well plate. The internal standard, penoxsulam stable isotope in methanol/water/acetic acid, is added to the extract prior to LC/MS/MS analysis. Residues are quantified using the ion transition m/z 484 to 195, and using the transition m/z 484 to 164 for confirmation. The validated limit of quantitation (LOQ) is 0.01 ppm, and the calculated limit of detection (LOD) was 0.003 ppm in all bovine matrices and fish tissue.

This method was adequately validated using fortified samples of bovine milk, fat, kidney, liver, and muscle and fish (edible tissue). Recoveries of penoxsulam averaged 93 ± 1.7% in milk, 98 ± 3.1% in bovine fat, 96 ± 3.3% in bovine kidney, 96 ± 2.3% in bovine liver, 95 ± 1.9% in bovine muscle, and 95 ± 2.4% in fish (edible tissue) fortified with penoxsulam at 0.01 (LOQ) and 0.1 ppm. In addition to the ion transition from the parent ion to a structurally significant product ion m/z 484/195 (quinazoline portion of the molecule), confirmation is available from a second transition of m/z 484/164. This second ion transition, which probably involves loss of a

methoxy group relative to the m/z 195 ion, should provide adequate confirmation unless homologous pesticides modified only at the phenyl end of the molecule are later introduced.

Method GRM 05.08 was successfully validated by an independent laboratory using milk and fish as the matrices. The ILV study included quantification of both ion transitions (484/195 m/z and 484/164 m/z) using two different HPLC systems (C8 or C18 column). A modification of Method GRM 05.08 was used for data-collection in the analysis of freshwater clam and catfish samples from a companion field accumulation study (MRID 46703507). However, no concurrent recoveries were submitted to support the modification of GRM 05.08 used in this catfish and clam study.

Conclusions: LC/MS/MS method GRM 01.25 is adequate for tolerance enforcement for rice commodities. LC/MS/MS Method GRM 05.08 has been submitted, apparently for tolerance enforcement. A modification of GRM 05.08 (no internal standard) is used for data-collection in the analysis of freshwater clam and catfish samples from the field accumulation study. Adequate method validation data, including those conducted by an independent laboratory, were submitted for Method GRM 05.08 using bovine matrices and fish tissues. The validated limit of quantitation (LOQ) is 0.01 ppm, and the calculated limit of detection (LOD) was 0.003 ppm in tested bovine matrices and fish tissue. Method GRM 05.08, however, uses an isotopic internal standard. Therefore, GRM 05.08 can also be acceptable for enforcement of tolerances for fish and shellfish, should tolerances be proposed, so long as the registrant agrees to supply the isotopic internal standard for as long as the method exists. Because no concurrent recoveries were submitted to support the modification of GRM 05.08 used in the catfish and clam study (MRID 46703507), this data must be submitted.

860.1360 Multiresidue Methods

PP#3F6542: DP Barcode D288152, 8/11/04, W. Cutchin (45830716.der.wpd)

Testing results of penoxsulam analyzed according to the FDA Multi-Residue Method Test guidelines in PAM Vol. I, Appendix II (1/94) were submitted in conjunction with the rice petition (PP# 3F6542). The multiresidue method data indicate that penoxsulam is not adequately recovered using any of the multiresidue methods. These data have been forwarded to FDA for further evaluation.

860.1380 Storage Stability

In the submitted magnitude of the residue study in freshwater clams and catfish (MRID 46703507), information pertaining to sample storage conditions and intervals was minimal with no analysis dates provided. The study submission reported the fish exposure dates as 8/26/05-9/23/05 (second study), and reported the study completion date as 10/14/05. Based on these dates, clam/catfish samples may have been stored for up to 49 days (1.6 months) from collection to analysis.

No supporting storage stability data were submitted to validate the storage conditions and intervals of samples taken from the magnitude of the residue study in freshwater clams and catfish. However, it appears that samples could not have been stored for longer than 1.6 months

prior to residue analysis. Because samples were stored for a relatively short interval and because the Agency has previously noted in the rice petition (PP#3F6542) that comparative analysis of goat milk and tissue extracts show that residue profiles are similar after 135 and 300 days of sample collection, no supporting storage stability data are required.

860.1400 Water, Fish, and Irrigated Crops

The rice petition (PP#3F6542; DP Barcode D288152, 8/11/04, W. Cutchin) reported the results of a bioaccumulation study (MRID 45831101; DP Barcode D288160, 4/22/04, L. Shanaman) which investigated the residues of penoxsulam in crayfish. An EFED review of the study concluded that penoxsulam has very low potential to bioconcentrate in edible tissues of crayfish. Following exposure to 494 ppb penoxsulam in water (>10x the 45 ppb screening level recommended by EFED for the use in rice; L. Shanaman, 7/8/04), total radioactive residues in crayfish tail muscle were 14.4 ppb (0.014 ppm); the average steady-state calculated bioconcentration factor (BCF) was 0.02 mL/g. It was concluded that the available data for crayfish indicate that tolerances for penoxsulam residues in crayfish are not required to support the rice use.

In the current petition, the petitioner has submitted separate studies investigating (i) the nature and potential for bioaccumulation of penoxsulam residues in bluegill sunfish and (ii) the magnitude and potential for bioaccumulation of penoxsulam residues in freshwater clams and catfish.

Bluegill sunfish

46703506.der.doc

Dow AgroSciences has submitted a study investigating the nature and potential for bioaccumulation of penoxsulam residues in bluegill sunfish. Bluegill sunfish were exposed for 28 consecutive days to the radiolabeled test substance, [phenyl-U-^{14}C]penoxsulam or [het-2-^{14}C]penoxsulam, under static conditions at concentrations of 0.150 mg ai/L or 1.50 mg ai/L. The applied rates are 1x and 10x the maximum allowed annual label rate. Separate aquatic chambers, containing ~150 fish each at the start, were designed for the two radiolabels, two concentration rates, and the control. The study was conducted by ABC Laboratories, Inc. (Columbia, MO). The concentrations of the test water solutions were periodically verified during the study period and were generally within the applied rate for each radiolabel with an overall range of 73-96% of the nominal rate.

Following exposure of sunfish at 1x, total radioactive residues (TRR) were below the minimum quantifiable limit (MQL; <7.10 to <7.45 ppb) in all fish samples exposed to [phenyl-U-^{14}C]penoxsulam, and ranged from <MQL to 11.4 ppb in fish samples exposed to [het-2-^{14}C]penoxsulam. Following exposure of fish at 10x, TRR ranged from 28.5 to 105 ppb in fish samples exposed to [phenyl-U-^{14}C]penoxsulam, and from <MQL to 42.6 ppb in fish samples exposed to [het-2-^{14}C]penoxsulam. TRR were highest in Day 1 fish treated with [phenyl-U-^{14}C]penoxsulam and Day 21 fish treated with [het-2-^{14}C]penoxsulam. The TRR appears to plateau after approximately 7 days of exposure.

The bioconcentration factors (BCF; TRR in tissue/TRR in water) were calculated where possible;

all calculated BCF values were ≤ 0.10, indicating that there is little potential for the test substance or its metabolites to bioaccumulate.

Select edible fish tissues from the 10x rate treatments were subjected to residue characterization/ identification; however, raw data were not submitted to support these reported results. In Day 7 fish samples exposed to [phenyl-U-^{14}C]penoxsulam, the following components were identified: penoxsulam (34% TRR, 0.039 ppm), 5-hydroxy penoxsulam (14% TRR, 0.016 ppm), penoxsulam sulfonamide (4% TRR, 0.005 ppm), and penoxsulam BSTCA (3% TRR, 0.003 ppm). In Day 28 fish samples exposed to [het-2-^{14}C]penoxsulam, only penoxsulam (23% TRR, 0.021 ppm) and 5-hydroxy penoxsulam (9% TRR, 0.009 ppm) were identified. Bound residues were reported as 18% TRR (0.021 ppm) and 25% TRR (0.023 ppm) in fish treated with [phenyl-U-^{14}C]penoxsulam or [het-2-^{14}C]penoxsulam, respectively. The %TRR reported by the petitioner represents the percent of the total residues that could be extracted and chromatographed plus bound residues. Compared to the TRR determined by combustion/LSC, the above accountabilities were 174-235% of the combustion TRR. No explanation was provided to account for this discrepancy between accountability and original TRR of about 200%. Because these accountabilities of 174-235% may arise from concentration of some components of the residue relative to others, until the discrepancy is explained it is not possible to determine the scientific acceptability of this study.

The chemical names and structures of metabolites identified in this study are presented in Appendix 1.

Information pertaining to sample storage conditions and intervals was minimal and no analysis dates were provided. The study submission reported the exposure dates of the initial and second study. Based on the study completion date, fish samples may have been stored for up to 280 days (9.2 months) from collection to completion of residue characterization. No storage stability data were provided. The Agency has previously noted in PP#3F6542 (DP Barcode D288152, 8/11/04, W. Cutchin) that comparative analysis of goat milk and tissue extracts show that residue profiles are similar after 135 and 300 days of sample collection, therefore supporting storage stability data will not be required for the subject fish metabolism study. We note for future submissions that the petitioner should submit individual sample TRR and chromatographic analysis dates.

Conclusions: An explanation of the 174-235% accountabilities must be submitted before it can be determined if the bluegill sunfish study is scientifically acceptable. In addition, confirmatory data/information must be submitted; that is, the petitioner is required to provide additional information regarding the chromatographic system (i.e., instrumentation and detection) and include the LOD/LOQ of the methodologies used for identification/characterization of the residues. In addition, raw data are required to support the reported characterization/identification of radioactive residues in the edible fish tissues.

Freshwater clams and catfish

46703507.der.doc

Dow AgroSciences has submitted a study investigating the magnitude and potential for bioaccumulation of penoxsulam residues in freshwater clams and catfish. The test organisms were exposed for 28 consecutive days to penoxsulam under static aquatic conditions at

concentrations of 0.150 mg ai/L and 1.50 mg ai/L. The applied rates are equivalent to 1x and 10x the maximum labeled rates. Separate aquatic chambers, each containing ~60 fish and 300 clams at initiation, were designed for the two concentration rates and control. Samples of the water solutions as well as clams and catfish were collected at regular intervals during the 28-day exposure period.

Water and edible tissues of fish and clams were analyzed for residues of penoxsulam using methodology based on Dow AgroSciences GRM 05.08 entitled "Determination of Residues of Penoxsulam in Bovine Tissues and Fish by High Performance Liquid Chromatography with Tandem Mass Spectrometry." This method is adequate for data collection based on validation data reported in the 860.1340 DER for MRIDs 46703504 and 46703505. The reported minimum quantifiable limits (MQLs) were 3.99 ppb for the clam and fish tissue samples, and 45 ppb for the water samples. The method actually used is only briefly described but appears to be the same as GRM 05.08 except that no isotopically labeled standard is used as an internal standard. Concurrent recovery data were not reported.

Samples could not have been stored for greater than 1.6 months prior to residue analysis; but actual study analysis dates were not provided. No supporting storage stability data were submitted, but translation of the available stability data from an animal metabolism study discussed in PP#3F6542 (DP Barcode D288152, 8/11/04, W. Cutchin) can be used to support stability of these samples.

The concentrations of the test water solutions were periodically verified during the study period. The water solutions were generally within the applied rate with an overall range of 68-115% except those samples collected from the 4 hour post application time period (163% in the 1x rate study); data from this time period may indicate that the test substance might not have been completely mixed in the test system by that time.

Residues of penoxsulam ranged from below the MQL (<3.99 ppb) to 18.3 ppb in clams, and <MQL to 4.16 ppb in catfish tissues following exposure at 0.15 mg ai/L (1x). Following exposure at 10x, residues ranged 6.16–141 ppb in clams and <MQL–56.3 ppb in catfish tissues. Residues were highest in clams exposed for 21/25 days, and in catfish exposed for 14 days (10x).

The bioconcentration factors (BCF; concentration in tissue/concentration in water) were calculated where possible. The calculated clam BCF values ranged 0.033-0.15 and 0.0041-0.098 for the 1x and 10x treatment levels, respectively. Except for the 4-hour post-application sample with a calculated BCF value of 0.017, all other catfish tissue concentrations from the 1x treatment were less than the MQL; therefore, no BCF could be calculated. The calculated catfish BCF values ranged 0.0029-0.033 for the 10x treatment level.

Conclusions:

Based on the available data, the submitted studies do not support the petitioner's request for tolerance exemptions on shellfish and finfish resulting from the proposed aquatic uses. At the 10X treatment rate (1.5 mg ai/L) for the proposed aquatic use, residues of penoxsulam, per se, in clams were up to 141 ppb (0.140 ppm), residues in catfish up to 56 ppb (0.056 ppm), and residues in bluegill, based upon the data as reported, were up to ppb (0.039 ppm). TRR (not

penoxsulam, per se) in crayfish in water treated at 10x, assuming linearity, are estimated to be about 50 ppb (0.05 ppm).

At 1x (0.15 mg ai/L) the data show that the maximum penoxsulam residues in catfish treated at were 4.16 ppb (0.004 ppm). At 1x (0.15 mg ai/L) in bluegills, the maximum TRR were 11.4 ppb (0.0114 ppm) (and times penoxsulam at the reported 34% TRR) the residues of penoxsulam residues, per se, would also be about 3.9 ppb (0.004 ppm). Penoxsulam residues, per se, in clams in water treated at 1x (0.15 mg/L) were up to 18.3 ppb (0.018 ppm). Based upon the crustacean study using rice treatment rates, and assuming linearity, TRR in crayfish treated at 0.15 mg ai/L are estimated to be at about 4.8 ppb (0.005 ppm). Based upon these studies the tolerance expression for fish should be penoxsulam, per se. Therefore, a tolerance of penoxsulam residues on mollusc is recommended at 0.02 ppm; and a tolerance on finfish and crustacean (or crayfish) is recommended at 0.01 ppm.

In general, from the bluefish study, 5-hydroxypenoxsulam is present in penoxsulam residues at about 40% of the parent penoxsulam. Thus, residues of concern for risk assessment based upon these studies should be penoxsulam plus 5-hydroxy penoxsulam.

In addition, the proposed use of GF-443 SC for use as an aquatic herbicide will require submission of magnitude of the residue data on irrigated crops because penoxsulam-treated water from lakes, ponds, canals, and reservoirs may unknowingly be used by second parties for irrigation of food/feed crops.

860.1460 Food Handling

There are no proposed uses in the current petition which are relevant to this guideline topic.

860.1480 Meat, Milk, Poultry, and Eggs

Penoxsulam-treated water may be ingested by livestock animals. The contribution of penoxsulam to the dietary burdens of livestock animals, resulting from the proposed aquatic use (and including the registered use on rice), is presented below in Table 3.

Table 3. Calculation of Maximum Theoretical Dietary Burdens of Penoxsulam to Livestock.

Feed Commodity	% Dry Matter[1]	% Diet[1]	Established Tolerance, ppm	Dietary Contribution, ppm[2]
Beef Cattle				
Rice, straw	90	10	0.50	0.056
Rice, grain	88	40	0.02	0.009
Rice, hulls	90	10	0.02 (grain)	0.002
Rice, bran	90	15	0.02 (grain)	0.003
Water	NA	NA	NA	0.150 [3]
TOTAL BURDEN	NA	75	NA	**0.220**

Table 3. Calculation of Maximum Theoretical Dietary Burdens of Penoxsulam to Livestock.

Feed Commodity	% Dry Matter[1]	% Diet[1]	Established Tolerance, ppm	Dietary Contribution, ppm[2]
Dairy Cattle				
Rice, straw	90	10	0.50	0.056
Rice, grain	88	40	0.02	0.009
Rice, hulls	90	10	0.02 (grain)	0.002
Rice, bran	90	15	0.02 (grain)	0.003
Water	NA	NA	NA	0.150 [3]
TOTAL BURDEN	NA	NA	NA	**0.220**
Poultry				
Rice, grain	NA	60	0.02	0.012
Rice, hulls	NA	15	0.02 (grain)	0.003
Rice, bran	NA	25	0.02 (grain)	0.005
Water	NA	NA	NA	0.150 [3]
TOTAL BURDEN	NA	100	NA	**0.170**
Swine				
Rice, grain	NA	65	0.02	0.013
Rice, bran	NA	15	0.02 (grain)	0.003
Water	NA	NA	NA	0.150 [3]
TOTAL BURDEN	NA	80	NA	**0.166**

[1] Table 1 (OPPTS Guideline 860.1000).

[2] Contribution = [tolerance / % DM (cattle)] x % diet). Poultry and swine diets are not corrected for % dry matter.

[3] This assumes that water consumed by livestock animal will be treated at 0.150 ppm (equal to the maximum application rate of 150 ppb).

The petitioner did not submit any livestock feeding studies with this petition. The maximum residues of penoxsulam observed in any matrix in the goat metabolism study were 0.047 ppm in kidney from the goat dosed with [PH-^{14}C]penoxsulam at 12.4 ppm (56x the dietary burden for beef and dairy cattle). When residues are extrapolated at 10x of this dietary burden, residues are expected to be 0.009 ppm, which is less than the (plant) method LOQ of 0.01 ppm. Note, however, that this dietary burden does not include potential residues on crops that are inadvertently irrigated with treated water.

The maximum residues of penoxsulam observed in any matrix in the poultry metabolism study were 0.017 ppm in liver from hens dosed with [TP-^{14}C]penoxsulam at 11 ppm (65x the dietary burden for poultry). When residues are extrapolated at 10x of the dietary burden, residues are expected to be 0.003 ppm, which is less than the (plant) method LOQ of 0.01 ppm.

Conclusions. Thus if no residues occur in crops inadvertently irrigated with penoxsulam treated water, the proposed aquatic uses of penoxsulam could result in a 40 CFR §180.6(a)(3) situation for ruminant and poultry commodities; i.e., there would no reasonable expectation of finite residues in ruminant and poultry commodities, and no ruminant or poultry feeding study would need to be submitted. This conclusion must, however, be considered tentative because a study on the magnitude of the residue in irrigated crops is required to support the proposed aquatic uses. If significant residues are found in crops irrigated with penoxsulam treated water, then livestock feeding studies may become necessary.

860.1500 Crop Field Trials

As previously stated in 860.1400, because penoxsulam-treated water from lakes, ponds, canals, and reservoirs may be unknowingly be used by second parties for irrigation of food/feed crops, the proposed use of GF-443 SC as an aquatic herbicide will also require magnitude of the residue data on crops that may potentially be irrigated, and certain such crops also will need processing data as appropriate. Technically, crops that may potentially be irrigated can include any U. S. grown crop, so selected crops for testing might need to include a representative crop from all crop groups. ChemSAC has discussed this requirement for penoxsulam (ChemSAC Minutes for 10 January 2007). Because this full requirement could be very onerous and because penoxsulam is intended for use only at concentrations less than 150 lbs ai/L, ChemSAC has suggested that the petitioner should initially propose studies at the maximum application rate to a very limited number of crops that are likely to be worst case for residues. Worst case overhead irrigation should be used and, because treatment would be done unknowingly, crops should be sampled with a zero day post treatment interval. A grass, a leafy vegetable, a cereal, and perhaps a root crop should be included. Either TRR should be measured, or based upon rice and confined rotational crop studies, analysis for penxulam and 5-OH penoxsulam should be adequate. However, HED also understands that other plant metabolism studies may currently exist and recommends that submission of these would allow for a more comprehensive understanding of the residues requiring analysis. If residues on this limited selection of worst case crops are found to be negligible then no further work will be needed. If measurable residues are found present, additional field trials, possibly with processing studies as needed, may be required.

860.1520 Processed Food and Feed

Processing data on representative irrigated crops are required to support the aquatic uses if residues may occur in the RAC.

860.1650 Submittal of Analytical Reference Standards

The rice petition (PP#3F6542) reported that an analytical reference standard for penoxsulam is available at the EPA National Pesticide Standards Repository (expiration date January 2007). In addition, several metabolite standards are available. If GRM 05.08 is to be used as a tolerance enforcement method the registrant must also supply the radiolabeled internal standard to the repository

860.1850 Confined Accumulation in Rotational Crops

PP#3F6542; DP Barcode D288152, 8/11/04, W. Cutchin (45830720.der.wpd)

An acceptable crop rotational crop study was reviewed in the rice petition (PP#3F6542). HED review of the study concluded that no quantifiable residues of penoxsulam or 5-OH XDE-638 are expected to be present in the raw agricultural commodities of small grains, leafy vegetables, and root crops planted 90 days following treatment with penoxsulam at 0.045 or 0.090 lb ai/A (1x or 2x the rate for rice). The data also indicate that residues of the metabolite penoxsulam BSTCA could be present at >0.01 ppm in the foliage of root crops planted 90 days following treatment at 0.090 lb ai/A (2x). The MARC determined that penoxsulam BSTCA is not a residue of concern for penoxsulam in rotated crops.

It was noted that the submitted confined rotational crop study only included one plantback interval, 90 days. If in the future plantback intervals other than 90 days are proposed, an additional confined rotational crop study reflecting the proposed plantback interval would be required.

860.1900 Field Accumulation in Rotational Crops

Based on data from the confined rotational crop study, no quantifiable residues of penoxsulam, its 5-OH metabolite, or BSTCA are expected to be present in the raw agricultural commodities of small grains, leafy vegetables, and root crops planted 90 days following treatment with penoxsulam at 1x the maximum seasonal rate. Therefore, field rotational crop studies are not required to support this petition.

860.1550 Proposed Tolerances

The MARC previously determined that for the tolerance expression and risk assessment the residue of concern for penoxsulam in plants, ruminants and rotated crops is parent only. Tolerances for residues of penoxsulam are listed in 40 CFR §180.605. The tolerance expression is in terms of the penoxsulam *per se*. Tolerances of 0.02 and 0.50 ppm have been established for rice grain and straw, respectively.

In the current petition, Dow AgroSciences requests an exemption from the requirements of tolerances on fish and shellfish when penoxsulam is applied in aquatic areas. HED has reviewed the available data, and finds that it does not support the petitioner's request for tolerance exemptions on shellfish and finfish, resulting from the proposed aquatic uses. The studies show real residues of penoxsulam at both the 10X rate and the 1X rate. At 1x the application rate (0.15 mg ai/L) the maximum penoxsulam residues in catfish were 4.16 ppb (0.004 ppm) and in clams were up to 18.3 ppb (0.018 ppm). In bluegills at 1X, the maximum TRR were 11.4 ppb (0.0114 ppm). If these results are multiplied by 34% of the TRR to estimate penoxsulam, per se, the estimated concentration of parent only becomes about 3.9 ppb (0.004 ppm). Based upon the crustacean study using rice treatment rates, and assuming linearity, TRR in crayfish in water treated at 0.15 mg ai/L are estimated to be at about 4.8 ppb (0.005 ppm). At 10X (1.5 mg ai/L) the maximum application rate residues of penoxsulam were found in catfish up to 56 ppb (0.6 ppm) and in clams up to 141 ppb (0.14 ppm). Based upon the data as submitted, residues in bluegills at 10X were up to 39 ppb (0.04 ppm). As extrapolated from the rice field study, TRR in

crayfish are expected to be up to up to 48 ppb (0.05 ppm).

Based upon the reviewed studies the tolerance expression for fish should be penoxsulam, per se. In general, from the bluefish study, 5-hydroxypenoxsulam is present in penoxsulam residues at about 40% of the parent penoxsulam. Thus, residues of concern in fish (finfish, mollusc and crustacean) for risk assessment based upon these studies should be penoxsulam plus 5-hydroxy penoxsulam.

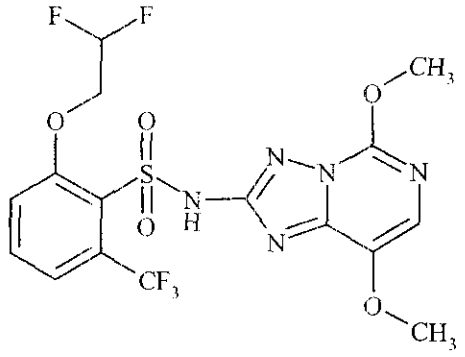
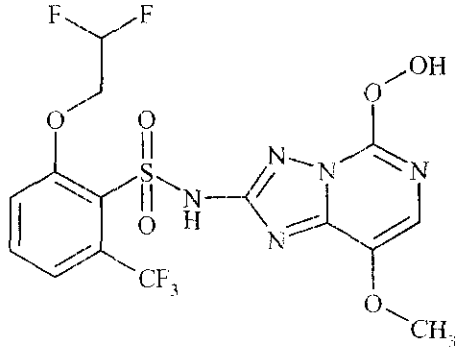
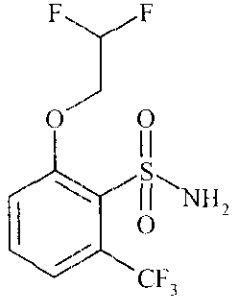
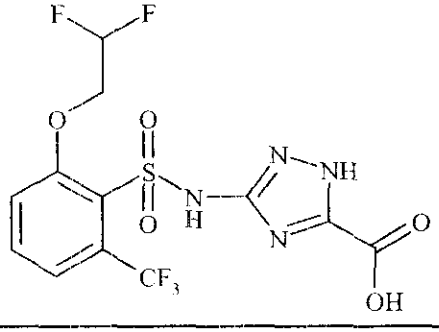
The petitioner is required to propose tolerances and submit a revised Section F.

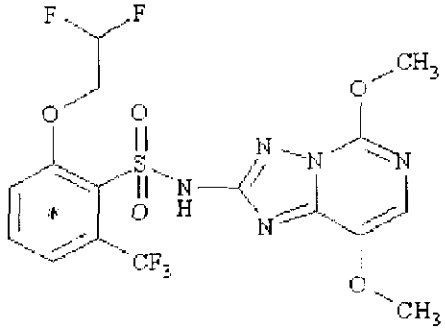
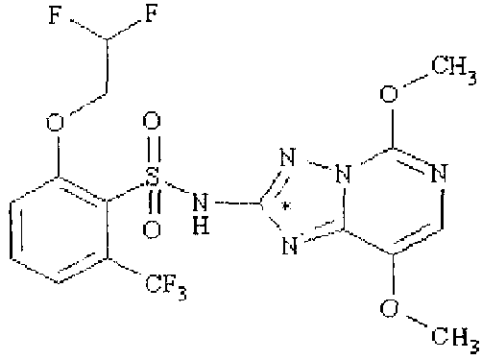
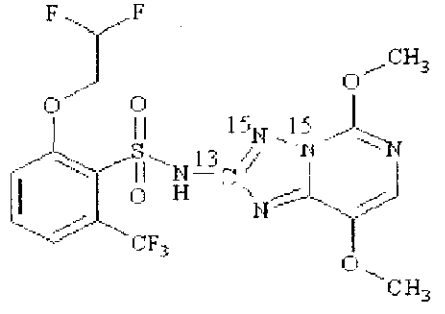
Based on the available residue data for freshwater clams and catfish, HED tentatively recommends a tolerances of 0.02 ppm for shellfish and 0.01 ppm for fish, pending verification of the label rates and aquatic use pattern.

References

DP Barcode: D288152
Subject: Penoxsulam. Petition for the Establishment of Permanent Tolerances for the Use on Rice. Summary of Analytical Chemistry and Residue Data. PP#3F6542
From: W. Cutchin
To: P. Errico/J. Miller
Date: 8/11/04
MRIDs: 45830712-45830717, 45830719-20, 46267601

Attachments
Appendix I - Chemical Name and Structure of Metabolites Identified in a Sunfish Metabolism Study

APPENDIX 1. Identification of Compounds from a Bluegill Sunfish Metabolism Study (MRID 46703506)		
Common name (Code name)	Chemical name	Chemical structure
Penoxsulam (DE-638, XDE-638, XR-638, X638177)	CAS name: 2-(2,2-difluoroethoxy)-N-(5,8-dimethoxy[1,2,4]triazolo[1,5-c] pyrimidin-2-yl)-6-(trifluoromethyl) benzenesulfonamide	
5-Hydroxy penoxsulam (5-Hydroxy-XDE-638)	2-(2,2-difluoroethoxy)-N-(5,6-dihydro-8-methoxy-5-oxo[1,2,4]triazolo[1,5-c] pyrimidin-2-yl)-6-trifluoromethyl) benzenesulfonamide	
Penoxsulam sulfonamide (XDE-638 sulfonamide)	2-(2,2-difluoroethoxy)-6-(trifluoromethyl)benzenesulfonamide	
Penoxsulam BSTCA (Triethylammonium of XDE-638)	Triethylammonium 5-(2,(2,2-difluoroethoxy)-6-trifluoromethyl benzenesulfonyl amino)-1H-1,2,4-triazole-3,carboxylate	

APPENDIX II. Radiolabeld Penoxulam Compounds Discussed in Text		
Common name (Code name)	Chemical name	Chemical structure
[phenyl-UL-^{14}C]penoxsulam)	N-((1-(2,2-difluoroethoxy)-3-trifluoromethyl-2-benzene-Ph-UL-^{14}C)sulfonyl)-2-amino-5,8-dimethoxy-1,2,4-*triazolo[1.5-c]pyrimidine*	
[het-2-^{14}C]penoxsulam 5-	N-[(1-(2,2-difluoroethoxy)-3-trifluoromethyl)-2-benzene)sulfonyl]-2-amino-5,8-dimethoxy-1,2,4-triazolo[1.5-c]pyrimidine-2-^{14}C	
Radiostable Penoxsulam Internal Standard	2-2,2-difluoroethoxy)-*N*-(5,8-dimethoxy[1,2,4]triazolo[1,5-*c*]pyrimidin-2-yl-2-^{13}C-3,4-$^{15}N_2$)-6-(trifluoromethyl)-benzenesulfonamide	

Primary Evaluator _____ Date: 18 Dec 2006
David Soderberg, Chemist, HED, RRB3

Approved by _____ Date: 18 Dec 2006
Danette Drew, Senior Scientist, HED, RRB3

This DER was originally prepared under contract by Dynamac Corporation (2275 Research Boulevard, Suite 300; Rockville, MD 20850; submitted 08/02/2006). The DER has been reviewed by the Health Effects Division (HED) and revised to reflect current Office of Pesticide Programs (OPP) policies.

STUDY REPORTS:

46703504 Shackelford, D. (2005) Validation Report for Method GRM 05.08 - Determination of Residues of Penoxsulam in Bovine Tissues and Fish by High Performance Liquid Chromatography with Tandem Mass Spectrometry. Project Number: 041029, GRM/05/08. Unpublished study prepared by Dow Agrosciences LLC. 62 p.

46703505 Class, T. (2005) Independent Laboratory Validation of Dow Agrosciences LLC Method GRM 05.08 - Determination of Residues of Penoxsulam in Bovine Tissues and Fish by High Performance Liquid Chromatography with Tandem Mass Spectrometry. Project Number: 050026, P/887/G, GRM/05/08. Unpublished study prepared by PTRL Europe Gmbh. 90 p.

EXECUTIVE SUMMARY:

Dow AgroSciences has submitted Method GRM 05.08 for the determination of residues of penoxsulam in bovine milk, fat, kidney, liver, and muscle, and fish edible tissues. Using this method, residues in milk or ground tissue samples are extracted for 1 hour with acetonitrile/water at 60 °C. An aliquot of the extract is diluted with 0.1 N HCl for cleanup with a solid phase extraction (SPE) well plate. The internal standard, penoxsulam stable isotope (in methanol/water/acetic acid), is added to the extract just prior to LC/MS/MS analysis. Residues are quantitated using the ion transition m/z 484 to 195, and using the transition m/z 484 to 164 for confirmation. The validated limit of quantitation (LOQ) is 0.01 ppm, and the calculated limit of detection (LOD) was 0.003 ppm in all bovine matrices and fish tissue.

The method was adequately validated using fortified samples of bovine milk, fat, kidney, liver, and muscle, and fish (edible tissue). Recoveries of penoxsulam averaged 93 ± 1.7% in milk, 98 ± 3.1% in bovine fat, 96 ± 3.3% in bovine kidney, 96 ± 2.3% in bovine liver, 95 ± 1.9% in bovine muscle, and 95 ± 2.4% in fish (edible tissue) fortified with penoxsulam at 0.01 ppm (LOQ) and 0.1 ppm. Because the submitted method monitors the transition of the molecular ion to a product ion at m/z 484/195, a second transition at m/z 484/164 can serve to provide adequate confirmation of the molecular structure.

Method GRM 05.08 was successfully validated by an independent laboratory using milk and fish as the matrices. The ILV study included quantitation of both ion transitions (484/195 m/z and 484/164 m/z) using two different HPLC systems (C8 or C18 column). Method GRM 05.08 was the data-collection method used in the analysis of freshwater clam and catfish samples from a companion field accumulation study (MRID 46703507) submitted in conjunction with DP Barcode D326985. Method GRM 05.08 is similar to Method GRM 01.25, the proposed enforcement method in an earlier rice petition (PP#3F6542; D288152, 8/11/04, W. Cutchin) which has been forwarded to ACB/BEAD for a regulatory method validation, except that GRM 05.08 uses an isotopic internal standard.

STUDY/WAIVER ACCEPTABILITY/DEFICIENCIES/CLARIFICATIONS:

Under the conditions and parameters used in the study, analytical method test data are scientifically acceptable. The acceptability of this study for regulatory purposes is addressed in the forthcoming U.S. EPA Residue Chemistry Summary Document, DP Barcode D326985.

COMPLIANCE:

Signed and dated Good Laboratory Practice (GLP), Quality Assurance and Data Confidentiality statements were provided. No deviations from regulatory requirements were reported which would have an impact on the validity of the study.

A. BACKGROUND INFORMATION

Penoxsulam is a sulfonamide herbicide currently registered for use on rice for selective control of grasses, broadleaf, and sedge weeds. Dow AgroSciences is proposing new aquatic uses of penoxsulam for the control aquatic plant pests such as hydrilla, water hyacinth, egeria, and various others in lakes, reservoirs, ponds, and canals. The mode of action at the cellular level involves the inhibition of acetolacetate synthase (ALS).

TABLE A.1. Test Compound Nomenclature.

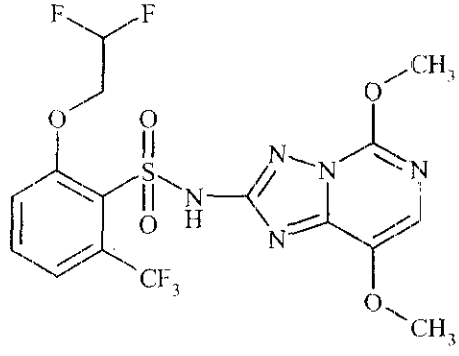
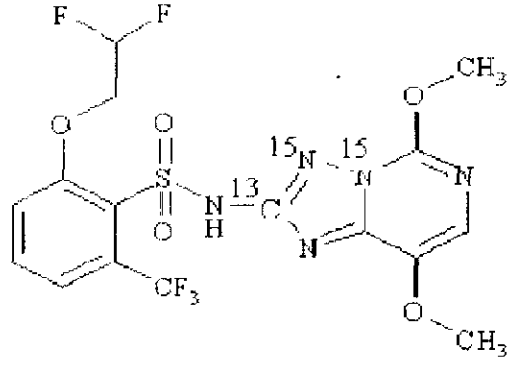
Compound	
Common name	Penoxsulam
Company experimental name	XDE-638
IUPAC name	6-(2,2-difluoroethoxy)-N-(5,8-dimethoxy-s-triazolo[1,5-c]pyrimidin-2-yl)-α,α,α-trifluoro-o-toluenesulfonamide

TABLE A.1. Test Compound Nomenclature.	
CAS name	2-(2,2-difluoroethoxy)-N-(5,8-dimethoxy[1,2,4]triazolo[1,5-c] pyrimidin-2-yl)-6-(trifluoromethyl) benzenesulfonamide
CAS registry number	219714-96-2
End-use product (EP)	GF-443 SC

TABLE A.2. Physicochemical Properties of the Technical Grade Test Compound: Penoxsulam.			
Parameter	Value		Reference
Melting point/range	Not available		
pH	5.2		MRID 45830707
Density	1.61 g/mL at 20 °C		MRID 45830707
Water solubility at 19 °C	Unbuffered pH 5 pH 7 pH 9	4.91 mg/L 5.66 mg/L 408 mg/L 1460 mg/L	MRID 45830720
Solvent solubility at 19 °C	Xylene 1-Octanol Methanol Ethyl acetate *Acetonitrile* Acetone Dimethylsulfoxide	0.017 g/L 0.035 g/L 1.48 g/L 3.23 g/L 15.3 g/L 20.3 g/L 78.4 g/L	MRID 45830720
Vapor pressure	7.16×10^{-16} *mm Hg at 25 °C*		MRID 45830720
Dissociation constant, pK_a	5.1 (ambient)		MRID 45830720
Octanol/water partition coefficient, $Log(K_{OW})$	Unbuffered pH 5 pH 7 pH 9	-0.354 1.137 -0.602 -1.418	MRID 45830720
UV/visible absorption spectrum	Not available		

TABLE A.3. Stable Isotope Structure.	
Penxsulam Stable Isotope 2-2,2-difluoroethoxy)-*N*-(5,8-dimethoxy[1,2,4]triazolo[1,5-c]pyrimidin-2-yl-2-^{13}C-3,4-$^{15}N_2$)-6-(trifluoromethyl)-benzenesulfonamide	

B. MATERIALS AND METHODS

B.1. Data-Gathering Method

Samples of freshwater clam and catfish from the field accumulation study (MRID 46703507) submitted in conjunction with DP Barcode D326985 were analyzed for residues of penoxsulam using Dow AgroSciences' Method GRM 05.08, "Determination of the Residues of Penoxsulam in Bovine Tissues and Fish by High Performance Liquid Chromatography with Tandem Spectrometry." The petitioner has submitted descriptions and method validation data for the method. The method is written for analysis of penoxsulam residues in bovine matrices (muscle, kidney, liver, fat and milk) and fish edible tissue.

B.1.1. Principle of the Method

Briefly, milk or ground tissue samples are extracted with acetonitrile (ACN)/water for one hour at 60 °C. An aliquot of the extract is diluted with 0.1 N HCl for cleanup with a solid phase extraction (SPE) well plate. The internal standard, a penoxsulam stable isotope in methanol/water/acetic acid, is added to the extract prior to LC/MS/MS analysis.

TABLE B.1.1. Summary Parameters for the Analytical Method Used for the Quantitation of Penoxsulam Residues in Bovine Matrices and Fish Tissue.

Method ID	Method GRM 05.08
Analyte(s)	Penoxsulam
Extraction solvent/technique	Milk or ground tissue samples are extracted with ACN:water (8:2, v:v) in a water bath at 60 °C for one hour and then shaken for a minimum of 30 minutes. The extract volume is adjusted for solvent loss with additional ACN/water and centrifuged.
Cleanup strategies	An aliquot of the extract is mixed with 0.1 N HCl (1:2, v:v) for cleanup on a polymeric sorbent SPE 96-well plate using ACN to elute the residues. The eluate is collected in a SPE collection plate, and the internal standard ($^{13}C^{15}N_2$-penoxsulam) in methanol:water (30:70, v:v) containing 0.2% acetic acid is added; the mixture is sonicated and vortexed to ensure uniformity of the final sample.
Instrument/Detector	LC/MS/MS using a C8 column and a gradient mobile phase of acetonitrile:methanol (50:50, v:v) and 0.1% acetic acid in water. Penoxsulam is detected by its daughter ions using an electrospray interface in the positive ion mode. The parent and daughter ions monitored are: m/z 484 and 195 for penoxsulam m/z 484 and 164 for confirmation of penoxsulam and m/z 487 and 198 for $^{13}C^{15}N_2$-penoxsulam (internal standard).
Standardization method	A calibration curve (linear regression) of penoxsulam and $^{13}C^{15}N_2$-penoxsulam standards is generated, plotting the concentration of penoxsulam (x-axis) versus the respective quantitation ratio (peak area of penoxsulam ion ÷ penoxsulam internal standard isotope; y-axis). Isotopic overlap between the analyte and the internal standard is empirically determined by analyzing standard solutions of each compound; the correction factor is applied to the measured quantitation ratio if isotopic crossover is determined. Samples calculations were provided.
Stability of std solutions	Not addressed
Retention times	~3 minutes

B.2. Enforcement Method

A previously submitted LC/MS/MS Method GRM 01.25 was proposed as an enforcement method for residues of penoxsulam in association with a petition for use on rice (PP#3F6542; D288152, 8/11/04, W. Cutchin). That method has been submitted to ACB/BEAD for a regulatory method validation. The method (Method GRM 05.08) reviewed here is similar to the

earlier rice enforcement method (Method GRM 01.25), except that an isotopic internal standard is used in GRM 05.08. With the submission of an ILV (MRID 46703505), GRM 05.08 meets all of the scientific requirements as an enforcement method for bovine tissues and fish, so long as the sponsor agrees to supply both the standard and the isotopically labeled internal standard to the Ft. Meade Repository for as long as the method exists.

C. RESULTS AND DISCUSSION

C.1. Data-Gathering Method

The method validation recoveries of penoxsulam using Method GRM 05.08 were adequate from fortified samples of bovine milk, fat, kidney, liver, and muscle, and fish (edible tissue). The method validation data are presented in Table C.1.1. Recoveries of penoxsulam averaged 93 ± 1.7% in milk, 98 ± 3.1% in bovine fat, 96 ± 3.3% in bovine kidney, 96 ± 2.3% in bovine liver, 95 ± 1.9% in bovine muscle, and 95 ± 2.4% in fish (edible tissue) fortified with penoxsulam at 0.01 (LOQ) and 0.1 ppm.

The method characteristics for Method GRM 05.08 are presented in Table C.1.2. The method is adequately specific for penoxsulam. It monitors the precursor ion plus one structurally significant product ion, and for confirmation, a second transition of m/z 484/164 may also be monitored. Typical chromatograms for the confirmation of penoxsulam in bovine muscle were provided, and recovery data for the confirmatory ion transition with milk and fish were generated in association with the ILV study (see Section C.3).

In the goat metabolism study (45830713.der.wpd, 7/19/04, W. Cutchin), kidney and liver samples from goats orally dosed for 5 days with either triazolo-pyrimidine (TP) or phenyl (PH) radiolabeled penoxsulam (at 10.1 or 12.4 ppm in the diet, respectively), were extracted by refluxing for one hour with ACN:water (1:1, v:v). The parent was identified in both kidney and liver as the major residue, but was present only at low concentrations (<0.05 ppm). Because Method GRM 05.08 uses a similar extraction process (ACN:water, 8:2, v:v at 60° C for one hour) as that used in the metabolism study, and the parent was found in the goat metabolism study, albeit at low concentrations, no further radiovalidation data for the residue method is needed.

TABLE C.1.1. Recovery Results from Method Validation of Bovine Matrices and Fish Tissues using the Data-Gathering Analytical Method. [1]

Matrix	Spiking Level [2] (ppm)	Recoveries Obtained (%)	Mean Recovery ± SD [CV] [3] (%)
Bovine milk	0.003	93	Not applicable (NA)
	0.01	92, 92, 92, 93, 95, 96	93 ± 2.0 [2.1]
	0.1	91, 92, 92, 93, 94	92 ± 1.4 [1.5]
Bovine fat	0.003	90	NA
	0.01	94, 95, 96, 98, 98, 102	97 ± 2.9 [3.0]
	0.1	97, 97, 98, 101, 104	99 ± 3.1 [3.1]
Bovine kidney	0.003	93	NA
	0.01	92, 94, 96, 99, 100, 103	97 ± 4.0 [4.1]

TABLE C.1.1. Recovery Results from Method Validation of Bovine Matrices and Fish Tissues using the *Data-Gathering Analytical Method.* [1]

Matrix	Spiking Level [2] (ppm)	Recoveries Obtained (%)	Mean Recovery ± SD [CV] [3] (%)
	0.1	93, 94, 94, 97, 98	95 ± 2.0 [2.1]
Bovine liver	0.003	97	NA
	0.01	94, 96, 96, 98, 98, 99	97 ± 2.1 [2.2]
	0.1	92, 94, 97, 98, 98	96 ± 2.6 [2.7]
Bovine muscle	0.003	93	NA
	0.01	91, 94, 95, 96, 97, 97	95 ± 2.3 [2.4]
	0.1	92, 93, 94, 96, 96	94 ± 1.6 [1.7]
Fish muscle (edible tissue)	0.003	93	NA
	0.01	93, 94, 95, 95, 96, 100	95 ± 2.5 [2.6]
	0.1	90, 93, 94, 94, 96	94 ± 2.1 [2.2]

[1] Fortification standard solutions were prepared in ACN.
[2] Fortification levels at 0.003 ppm are representative of the LOD and have a lower degree of confidence than those at or above the LOQ (0.01 ppm).
[3] As reported by the petitioner.

TABLE C.1.2. Characteristics for the Data-Gathering Analytical Method Used for the Quantitation of Penoxsulam Residues in Bovine Matrices and Fish Tissues.

Analyte(s)	Penoxsulam
Equipment ID	Applied Biosystems MDS Sciex API 3000 LC/MS/MS System; Zorbax SB-C8 column (4.6 x 75 mm, 3.5-µm).
Limit of quantitation (LOQ)	*0.01 ppm in all bovine matrices and fish tissue* (validated LOQ) 0.00197-0.00395 ppm (LOQ calculated as 10x the standard deviation from the 0.01 ppm recovery results)
Limit of detection (LOD)	0.003 ppm in all bovine matrices and fish tissue (validated LOD) 0.00059-0.00119 ppm (LOD calculated as 3x the standard deviation from the *0.01* ppm recovery results)
Accuracy/Precision	Percent recoveries and CVs indicate acceptable accuracy/precision for residues of penoxsulam from bovine matrices and fish tissue at the LOQ and 10x LOQ. The recovery ranges were 91-96% (average = 93%; CV = 1.9%) for bovine milk, 94-104% (average = 98%; CV = 3.1%) for bovine fat, 92-103% (average = 96%; CV = 3.4%) for bovine kidney, 92-99% (average = 96%; CV = 2.4%) for bovine liver, 91-97% (average = 95%; CV = 2.1%) for bovine muscle, and 90-100% (average = 95%; CV = 2.6%) for fish. See Table C.1.1 above.
Reliability of the Method [ILV]	An ILV was conducted to verify the reliability of Method GRM 05.08 (using both C8 and C18 columns) for the determination of residues of penoxsulam in bovine milk and fish edible tissue with transition ions m/z 484/195 and 484/164 (for confirmation). The values obtained are indicative that the LC/MS/MS method is reliable. See Section C.3.
Linearity	The method/detector response was linear (coefficient of determination, r^2= 1.0000) within the range of 0.05-50 ng/mL.
Specificity	The control chromatograms generally have no peaks above the chromatographic background and the spiked sample chromatograms contain only the analyte peak of interest. Peaks were well defined and symmetrical. There appeared to be no carryover to the following chromatograms.

C.2. Enforcement Method

GRM 05.08 is also scientifically supportable as an enforcement method so long as the sponsor agrees to supply the isotopically labeled internal standard to the EPA Ft. Meade repository for as

long as the method exists. The registrant must also submit the raw data that support their calculated results.

C.3. Independent Laboratory Validation

An independent laboratory validation study was conducted for Method GRM 05.08 using samples of bovine milk and fish tissue (MRID 46703505). The ILV was conducted at PTRL Europe GmbH (Ulm, Germany). Samples of commercially obtained bovine whole milk and fish (pollack) edible tissue were fortified with a penoxsulam standard in ACN at 0.01 ppm (LOQ) and 0.10 ppm. Fortified and unfortified (control) samples were analyzed using Method GRM 05.08 as described in Table B.1.1. No one matrix appeared to be more difficult to analyze than another, therefore, fish was chosen as the representative matrix for a meat tissue and bovine milk was chosen "because it is of a highly aqueous nature."

Prior to initiation of the first ILV trial, the laboratory conducted preliminary studies necessary to establish acceptable performance of the chromatographic instrumentation being used (i.e., HPLC retention time and detector sensitivity). The ILV trial included quantitation of both ion transitions (484/195 m/z and 484/164 m/z) using two different HPLC systems (C8 or C18 column). The ILV was successful for both fish and milk with the first trial. The recoveries of penoxsulam from the ILV study are reported in Table C.3.1. Residues of penoxsulam were less than the LOQ (<0.01 ppm) in/on two unfortified samples of milk and fish.

The laboratory reported that a set of 26 samples required 8 person-hours or ~1 work day in the laboratory, with overnight (unattended) LC/MS/MS analysis, and ~2 hours for data evaluation. No communication between the ILV laboratory personnel with the method developers was necessary, and the ILV laboratory did not identify any critical steps or recommend any modifications to the method.

TABLE C.3.1. Recovery Results Obtained by an Independent Laboratory Validation of Method GRM 05.08 for the Determination of Penoxsulam in Bovine Milk and Fish.

Matrix	Spiking Level (ppm)	Transition Ions 484 →195 m/z		Transition Ions 484 →164 m/z	
		Recoveries Obtained %	Mean Recovery ± SD [CV]	Recoveries Obtained %	Mean Recovery ± SD [CV]
LC/MS/MS using a C8 column					
Bovine milk	0.01	96, 97, 97, 98, 103	98 ± 2.8 [2.8]	95, 96, 98, 98, 99	97 ± 1.6 [1.7]
	0.1	93, 94, 95, 100, 100	96 ± 3.4 [3.5]	91, 94, 96, 97, 97	95 ± 2.5 [2.7]
Fish	0.01	93, 96, 97, 100, 104	98 ± 4.2 [4.3]	95, 96, 97, 100, 103	98 ± 3.3 [3.3]
	0.1	93, 94, 94, 95, 98	95 ± 1.9 [2.0]	93, 94, 94, 95, 100	95 ± 2.8 [2.9]
LC/MS/MS using a C18 column					
Bovine milk	0.01	97, 97, 99, 100, 104	99 ± 2.9 [2.9]	93, 96, 101, 102, 103	99 ± 4.3 [4.3]
	0.1	92, 95, 97, 98, 99	96 ± 2.8 [2.9]	94, 94, 95, 95, 99	95 ± 2.1 [2.2]
Fish	0.01	98, 98, 98, 100, 108	100 ± 4.3 [4.3]	94, 101, 101, 103, 104	101 ± 3.9 [3.9]
	0.1	94, 96, 96, 97, 98	96 ± 1.5 [1.5]	93, 95, 97, 98, 100	97 ± 2.7 [2.8]

D. CONCLUSION

Adequate method validation data, including those provided by an independent laboratory, have been submitted to support Method GRM 05.08 for use on bovine and fish tissues, and milk. Except for use of an isotopic internal standard, Method GRM 05.08 is similar to Method GRM 01.25, the proposed enforcement method in an earlier rice petition (PP#3F6542; D288152, 8/11/04, W. Cutchin) which has been forwarded to ACB/BEAD for a regulatory method validation. No additional radiovalidation data is required for GRM 05.08. The data requirements to support an enforcement method have been scientifically fulfilled.

E. REFERENCES

DP Barcode: D288152
Subject: Penoxsulam. Petition for the Establishment of Permanent Tolerances for the Use on Rice. Summary of Analytical Chemistry and Residue Data. PP#3F6542
From: W. Cutchin
To: P. Errico/J. Miller
Date: 8/11/04
MRIDs: 45830712-17, 45830719-20, 46267601

F. DOCUMENT TRACKING

RDI: David Soderberg (29 Sept 2006); Name2 (Date); Richard Loranger (29 September 2006).
Petition Number(s): 5F7012
DP Barcode(s): D326985
PC Code: 119031

Template Version June 2005

Primary Evaluator _____ Date 28 Dec 2006:
David Soderberg, Chemist, RRB3, HED

Approved by _____ Date: 28 Dec 2006
Danette Drew, Senior scientist, RRB3, HED

This DER was originally prepared under contract by Dynamac Corporation (2275 Research Boulevard, Suite 300; Rockville, MD 20850; submitted 08/02/2006). The DER has been reviewed by the Health Effects Division (HED) and revised to reflect current Office of Pesticide Programs (OPP) policies.

STUDY REPORT:

46703506 Autderheide, J. (2005) Penoxsulam: Field Accumulation in Bluegill Sunfish (Lepomis macrochirus). Project Number: 48935. Unpublished study prepared by Analytical Bio-Chemistry Labs., Inc. 65 p.

EXECUTIVE SUMMARY:

Dow AgroSciences has submitted a study investigating the total radioactive residues (TRR) in bluegill sunfish muscle tissue after treatment of their water with radiolabeled penoxsulam; and the potential for bioaccumulation of TRR was calculated.. In addition, where there was adequate tissue available, an investigation was made to characterize or identify these radioactive residues.

Bluegill sunfish were exposed for 28 consecutive days to the radiolabeled penoxsulam, either [phenyl-U-^{14}C]penoxsulam or [het-2-^{14}C]penoxsulam, under static conditions at concentrations of 0.150 mg ai/L or 1.50 mg ai/L. The petitioner stated that the applied rates are equivalent to 1x and 10x the maximum expected environmental conditions. Separate aquatic chambers, containing ~150 fish at initiation, were designed for the two radiolabels, two concentration rates, and the control. The study was conducted by ABC Laboratories, Inc. (Columbia, MO).

The concentration of penoxsulam in the water of each test chamber was periodically verified during the study period. The water solutions were generally found to stay within an overall range of 73-96% of the intended treatment concentration.

Following exposure of fish at 1x, total radioactive residues (TRR) were below the minimum quantifiable limit (MQL; <7.10 to <7.45 ppb) in all fish tissue samples exposed to [phenyl-U-^{14}C]penoxsulam, and ranged from <MQL to 11.4 ppb in fish tissue samples exposed to [het-2-^{14}C]penoxsulam. Following exposure of fish at 10x, TRR ranged from 28.5 to 105 ppb in fish samples exposed to [phenyl-U-^{14}C]penoxsulam, and from <MQL to 42.6 ppb in fish samples exposed to [het-2-^{14}C]penoxsulam. TRR were highest in Day 1 fish treated with [phenyl-U-^{14}C]penoxsulam and in Day 21 fish treated with [het-2-^{14}C]penoxsulam. The TRR appears to plateau after approximately 7 days of exposure. It appears as though these reported TRR needed to be corrected for a relatively large background count.

Bioconcentration factors based upon the measured TRR (BCF; TRR in tissue/TRR in water) were calculated where possible; all calculated BCF values were ≤ 0.10, indicating that there is little potential for the test substance and/or its metabolites to bioaccumulate.

Selected edible fish tissues from the 1.5 mg ai/L (10x) rate treatments were subjected to further identification or characterization of the residues. However, only a summary of these results plus two chromatograms were provided. Raw data and information about the chromatographic conditions were not submitted.

In Day 7 fish samples exposed to [phenyl-U-^{14}C]penoxsulam, the following components were identified: penoxsulam (0.039 ppm), 5-hydroxy penoxsulam (0.016 ppm), penoxsulam sulfonamide (0.005 ppm), and penoxsulam BSTCA (0.003 ppm), plus non-discriminated residues (0.031 ppm) and bound residues (0.21 ppm). These identified/characterized residues add to 0.115 ppm while the original TRR for this sample was 0.066 ppm.

In Day 28 fish samples exposed to [het-2-^{14}C]penoxsulam, only penoxsulam (0.021 ppm) and 5-hydroxy penoxsulam (0.009 ppm) were identified. Non-discriminated residues were 0.40 ppm and bound residues were 0.023 ppm. These identified/characterized residues add to 0.093 ppm while the original TRR for this sample was 0.040 ppm.

The components for the day 7 ([phenyl-U-^{14}C]penoxsulam) fish thus totaled to 174% of the TRR reported for those fish; and the components for the day 28 ([het-2-^{14}C]penoxsulam) totaled to 235% of the TRR reported for those fish. The difference may simply be due to unexplained sample concentration, but without raw data or an explanation it is conceivable that the cause instead is overestimation of only some portion of the residues, compared to other portions. Therefore this study cannot be concluded to be scientifically acceptable until this discrepancy is resolved.

Information pertaining to sample storage conditions and intervals was minimal; no analysis dates were provided. The study submission reported the only exposure dates of the initial and second study. Based on the study completion date, fish samples may have been stored for up to 280 days (9.2 months) from collection to completion of residue characterization. No storage stability data were provided. The Agency has previously noted in PP#3F6542 (DP Barcode D288152, 8/11/04, W. Cutchin) that comparative analysis of goat milk and tissue extracts show that residue profiles are similar after 135 and 300 days of sample collection, therefore supporting storage stability data will not be required for the subject fish metabolism study. We note for future submissions that the petitioner should submit individual sample TRR and chromatographic analysis dates.

STUDY/WAIVER ACCEPTABILITY/DEFICIENCIES/CLARIFICATIONS:

Only data summaries were submitted for this study. The residue data cannot be considered to be scientifically acceptable until an explanation of the high recoveries found during residue identification characterization of fish tissues compared to TRRs originally determined for these tissues are explained and the difference supported by submission of appropriate data or

methodological information. In addition the petitioner must also provide information regarding the chromatographic system (i.e., instrumentation, columns, eluants, and detectors) and include the LOD/LOQ of the methodologies used for identification/characterization of the residues.

Although these data are not scientifically acceptable as submitted, they may be adequate to serve some regulatory purposes. The acceptability of this study for regulatory purposes is addressed in the forthcoming U.S. EPA Residue Chemistry Summary Document, DP Barcode D326985.

COMPLIANCE:

Signed and dated Good Laboratory Practice (GLP), Quality Assurance and Data Confidentiality statements were provided. No deviations from regulatory requirements were reported which would have an impact on the validity of the study.

A. BACKGROUND INFORMATION

Penoxsulam is a sulfonamide herbicide currently registered for use on rice for selective control of grasses, broadleaf, and sedge weeds. Dow AgroSciences is now proposing new aquatic uses for penoxsulam for the control aquatic plant pests such as hydrilla, water hyacinth, egeria, and various others in lakes, reservoirs, ponds, and canals. The mode of action at the cellular level involves the inhibition of acetolacetate synthase (ALS).

TABLE A.1. Test Compound Nomenclature.

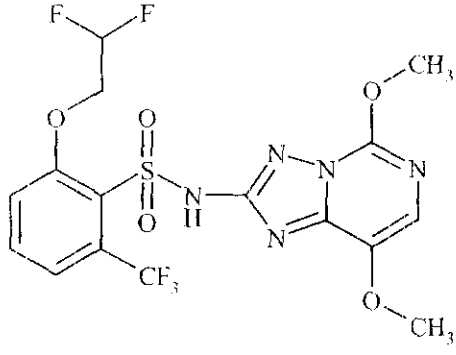
Compound	
Common name	Penoxsulam
Company experimental name	XDE-638
IUPAC name	6-(2,2-difluoroethoxy)-N-(5,8-dimethoxy-s-triazolo[1,5-c]pyrimidin-2-yl)-α,α,α-trifluoro-o-toluenesulfonamide
CAS name	2-(2,2-difluoroethoxy)-N-(5,8-dimethoxy[1,2,4]triazolo[1,5-c] pyrimidin-2-yl)-6-(trifluoromethyl) benzenesulfonamide
CAS registry number	219714-96-2
End-use product (EP)	GF-443 SC

TABLE A.2. Physicochemical Properties of the Technical Grade Test Compound: Penoxsulam.

Parameter	Value	Reference
Melting point/range	Not available	
pH	5.2	MRID 45830707

TABLE A.2. Physicochemical Properties of the Technical Grade Test Compound: Penoxsulam.			
Parameter	Value		Reference
Density	1.61 g/mL at 20 °C		MRID 45830707
Water solubility at 19 °C	Unbuffered pH 5 pH 7 pH 9	4.91 mg/L 5.66 mg/L 408 mg/L 1460 mg/L	MRID 45830720
Solvent solubility at 19 °C	Xylene 1-Octanol Methanol Ethyl acetate Acetonitrile Acetone Dimethylsulfoxide	0.017 g/L 0.035 g/L 1.48 g/L 3.23 g/L 15.3 g/L 20.3 g/L 78.4 g/L	MRID 45830720
Vapor pressure	7.16×10^{-16} mm Hg at 25 °C		MRID 45830720
Dissociation constant, pK_a	5.1 (ambient)		MRID 45830720
Octanol/water partition coefficient, $Log(K_{OW})$	Unbuffered pH 5 pH 7 pH 9	-0.354 1.137 -0.602 -1.418	MRID 45830720
UV/visible absorption spectrum	Not available		

B. EXPERIMENTAL DESIGN

B.1. Fish Metabolism

To investigate the nature of penoxsulam residues in freshwater predator fish, bluegill sunfish were exposed for 28 consecutive days to the radiolabeled test substance, [phenyl-U-^{14}C]penoxsulam or [het-2-^{14}C]penoxsulam, under static conditions at concentrations of 0.150 mg ai/L or 1.50 mg ai/L. The petitioner stated that the applied rates are equivalent to 1x and 10x the maximum expected environmental conditions (label rate). Five separate aquatic chambers, containing ~150 fish/chamber at initiation, were designed for each of the two radiolabels, both at the two concentration rates, and one chamber for the control. The study was conducted by ABC Laboratories, Inc. (Columbia, MO).

The radiolabeled test substance diluted in acetone was added to the aquatic chamber containing "laboratory freshwater" (well water with a hardness of 130-160 mg/L as $CaCO_3$) using procedures to minimize damage to the fish; acetone only was added to the control chamber water. The initial water level in each test vessel was marked and test water was periodically added to make up any water sampling or evaporative losses during the 28-day exposure period. The aquatic chambers were aerated regularly to maintain the dissolved oxygen at ≥65% saturation.

Due to excessive mortality during initial exposure, the 10x rate treatment with phenyl radiolabeled penoxsulam was repeated (second testing). The initial exposure mortality was not attributed to the exposure of the test substance, but was thought to be most likely an effect of a contaminant within that test vessel.

TABLE B.1.1.1. General Test Organism Information.					
Species	Scientific name	Age	Weight at study initiation (kg)	Health status	Description of housing/holding area
Bluegill sunfish	*Lepomis macrochirus*	Juvenile	Not specified (NS)	Healthy	Fish were housed in circular metal vessels, ~66 inch diameter by 24 inch deep, coated on the inside with fiberglass resin. Each test vessel contained ~440 L of laboratory freshwater, and was maintained in an open-air greenhouse with a roof.

TABLE B.1.1.2. Test Organism Dietary Regime.		
Diet	Acclimation period	Predosing
Fish were fed a commercial fish feed at least once daily. The study protocol stated that fish food may be supplemented with live brine shrimp nauplii (*Artemia* sp.) and/or other aquatic invertebrates.	Fish were maintained in the laboratory for a minimum of 14 days before starting the test. The test fish were gradually acclimated to the aquatic chamber at least 48 hours prior to testing.	None.

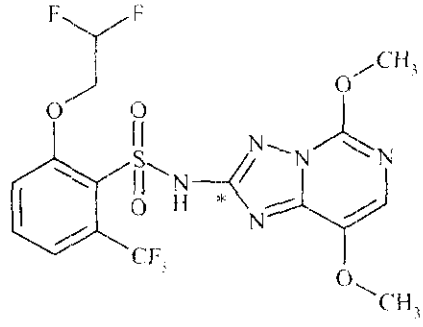
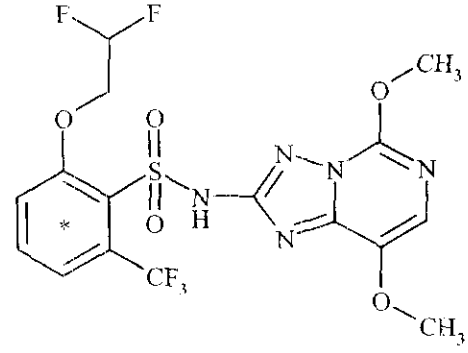
TABLE B.1.1.3. Test Organism Dosing Regime.			
Regime	Level of administered dose	Food consumption (kg/day)	Timing/Duration
Aquaculture	Test substances were administered once on Day 0 at 0.15 mg ai/L for the low rate and 1.5 mg ai/L for the high rate.	Not provided.	28 days

Environmental Conditions of the Test Vessels

During the 28 days of exposure, temperature, dissolved oxygen concentration, and pH of the water remained within acceptable limits. The measured temperatures ranged 9.3-29.9 °C, the pH ranged 6.5-9.5, and the measured dissolved oxygen concentration ranged 6.7-18.9 mg/L (77-212% of air saturation). The supersaturation of the water test systems with oxygen was attributed to an algal bloom which formed in each aquatic chamber after study initiation. The algae bloom may have increased the light attenuation and diminished possible photolysis of the test substance.

B.1.2. Test Materials

TABLE B.1.2.1. Test Material Characteristics.

Chemical structure		
Radiolabel position	Labeled on the 2-heterocyclic ring (Het-2) [1]	Uniformly labeled on the phenyl ring (PH)
Lot No.	Inv# 1861	Inv# 1573
Purity	>99%	97.5%
Specific activity	30.0 mCi/mmol	28.1 mCi/mmol

[1] We note that the petitioner did not provide a structure or sufficient description of the location of the radiolabel for the Het-2 test substance; the test substance was identified as N-[(1-(2,2-difluoroethoxy)-3-trifluoromethyl)-2-benzene)sulfonyl]-2-amino-5,8-dimethoxy-1,2,4-triazolo[1,5-c]pyrimidine-2-^{14}C. The radiolabel position indicated here is the same as the label position for the hetero-ring-labeled test substance used in a previously submitted rice metabolism study (refer to DER 45830712.der.wpd), which was described as the "2-triazolopyrimidine label." We have concluded that this location would be reasonable given the description provided in MRID 46703506.

B.1.3. Sampling Information

TABLE B.1.3.1. Sample Collection Information.

Samples Collected	Sampling Intervals
Water	Pretreatment and posttreatment (4 hours, 1 day, 3 days, 7 days, 14 days, 21 days, and 28 days)
Edible fish tissues	Pretreatment and posttreatment (4 hours, 1 day, 3 days, 7 days, 14 days, 21 days, and 28 days)

B.1.4. Analytical Methods for the Identification/Characterization of Residues

B.1.4.1. Sample Handling and Preparation

Water sampling: Duplicate water samples were collected from each exposure system for measurement of total radioactivity and calculation of the test substance concentration. Samples were collected prior to exposure, ~4 hours post-application, and then 1, 3, 7, 14, 21, and 28 days after application of the test substance. Single water samples from the control test vessel were collected and analyzed at days 0, 14, and 28 of the study. Water samples were taken from a point at least one foot away from the side and bottom of the test vessels. No information was provided regarding how the collected water samples were stored after sampling.

Fish sampling: Samples of fish (a minimum of four fish) from the exposure system were collected for analysis each time water samples were collected (prior to exposure, ~4 hours post-application, and then 1, 3, 7, 14, 21, and 28 days after application). Fish were divided into edible

(fillet) and non-edible portions (remainder of fish). Control samples were collected on days 0, 14, and 28. All collected tissue samples were homogenized cryogenically with dry ice and then immediately frozen at ≤ 20 °C or until analyzed.

The following procedures were employed to extract residues in fish tissue samples. A known amount of tissue was blended with acetonitrile:water (50:50, v:v), the blended mixture was heated in a sand bath at ~60 °C for one hour, and the extract mixture was centrifuged at 20 °C for 20 minutes. The supernatant was mixed with NaCl, shaken, and centrifuged. The tissue remaining after removal of the supernatant was re-extracted using the steps described above. The supernatant was then extracted with NaCl + saturated NaCl in water and centrifuged at 20 °C for 20 minutes. The liquid fractions from the various extraction steps were combined and centrifuged. The upper acetonitrile fraction was extracted once more with NaCl + saturated NaCl in water followed by centrifugation. The tissue remaining after removal of the supernatant was then extracted again with acetonitrile:0.01 N HCl (90:10, v:v), heated in a sand bath, and centrifuged. The acetonitrile, aqueous, and solid fractions were reserved for analysis.

B.1.4.2. Analytical Methodology

Water samples were directly assayed by LSC for total radioactivity. The MQL for LSC analysis of water was 2.8 ppb. Fish tissue samples were measured in triplicate for total radioactivity by combustion LSC. The MQL for LSC determination of fish tissues was <7.10 to <7.45 ppb.

The study submission states that residues were quantified and identified by LSC analysis of the collected UV detector fractions and comparison of the retention times with those of reference standards. Sample chromatograms were provided. The chemical names and structures of the reference standards are presented in Appendix I. However, although sample chromatograms were included, there was virtually no information explaining the chromatography used for separation and identification/characterization of the residues in fish edible tissue. This is a data deficiency. In addition, it appears from the submitted chromatograms that significant radiolabeled peaks were seen for [het-2-^{14}C]penoxsulam that went unidentified.

The petitioner should further describe the chromatographic system (i.e., instrumentation and detection), include the LOD/LOQ of the methodologies employed and explain if the unidentified peaks mentioned above were further characterized in any way.

B.2. Magnitude of the Residue

A separate field accumulation study with penoxsulam in freshwater calm and catfish was submitted in conjunction with DP Barcode D326985; refer to the 860.1400 DER for MRID 46703507.

C. RESULTS AND DISCUSSION

Information pertaining to sample storage conditions and intervals was minimal (Table C.2.) and no analysis dates were provided. The study submission did report the exposure dates of the

initial study (7/25/04-9/17/04) and for the second study (7/27/04-8/24/04). Representative chromatograms for analysis of fish tissue were dated 4/7 or 4/8/05, and the study completion date was reported as 5/3/05. Based on these dates, fish samples may have been stored for up to 280 days (9.2 months) from collection to completion of residue characterization. No storage stability data were provided. The Agency has previously noted in PP#3F6542 (DP Barcode D288152, 8/11/04, W. Cutchin) that comparative analysis of goat milk and tissue extracts show that residue profiles are similar after 135 and 300 days of sample collection, therefore additional supporting storage stability data will not be required for the subject fish metabolism study.

The concentrations of the test water solutions were periodically verified during the study period. The water solutions were generally within the applied rate for each radiolabel with an overall range of 73-96% of the nominal rate (Table C.2.1).

The TRR in edible tissues of bluegill sunfish exposed to the test system are presented in Table C.2.1. Following exposure of fish at 0.15 mg ai/L (1x), TRR were below the MQL (<7.10 to <7.45 ppb) in all fish samples exposed to [phenyl-U-^{14}C]penoxsulam, and ranged from <MQL to 11.4 ppb in fish samples exposed to [het-2-^{14}C]penoxsulam. Following exposure of fish at 1.5 mg ai/L (10x), TRR ranged from 28.5 to 105 ppb in fish samples exposed to [phenyl-U-^{14}C]penoxsulam, and from <MQL to 42.6 ppb in fish samples exposed to [het-2-^{14}C]penoxsulam. TRR were highest in Day 1 fish treated with [phenyl-U-^{14}C]penoxsulam and Day 21 fish treated with [het-2-^{14}C]penoxsulam. The TRR appears to plateau after approximately 7 days of exposure.

The bioconcentration factors (BCF; TRR in tissue/TRR in water) were calculated where possible, and ranged from, 0.0244-0.0833, 0.079-0.090, and 0.028-0.033 for the 10x phenyl label, 1x het-2 label, and 10x Het-2 label, respectively. All tissue concentrations in the 1x phenyl treatment were less than the MQL; therefore no BCF could be calculated (see Table C.2.1).

The results of characterization/identification of radioactive residues in the edible fish tissues are presented in Tables C.2.2 (phenyl-label) and C.2.3. (het-label); however, no raw data were submitted to support these reported results. In Day 7 fish samples exposed to [phenyl-U-^{14}C]penoxsulam, the following components were reported to have been identified: penoxsulam (34% TRR, 0.039 ppm), 5-hydroxy penoxsulam (14% TRR, 0.016 ppm), penoxsulam sulfonamide (4% TRR, 0.005 ppm), and penoxsulam BSTCA (3% TRR, 0.003 ppm). In Day 28 fish samples exposed to [het-2-^{14}C]penoxsulam, only penoxsulam (23% TRR, 0.021 ppm) and 5-hydroxy penoxsulam (9% TRR, 0.009 ppm) were identified. Bound residues were reported as 18% TRR (0.021 ppm) and 25% TRR (0.023 ppm) in fish treated with [phenyl-U-^{14}C]penoxsulam or [het-2-^{14}C]penoxsulam, respectively. The %TRR reported by the petitioner represents the percent of the total residues chromatographed and bound residues. Based on the TRR determined by combustion/LSC, accountabilites were 174-235%.

TABLE C.2. Summary of Concurrent Recoveries of Penoxsulam from Fish Edible Tissues.				
Matrix	Spike level (ppm)	Sample size (n)	Recoveries (%)	Mean ∀ std dev (%)

TABLE C.1. Summary of Concurrent Recoveries of Penoxsulam from Fish Edible Tissues.				
Matrix	Spike level (ppm)	Sample size (n)	Recoveries (%)	Mean ∀ std dev (%)
Not applicable				

TABLE C.2. Summary of Storage Conditions.			
Matrix	Storage Temperature (°C)	Estimated Storage Duration [1]	Interval of Demonstrated Storage Stability [2]
Water	Not provided	TRR dates not reported	None provided
Edible fish tissues	≤ 20 °C	≤280 days (9.2 months)	None provided

[1] Actual analysis dates were not provided; the storage duration for fish tissues is based on the study exposure and completion dates.

[2] The Agency has previously noted in PP#3F6542 (DP Barcode D288152, 8/11/04, W. Cutchin) that comparative analysis of goat milk and tissue extracts show that residue profiles are similar after 135 and 300 days of sample collection.

C.2. Identification, Characterization, and Distribution of Residues

Data in Table C.2.1. are presented as reported by the petitioner; no raw data were provided.

TABLE C.2.1. Total Radioactive Residues (Expressed as ppb) in Edible Tissues of Bluegill Sunfish and Test Solutions Exposure Study.

Sample Day	[phenyl-U-^{14}C]penoxsulam Applied at 0.15 mg ai/L (1x)			[phenyl-U-^{14}C]penoxsulam applied at 1.5 mg ai/L (10x)			[het-2-^{14}C]penoxsulam applied at 0.15 mg ai/L (1x)			a
	Edible tissue [1]	Water [1] (% nom)	BCF [2]	Edible tissue [1]	Water [1] (% nom)	BCF [2]	Edible tissue [1]	Water [1] (% nom)	BCF [2]	Edil tissu
Pre-trt	<MQL [3]	<MQL [3]	NA [4]	<MQL [3]	<MQL [3]	NA [4]	<MQL [3]	<MQL [3]	NA [4]	<MC
4 Hour	<MQL [3]	119 (79)	NA [4]	28.5	1,270 (84)	0.0244	<MQL [3]	131 (88)	NA [4]	<MC
1	<MQL [3]	120 (80)	NA [4]	105	1,260 (84)	0.0833	<MQL [3]	129 (86)	NA [4]	<MC
3	<MQL [3]	116 (77)	NA [4]	59.6	1,300 (86)	0.0458	<MQL [3]	127 (84)	NA [4]	<MC
7	<7.10 [3]	115 (77)	NA [4]	66.2	1, 220 (81)	0.0543	11.4	126 (84)	0.090	39
14	<7.10 [3]	110 (74)	NA [4]	61.7	1,260 (84)	0.0490	9.56	121 (81)	0.079	40
21	<7.45 [3]	110 (73)	NA [4]	65.4	1,210 (81)	0.0540	<7.74 [3]	117 (78)	NA [4]	42
28	<7.45 [3]	117 (78)	NA [4]	43.5	1,120 (74)	0.0388	<7.74 [3]	126 (84)	NA [4]	39

[1] TRR concentration was based upon the DPM value of the sample minus the background DPM values of the control matrix. The tissue and water values are expressed as parts per billion (ppb).

[2] The bioconcentration factor (BCF) was determined by dividing the tissue TRR concentration by the water TRR concentration.

[3] The minimum quantifiable limit (MQL) was 2.8 ppb for water samples and <7.10 to <7.45 ppb for fish tissues.

[4] NA = Not applicable. The values could not be determined from the data.

TABLE C.2.2. Characterization/Identification of Radioactive Residues in the Day 7 Edible Tissues of Bluegill Sunfish Exposed to [phenyl-U-^{14}C]penoxsulam at 1.5 mg/L (10x).		
Compound Name	% TRR [1]	Calculated Tissue Concentration (ppm)
Penoxsulam	34	0.039
5-Hydroxy penoxsulam	14	0.016
Penoxsulam sulfonamide	4	0.005
Penoxsulam BSTCA	3	0.003
Non-discriminated components [2]	27	0.031
Bound residues	18	0.021

[1] The %TRR reported by the petitioner is based on the total residues chromatographed and bound residues (0.115 ppm). Based on the TRR determined by combustion/LSC (66.2 ppb; Table C.2.1.), accountability was 174%.
[2] No individual components were greater than 0.010 ppm.

TABLE C.2.3. Characterization/Identification of Radioactive Residues in the Day 28 Edible Tissues of Bluegill Sunfish Exposed to [het-2-^{14}C]penoxsulam at 1.5 mg/L (10x).		
Compound Name	% TRR [1]	Calculated Tissue Concentration (ppm)
Penoxsulam	23	0.021
5-Hydroxy penoxsulam	9	0.009
Non-discriminated components [2]	43	0.040
Bound residues	25	0.023

[1] The %TRR reported by the petitioner is based on the total residues chromatographed and bound residues (0.093 ppm). Based on the TRR determined by combustion/LSC (39.6 ppb; Table C.2.1.), accountability was 235%.
[2] No individual components were greater than 0.010 ppm.

C.3. Proposed Metabolic Profile

No qualitative description of the metabolic pathway in bluegill sunfish was provided.

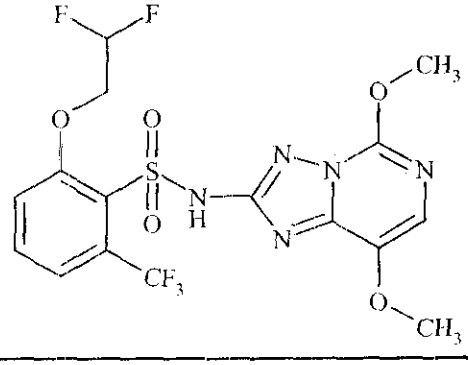
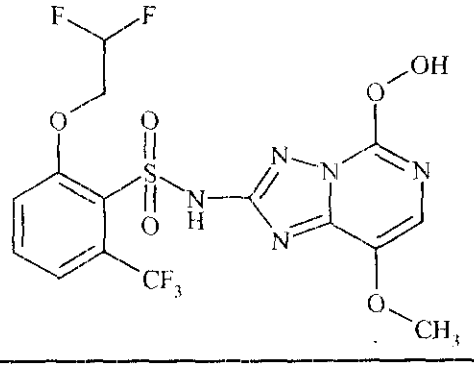
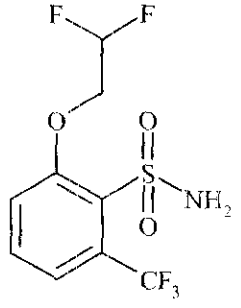
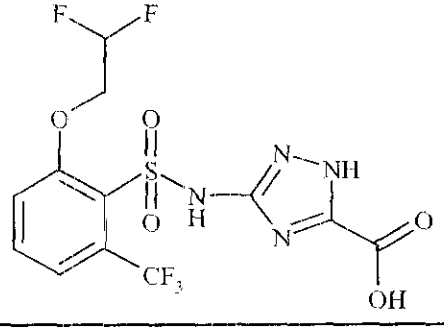
TABLE C.3.1. Identification of Compounds from Metabolism Study		
Common name (Code name)	Chemical name	Chemical structure
Penoxsulam (DE-638, XDE-638, XR-638, X638177)	CAS name: 2-(2,2-difluoroethoxy)-N-(5,8-dimethoxy[1,2,4]triazolo[1,5-c] pyrimidin-2-yl)-6-(trifluoromethyl) benzenesulfonamide	

TABLE C.3.1. Identification of Compounds from Metabolism Study		
Common name (Code name)	*Chemical name*	Chemical structure
5-Hydroxy penoxsulam (5-Hydroxy-XDE-638)	2-(2,2-difluoroethoxy)-N-(5,6-dihydro-8-methoxy-5-oxo[1,2,4]triazolo[1,5-c] pyrimidin-2-yl)-6-trifluoromethyl) benzenesulfonamide	
Penoxsulam sulfonamide (XDE-638 sulfonamide)	2-(2,2-difluoroethoxy)-6-(*trifluoromethyl*)benzenesulfonamide	
Penoxsulam BSTCA (*Triethylammonium* of XDE-638)	Triethylammonium 5-(2,(2,2-difluoroethoxy)-6-trifluoromethyl benzenesulfonyl amino)-1H-1,2,4-triazole-3,carboxylate	

D. CONCLUSION

The identified/characterized components of the TRR totaled to 174% to 235% of the TRR reported for the samefish samples. The difference may simply be due to unexplained sample concentration, but without raw data or an explanation it is conceivable that the cause instead is overestimation of only some portion of the residues, compared to other portions. Therefore this study cannot be concluded to be scientifically acceptable until this discrepancy is resolved. Additional information pertaining to the principle of the methods used for identification/ characterization of the residues in fish, and raw data regarding the quantitative assessment of metabolites identified must be submitted. Although incomplete, the results indicate that following a 28-day exposure of bluegill sunfish to [phenyl-U-^{14}C]penoxsulam or [het-2-^{14}C]penoxsulam at 0.15 mg ai/L (1x), TRR ranged from below the MQL (<7.45 ppb) to 11.4 ppb

in the edible fish tissues. All calculated bioconcentration factors were ≤0.10, indicating that there is little potential for the test substances or its metabolites to bioaccumulate.

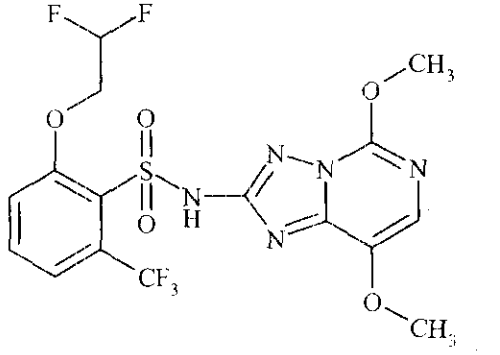
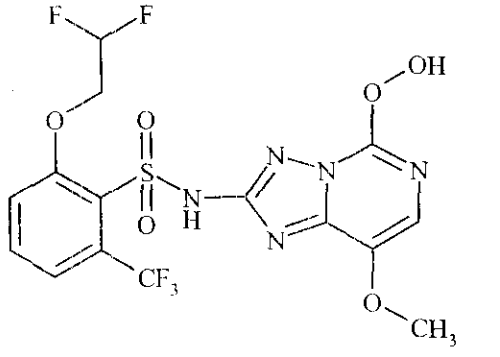
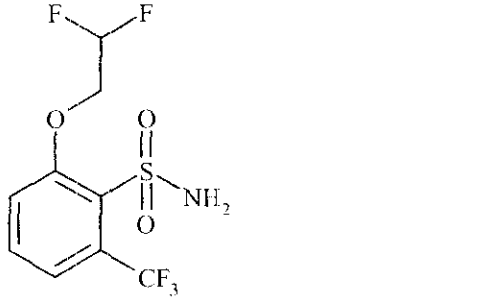
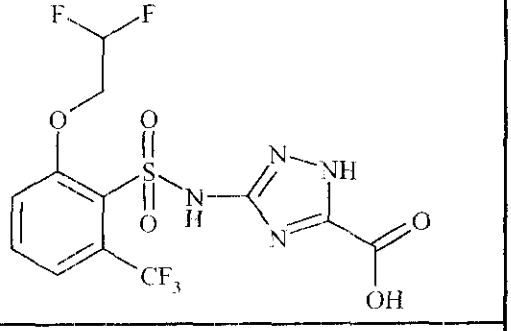
E. REFERENCES

DP Barcode: D288152
Subject: Penoxsulam. Petition for the Establishment of Permanent Tolerances for the Use on Rice. Summary of Analytical Chemistry and Residue Data. PP#3F6542
From: W. Cutchin
To: P. Errico/J. Miller
Date: 8/11/04
MRIDs: 45830712-45830717, 45830719-20, 46267601

F. DOCUMENT TRACKING

RDI: David Soderberg (28 December 2006); Danette Drew (28 December 2006).
Petition Number(s): 5F7012
DP Barcode(s): D326985
PC Code: 119031

Template Version June 2005

APPENDIX I. Chemical Names and Structures of Reference Standards Used in Fish Metabolism Study.		
Common name; (Company code)	Chemical name	Chemical structure
Penoxsulam (XDE-638; radiolabeled)	2-(2,2-difluoroethoxy)-N-(5,8-dimethoxy[1,2,4]triazolo[1,5-c] pyrimidin-2-yl)-6-(trifluoromethyl) benzenesulfonamide	
5-Hydroxy penoxsulam (5-Hydroxy-XDE-638)	2-(2,2-difluoroethoxy)-N-(5,6-dihydro-8-methoxy-5-oxo[1,2,4]triazolo[1,5-c] pyrimidin-2-yl)-6-trifluoromethyl) benzenesulfonamide	
Penoxsulam sulfonamide (XDE-638 sulfonamide)	2-(2,2-difluoroethoxy)-6-(trifluoromethyl)benzenesulfonamide	
Penoxsulam BSTCA (Triethylammonium of XDE-638)	Triethylammonium 5-(2,(2,2-difluoroethoxy)-6-trifluoromethyl benzenesulfonyl amino)-1H-1,2,4-triazole-3,carboxylate	
Penoxsulam TPSA (XDE-638 TPSA)	Not provided	Not provided

APPENDIX I. Chemical Names and Structures of Reference Standards Used in Fish Metabolism Study.		
Common name (Company code)	Chemical name	Chemical structure
5,8-dimethoxy penoxsulam (5,8-dimethoxy XDE-638)	Not provided	Not provided
2-amino-8-methoxy penoxsulam (2-amino-8-methoxy XDE-638)	Not provided	Not provided
Penoxsulam sulfonic acid (XDE-638 sulfonic acid)	Not provided	Not provided

Primary Evaluator: David Soderberg, Chemist, HED, RRB3 — Date: 28 Dec 2006

Approved by: Danette Drew, Senior scientist, HED, RRB3 — Date: 28 Dec 2006

This DER was originally prepared under contract by Dynamac Corporation (2275 Research Boulevard, Suite 300; Rockville, MD 20850; submitted 08/02/2006). The DER has been reviewed by the Health Effects Division (HED) and revised to reflect current Office of Pesticide Programs (OPP) policies.

STUDY REPORT:

46703507 Autderheide, J. (2005) Penoxsulam: Field Accumulation in Freshwater Clam and Catfish. Project Number: 49828. Unpublished study prepared by Analytical Bio-Chemistry Labs., Inc. 56 p.

EXECUTIVE SUMMARY:

Dow AgroSciences has submitted a study investigating the magnitude and potential for bioaccumulation of penoxsulam residues in freshwater clams and catfish. The test organisms were exposed for 28 consecutive days to penoxsulam under static aquatic conditions at concentrations of 0.150 mg ai/L and 1.50 mg ai/L. The petitioner stated that the applied rates are equivalent to 1x and 10x the maximum expected environmental conditions (proposed label rate). Separate aquatic chambers, initially containing ~60 fish and 300 clams each, were designed for the two concentration rates and control. Samples of the water solutions as well as clams and catfish were collected at regular intervals during the 28-day exposure period.

Edible tissues of fish and clams were analyzed for residues of penoxsulam using Dow AgroSciences method GRM 05.08 entitled "Determination of Residues of Penoxsulam in Bovine Tissues and Fish by High Performance Liquid Chromatography with Tandem Mass Spectrometry." This method is adequate for data collection based on validation data reported in the 860.1340 DER for MRIDs 46703504 and 46703505. Water was analyzed using a modification of this method. The reported minimum quantifiable limits (MQLs) were 3.99 ppb for the clam and fish tissue samples, and 45 ppb for the water samples.

Only summaries of the results were submitted. Concurrent recoveries and similar raw data and quality assurance information were not provided. This information must be submitted.

No supporting storage stability data were submitted. Actual study analysis dates were not provided but samples could not have been stored for a very long period of time (<49 days) prior to residue analysis. The study began on 26 August 2005 and the final report was completed on 14 October 2005. Given that no storage stability is required for samples held less than 30 days prior to analysis there is only a marginal reason to require any storage stability data. Available

storage stability data from an animal metabolism study discussed in PP#3F6542 (DP Barcode D288152, 8/11/04, W. Cutchin) show similar residue profiles in goat tissue extracts before and after 300 days of storage, and provides further evidence that for this study storage stability is not an issue.

The concentrations of the test water solutions were periodically verified during the study. The water solutions were generally within the applied rate with an overall range of 68-115% except those samples collected from the 4 hour post application time period (163% in the 1x rate study). Such results from this early time period may indicate that the test substance might not have been completely mixed in the test system yet.

Residues of penoxsulam ranged from below the MQL (<3.99 ppb) to 18.3 ppb in clams, and <MQL to 4.16 ppb in catfish following exposure at 0.15 mg ai/L (putatively 1x). Following exposure at 1.5 mg ai/L (putatively 10x), residues ranged 6.16–141 ppb in clams and <MQL–56.3 ppb in catfish. Residues were highest in clams exposed for 21/25 days, and in catfish (10X only) exposed for 14 days.

The bioconcentration factors (BCF; concentration in tissue/concentration in water) were calculated where possible. The calculated clam BCF values ranged 0.033-0.15 and 0.0041-0.098 for the 1x and 10x treatment levels, respectively. Except for the 4-hour post-application sample with a calculated BCF value of 0.017, all other catfish tissue concentrations from the 1x treatment were less than the MQL; therefore, no BCF could be calculated. The calculated catfish BCF values ranged 0.0029-0.033 for the 10x treatment level.

STUDY/WAIVER ACCEPTABILITY/DEFICIENCIES/CLARIFICATIONS:

Based upon the information that was submitted this study, it appears that this method should be scientifically acceptable. However, only a summary of analytical results was provided. No concurrent recoveries or other aspects of the analytical raw data were submitted. Without this information it is not possible to conclude that the analytical portion of this study is scientifically acceptable. The analytical raw data, including concurrent recoveries, must be submitted. Because the "in life" portion of the study was performed properly HED will also assume that the full study is acceptable pending submission of that data. The acceptability of this study for regulatory purposes is addressed in the forthcoming U.S. EPA Residue Chemistry Summary Document. DP Barcode D326985.

COMPLIANCE:

Signed and dated Good Laboratory Practice (GLP), Quality Assurance and Data Confidentiality statements were provided. No deviations from regulatory requirements were reported which would have an impact on the validity of the study.

A. BACKGROUND INFORMATION

Penoxsulam is a sulfonamide herbicide currently registered for use on rice for selective control of grasses, broadleaf, and sedge weeds. Dow AgroSciences is now proposing new aquatic uses for penoxsulam for the control aquatic plant pests such as hydrilla, water hyacinth, egeria, and various others in lakes, reservoirs, ponds, and canals. The mode of action at the cellular level involves the inhibition of acetolacetate synthase (ALS).

TABLE A.1. Test Compound Nomenclature.

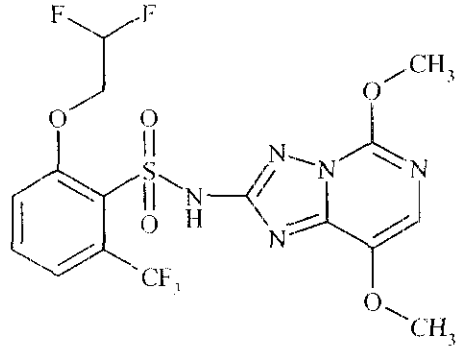
Compound	
Common name	Penoxsulam
Company experimental name	XDE-638
IUPAC name	6-(2,2-difluoroethoxy)-N-(5,8-dimethoxy-s-triazolo[1,5-c]pyrimidin-2-yl)-α,α,α-trifluoro-o-toluenesulfonamide
CAS name	2-(2,2-difluoroethoxy)-N-(5,8-dimethoxy[1,2,4]triazolo[1,5-c] pyrimidin-2-yl)-6-(trifluoromethyl) benzenesulfonamide
CAS registry number	219714-96-2
End-use product (EP)	GF-443 SC

TABLE A.2. Physicochemical Properties of the Technical Grade Test Compound: Penoxsulam.

Parameter	Value	Reference
Melting point/range	Not available	
pH	5.2	MRID 45830707
Density	1.61 g/mL at 20 °C	MRID 45830707
Water solubility at 19 °C	*Unbuffered* 4.91 mg/L pH 5 5.66 mg/L pH 7 408 mg/L pH 9 1460 mg/L	MRID 45830720
Solvent solubility at 19 °C	Xylene 0.017 g/L 1-Octanol 0.035 g/L Methanol 1.48 g/L Ethyl acetate 3.23 g/L Acetonitrile 15.3 g/L Acetone 20.3 g/L Dimethylsulfoxide 78.4 g/L	MRID 45830720
Vapor pressure	7.16×10^{-16} mm Hg at 25 °C	MRID 45830720
Dissociation constant, pK_a	5.1 (ambient)	MRID 45830720
Octanol/water partition coefficient, $Log(K_{OW})$	*Unbuffered* -0.354 pH 5 1.137 pH 7 -0.602 pH 9 -1.418	MRID 45830720
UV/visible absorption spectrum	Not available	

B. EXPERIMENTAL DESIGN

B.1. Fish Metabolism

A separate study investigating the nature of the residue and potential for bioaccumulation of penoxsulam in bluegill sunfish was submitted in conjunction with DP Barcode D326985; refer to MRID 46703506.

B.2. Magnitude of the Residue

To investigate the magnitude of penoxsulam residues in freshwater shellfish (mollusc) and bottom dwelling fish, freshwater clams and catfish were exposed for 28 consecutive days to penoxsulam under static aquatic conditions at concentrations of 0.150 mg ai/L and 1.50 mg ai/L. The petitioner stated that the applied rates are equivalent to 1x and 10x the maximum expected environmental conditions (label rate). Separate aquatic chambers, containing ~60 fish and 300 clams each at initiation, were designed for the two concentration rates and control. The study was conducted by ABC Laboratories, Inc. (Columbia, MO).

Penoxsulam standard diluted in acetone was added to the aquatic chamber containing approximately 440 L of "laboratory freshwater" using procedures to minimize damage to the fish; acetone without standard was also added to the water in the control chamber. The initial water level in each test vessel was marked and test water was periodically added to make up any water sampling or evaporative losses during the 28-day exposure period. These aquatic chambers were aerated regularly to maintain the dissolved oxygen at ≥65% saturation.

During initial exposure (August 11 - 17) there was excessive mortality of the clams, so the study was terminated and restarted on 26 August 2005 with a new batch of clams and catfish. The early mortality was not attributed to the exposure of the test substance, but was thought to be most likely due to a contaminant. Clams from the initial study were from Osage Catfisheries and likely collected from coldwaters, but the test vessels were maintained at elevated temperatures (>24 °C) and this may have affected the clam survival rate. Clams for the second study were collected from the USGS Complex.

B.2.1. Study Site Information/Test Organisms

TABLE B.2.1.1. General Test Organism Information

Species	Scientific Name	Age	Holding and Acclimation Information
Catfish	*Ictalurus punctatus*	Juvenile	Catfish were obtained from Osage Catfisheries (Osage, MO) and acclimated to the test conditions for a minimum of 48 hours. The fish were netted, and added to polyethylene bags which were floated in the outdoor tanks to allow the temperatures to equilibrate before the fish were carefully released into the tanks. Approximately 60 fish were added to each control and treatment vessel. During the holding and acclimation, the fish were fed a commercial fish feed

TABLE B.2.1.1. General Test Organism Information			
Species	Scientific Name	Age	Holding and Acclimation Information
Freshwater clams	*Corbicula fluminea*	Not specified	Freshwater clams (second study) were obtained from the USGS Complex (Columbia, MO). The clams were impartially distributed evenly between three stainless steel wire baskets within each test vessel. The baskets were evenly spaced within the tank to minimize personal bias. The clams were acclimated to the test conditions for ~48 hours prior to the chemical application. During the holding and acclimation, the clams were fed a commercial shellfish diet at least once daily.

TABLE B.2.1.2 Water Characterization.					
Study site	Water characteristics				
	Type	Hardness/Salinity	Temp. (°C)	pH	Dissolved O_2
Aquatic vessels were maintained in an open-air greenhouse with a roof at ABC Laboratories (Columbia, MO).	"laboratory freshwater"	well water with a hardness of 130-160 mg/L as $CaCO_3$	18.4-26.9	7.87-9.16	5.47-15.8 mg/L (82-195% of air saturation)

Measured concentrations of dissolved oxygen ranged up to 195% of air saturation. This supersaturation of oxygen in the water was attributed to an algal bloom which formed in each aquatic chamber 3 days after study initiation and continued for the duration of the study. The algae bloom may have increased the light attenuation and diminished possible photolysis of the test substance.

B.2.2. Sample Handling and Preparation

Water sampling: Single test solution samples were collected from each exposure system for the measurement of the test substance concentration. Samples were collected prior to exposure, ~4 hours post-application, and then 1, 3, 7, 14, 21, and 28 days after application of the test substance. Single water samples from the control test vessel were collected and analyzed at days 0, 14, and 28 of the study. Water samples were taken from a point at least one foot away from the side and bottom of the test vessels. Water samples were stored in amber glass bottles and frozen prior to analysis.

Fish and clam sampling: Samples of fish (a minimum of four) and clams (~25 g) from the exposure system were collected for analysis each time water samples were collected (prior to exposure, ~4 hours post-application and 1, 3, 7, 14, 21, and 28 days after application). In addition, clams were collected from the control and both treatment levels on day 25, and from the control and the 1.5 mg ai/L (10X) treatment on day 26. On day 28 clams could not be collected from the control and the 1.5 mg ai/L (10X) treatment tanks because there were no more surviving clams.

Fish were divided into edible (fillet) and non-edible portions (remainder of fish). The clams were shucked, and the tissue was separated from the shell. The number of clams collected at each time point was selected to produce a target of 25 g of edible tissue. Towards the end of the exposure, there was a die off of clams in all treatment groups, so that fewer clams were collected at each subsequent sampling period in order to have sufficient numbers of samples to determine

the uptake curves as well as sufficient tissue mass in each sample for residue analysis. Control samples were collected from each sampling event, but were only analyzed on days 0, 14, and 28 of the study for fish tissues, and 0, 14, 25, 26, and 28 for clam tissues. All fish and clam samples were immediately frozen in liquid nitrogen and maintained frozen until processing or analysis.

B.2.3. Analytical Methodology

Water (test solutions) and edible fish and clam samples were analyzed for residues of penoxsulam using methodology based on Dow AgroSciences GRM 05.08 entitled "Determination of Residues of Penoxsulam in Bovine Tissues and Fish by High Performance Liquid Chromatography with Tandem Mass Spectrometry." Detailed descriptions of the method along with independent laboratory validation data are reported in MRIDs 46703504 and 46703505.

Briefly, residues in tissue samples were extracted with acetonitrile (ACN)/water and held at 60 °C for one hour; water samples were simply mixed with ACN. An aliquot of the water or tissue extract was diluted with 0.1 N HCl for cleanup with a solid phase extraction (SPE) cartridge; residues were eluted with ACN. The eluate was diluted with methanol/water/acetic acid prior to LC/MS/MS analysis. Based on the brief method description in the clam/catfish study, it appears that an internal standard was not used for quantitation, and the reported minimum quantifiable limits (MQLs) were 3.99 ppb for the tissue samples and 45 ppb for the water samples. Although the study protocol required that the validity of the method be demonstrated by the analysis of concurrent recovery samples, no concurrent recoveries or other raw data were reported in conjunction with the exposure samples.

C. RESULTS AND DISCUSSION

Information pertaining to sample storage conditions and intervals was minimal (Table C.2.) and no analysis dates were provided. The study submission reported the exposure dates (8/26/05-9/23/05; second study), and the study completion date was 10/14/05. Based on these dates, clam/catfish samples may have been stored for up to 49 days (1.6 months) from collection to analysis. No storage stability data were provided. The Agency will not request storage stability data to support this study because samples were stored for an interval that might have been less than 30 days and at worst was not much greater than 30 days. Supporting this decision, the Agency has also previously noted, in PP#3F6542 (DP Barcode D288152, 8/11/04, W. Cutchin), that comparative analysis of goat milk and tissue extracts show that residue profiles are similar before and after 135 and 300 days of sample collection.

Water and edible tissues of fish and clams were analyzed for residues of penoxsulam using methodology based on Dow AgroSciences GRM 05.08, but no concurrent method validation data were reported. However, this method is adequate for data collection based on validation data reported in MRIDs 46703504 and 46703505. (LLMV = 0.01 ppm) (Note, however, that some residue levels found in fish samples from the higher rate exposure are not covered by the available method validation data.) The reported MQLs were 3.99 ppb for the clam and fish

tissue samples, and 45 ppb for the water samples. Residue data for the control samples were not reported and raw data were not provided.

The concentrations of the test water solutions were periodically verified during the study period. The water solutions were generally within the applied rate with an overall range of 68-115% of the nominal rate (Table C.4.1) except those samples collected from the 4 hour post application time period (163% in the 1x rate study); data from this time period may indicate that the test substance might not have been completely mixed in the test system by that time.

The concentrations of penoxsulam in edible clams and tissues of catfish exposed to penoxsulam over 28 days are presented in Table C.4.1. A summary of the residues in clams and catfish over the 28-day study duration is presented in Table C.4.2. Following exposure at 0.15 mg ai/L, residues were below the MQL (<3.99) to 18.3 ppb in clams, and <MQL to 4.16 ppb in fish tissues. Following exposure at 1.5 mg ai/L, residues ranged 6.16-141 ppb in clams and <MQL-56.3 ppb in fish tissues. Residues were highest in clams exposed for 21/25 days, and in catfish exposed for 14 days (10x).

The bioconcentration factors (BCF; concentration in tissue/concentration in water) were calculated where possible (see Table C.4.1). The calculated clam BCF values ranged 0.033-0.15 and 0.0041-0.098 for the 1x and 10x treatment levels, respectively. Except for the 4-hour post-application sample with a calculated BCF value of 0.017, all other catfish tissue concentrations from the 1x treatment were less than the MQL; therefore, no BCF could be calculated. The calculated catfish BCF values ranged 0.0029-0.033 for the 10x treatment level.

TABLE C.1. Summary of Concurrent Recoveries of Penoxsulam from Clam and Fish Edible Tissues.

Matrix	Spike level (ppm)	Sample size (n)	Recoveries (%)	Mean ± std dev (%)
None reported [1]				

[1] Method validation and independent laboratory validation data with fish fortified at 0.01 and 0.1 ppm were submitted in conjunction with the method description (860.1340 DER for MRIDs 46703504-05).

TABLE C.2. Summary of Storage Conditions.

Matrix	Storage Temperature (°C)	Estimated Storage Duration [1]	Interval of Demonstrated Storage Stability [2]
Water	frozen	≤49 days (1.6 months)	None provided
Edible clam fish tissues			None provided

[1] Actual analysis dates were not provided; the storage duration is based on the study exposure and completion dates.
[2] The Agency has previously noted in PP#3F6542 (DP Barcode D288152, 8/11/04, W. Cutchin) that comparative analysis of goat milk and tissue extracts show that residue profiles are similar after 135 and 300 days of sample collection.

C.2. Identification, Characterization, and Distribution of Residues

C.3. Proposed Metabolic Profile

The nature of the residue was determined in Bluefish and reported in MRID 46703606. In that study, on day 7 of exposure penoxsulam, per se, was 34% of the TRR, and the hydroxy metabolite 5-OH XDE638 constituted 14% of the TRR based upon phenyl labeled penoxsulam.

On day 28 parent penoxsulam constituted 23% of the TRR and 5-OH-XDE638 was 9% of the TRR based upon based upon triazine labed penoxsulam.

C.4. Residue Trials (Exposure Studies)

Data in Table C.4.1. are presented as reported by the petitioner; no raw data were provided.

TABLE C.4.1. Concentrations of Penoxsulam Residues (Expressed as ppb) in Edible Tissues of Freshwater Clams, Catfish, and Water Test *Solutions During the 28-Day Exposure Period.*

Sample Day	Clams exposed at 0.15 mg ai/L (1x)			Clams exposed at 1.5 mg ai/L (10x)			Catfish exposed at 0.15 mg ai/L (1x)			Catfish exposed at 1.5 mg ai/L (10x)		
	Edible tissue [1]	Water [1] (% nom)	BCF [2]	Edible tissue [1]	Water [1] (% nom)	BCF [2]	Edible tissue [1]	Water [1] (% nom)	BCF [2]	Edible tissue [1]	Water [1] (% nom)	BCF [2]
4 Hr	8.04	244 (163)	0.033	62.0	1,520 (101)	0.041	4.16	244 (163)	0.017	32.9	1,520 (101)	0.022
1	<MQL	139 (93)	NA [3]	6.62	1,570 (105)	0.0042	<MQL	139 (93)	NA [3]	4.53	1,570 (105)	0.0029
3	<MQL	136 (91)	NA [3]	6.16	1,490 (99)	0.0041	<MQL	136 (91)	NA [3]	4.64	1,490 (99)	0.0031
7	6.58	148 (99)	0.044	29.2	1,730 (115)	0.017	<MQL	148 (99)	NA [3]	18.2	1,730 (115)	0.011
14	13.7	155 (103)	0.088	70.1	1,710 (114)	0.041	<MQL	155 (103)	NA [3]	56.3	1,710 (114)	0.033
21	15.2	102 (68)	0.15	141	1,470 (98)	0.096	<MQL	102 (68)	NA [3]	<MQL	1,470 (98)	NA [3]
25	18.3	132 (88)	0.14	137	1,400 (93)	0.098	No sample	132 (88)	NA [3]	No sample	1,400 (93)	NA [3]
26	No sample	No sample	NA [3]	127	1,430 (95)	0.089	No sample	No sample	NA [3]	No sample	1,430 (95)	NA [3]
28	11.9	132 (88)	0.090	No sample	1,360 (91)	NA [3]	<MQL	132 (88)	NA [3]	16.0	1,360 (91)	0.012

[1] Residues are reported as parts per billion (ppb). The MQLs were 3.99 ppb for the clam/fish tissue samples and 45 ppb for the water samples.
[2] The bioconcentration factor (BCF) was determined by dividing the tissue concentration by the water concentration.
[3] NA = Not applicable. The values could not be determined from the data.

TABLE C.4.2. Summary of Residue Data from Fish Exposure Study with Penoxsulam.

Commodity/ Matrix	Total Applic. Rate (lb ai/A)	PHI (days)	Residue Levels (ppb)						
			n	Min.	Max.	HAFT[1]	Median (STMdR)	Mean (STMR)	Std. Dev.
Freshwater clams	0.15 mg ai/L	Study duration (0-28 days)	8	<3.99	18.3	NA	9.97	10.21	5.35
Catfish			7	<3.99	4.16	NA	3.99	4.01	0.06
Freshwater clams	1.5 mg ai/L	Study duration (0-28 days)	8	6.16	141	NA	66.1	72.4	56.8
Catfish			7	<3.99	56.3	NA	16.0	19.5	19.3

[1] HAFT = Highest average field trial value. Not applicable (NA).

D. CONCLUSION

Pending submission of supporting data as described above, HED will assume that this study is scientifically valid. The results indicate that following a 28-day exposure of freshwater clams and catfish to penoxsulam under a static aquatic conditions at 0.15 mg ai/L (1x), penoxsulam residues, per se, ranged from below the MQL (<3.99 ppb) to 18.3 ppb in clams, and <MQL to 4.16 ppb in catfish tissues. All calculated bioconcentration factors were ≤0.15, indicating that there is little potential for penoxsulam, per se, to bioaccumulate in the tested organisms.

E. REFERENCES

DP Barcode: D288152
Subject: Penoxsulam. Petition for the Establishment of Permanent Tolerances for the Use on Rice. Summary of Analytical Chemistry and Residue Data. PP#3F6542
From: W. Cutchin
To: P. Errico/J. Miller
Date: 8/11/04
MRIDs: 45830712-45830717, 45830719-20, 46267601

F. DOCUMENT TRACKING

RDI: D. Soderberg (28 Dec 2006); D. Drew (28 Dec 2006)
Petition Number(s): 5F7012
DP Barcode: D326985
PC Code: 119031

Template Version June 2005

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Chemical: Penoxsulam

PC Code:
119031
HED File Code: 11500 Petition Files Chemistry
Memo Date: 1/30/2007
File ID: DPD326985
Accession #: 000-00-0117

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