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Orthosulfamuron/IR5878/PC Code 108209/Isagro S.p.A.
DACO 7.2.1, 7.2.2, and 7.2.3 /OPPTS 860.1340/OECD IIA 4.2.5, 4.26, and 4.3
Residue Analytical Method

Primary Evaluator Douglas Dotson, Chemist, RAB2 D. Dotson Date: 2/14/2007

Peer Reviewer Dennis McNeilly, Chemist, RAB2 Date: 2/14/2007

This DER was originally prepared under contract by Dynamac Corporation (2275 Research Boulevard, Suite 300; Rockville, MD 20850; submitted 06/12/2006). The DER has been reviewed by the Health Effects Division (HED) and revised to reflect current Office of Pesticide Programs (OPP) policies.

STUDY REPORT:

46578969 Anspach, T. (2005) Enforcement Multiresidue Method (Including Validation) for the Determination of Residues of IR5878 in Matrix with Low Water Content (Rice Grain): Final Report. Project Number: ISA-0501V, AZ.G04-0112, A2/04/02/13. Unpublished study prepared by Dr. Specht and Partner. 45 p.

EXECUTIVE SUMMARY:

Isagro S.p.A. has submitted validation data for a QuEChERS multiresidue method for the determination of orthosulfamuron residues in/on rice grain (matrix with low water content). The QuEChERS multiresidue method used in the validation study is based on:

M. Anastassaides, S.J. Lehotay, D. Stajnbaher, and F.J. Schenck (2003). "Fast and easy multiresidue method employing acetonitrile extraction/partitioning and "dispersive solid-phase extraction" for the determination of pesticide residues in produce." Journal of AOAC International 86(2): 412-431.

The petitioner stated that the study was conducted according to the guidance documents of: (i) SANCO/825/00 (rev. 7 of March 17, 2004) and SANCO3029/99 (rev. 4 of July 11, 2000) of the European Commission; (ii) BBA Guideline: Residue Analytical methods for Post-registration Control Purposes of July 21, 1983; and (iii) U.S. EPA Residue Chemistry Test Guidelines, OPPTS 860.1340.

Briefly, the QuEChERS method uses a single-step buffered ACN extraction and salting out liquid-liquid partitioning from the water in the sample with magnesium sulfate and sodium acetate. The sample is cleaned up using dispersive solid phase extraction (SPE) to remove organic acids, excess water, and other components with primary secondary amine (PSA) and magnesium sulfate. An aliquot of the supernatant is diluted with 0.1% formic acid for HPLC/MS/MS analysis. The limit of quantitation (LOQ) is 0.01 ppm. Adequate recoveries ranging from 70% to 105% were obtained for rice grain samples fortified with orthosulfamuron at the LOQ (0.01 ppm) and 10x the LOQ.

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STUDY/WAIVER ACCEPTABILITY/DEFICIENCIES/CLARIFICATIONS:

Under the conditions and parameters used in the study, analytical method test data are classified as scientifically acceptable. However, the QuEChERS method does not conform to the FDA multiresidue methods data requirements under OPPTS 860.1360. To fulfill these data requirements, the petitioner needs to follow specific directions for each multiresidue method used by FDA published in that Agency's Pesticide Analytical Manual, Vol. I (PAM Vol. I) and provide recovery data for orthosulfamuron through these methods. The acceptability of this study for regulatory purposes is addressed in the U.S. EPA Residue Chemistry Summary Document, D332290, D. Dotson, 2/14/2007.

COMPLIANCE:

Signed and dated Good Laboratory Practice (GLP), Quality Assurance and Data Confidentiality statements were provided. The petitioner stated that the study was conducted in accordance with GLP standards as defined in 40 CFR 160, and is at variance with GLP standards as defined by the US EPA in the following manner: (a) The signed GLP-compliance statement by the Study Director does not conform with requirements of US EPA; and (b) GLP requirements specified in §19a para 1 Chemical Law (ChemG) are not necessary for analytical method development. Nevertheless the study was conducted according to the procedures described in this report and is in compliance with the requirement of GLP as set forth in the following: OECD Principles of Good Laboratory Practice (as revised in 1997) OECD Document; ENV/MC/CHEM(98)17, Paris, France, 1998, and EPA/FIFRA Good Laboratory Practice Standards, 40 CFR 160, FR 54, No. 158, August 17, 1989.

A. BACKGROUND INFORMATION

Orthosulfamuron is a postemergence herbicide that Isagro S.p.A. is proposing for use on rice grown in the United States for the control of annual and perennial broadleaf weeds, sedges, and barnyard grass. Orthosulfamuron belongs to the sulfamoylurea class of herbicides. It reportedly acts by inhibiting the plant enzyme acetolactate synthase which is active in the biosynthesis of valine, leucine, and isoleucine.

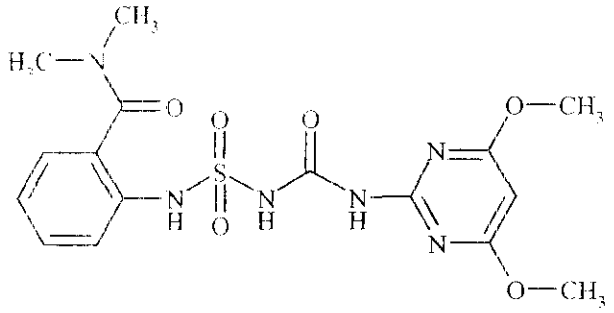
TABLE A.1. Test Compound Nomenclature.	
Compound	
Common name	Orthosulfamuron
Company experimental name	IR5878
IUPAC name	1-(4,6-dimethoxypyrimidin-2-yl)-3-[2-(dimethylcarbamoyl)phenylsulfamoyl]urea
CAS name	2-[[[[[(4,6-dimethoxy-2-pyrimidinyl)-amino]carbonyl]amino]sulfonyl]amino]-N,N-dimethylbenzamide
CAS registry number	213464-77-8
End-use product (EP)	0.51% G formulation (IR5878 0.5 GR; EPA Co. No. 80289) 51.5% WG formulation (IR5878 50 WG; EPA Co. No. 80289)

Table 2. Physicochemical Properties of the Technical Grade of Orthosulfamuron.		
Parameter	Value	Reference (MRID)
Color	White	46219004
Physical State	Fine Powder at 20°C	46219005
Odor	Odorless	46219006
pH	4.35 at 25°C (1% aqueous dispersion)	46219013
Density	1.45 g/mL at 20°C	46219008
Water solubility at 20°C	pH 4 buffer: 0.062 g/L pH 7 buffer: 0.63 g/L pH 8.5 buffer: 39 g/L	46219009
Solvent solubility at 20°C	n-heptane: 0.23 mg/L xylene: 130 mg/L acetone: 20 g/L ethyl acetate: 3.3 g/L dichloromethane: 56 g/L methanol: 8.3 g/L	Electronic communication, J. Messina to E. Kraft, 9/6/2006
Vapor pressure	1.1×10^{-7} at 20°C	46219010
Dissociation constant, pK_a	The test material becomes increasingly less soluble in water as the pH is lowered and undergoes degradation (hydrolysis) at neutral to acidic pHs. The test material is predicted to have 5 overlapping dissociation constants.	46219011
Octanol/water partition coefficient, $Log(K_{OW})$	pH 4: 2.0 pH 7: 1.3	46219012
UV/visible absorption spectrum	at pH 6.9, A=0.49 and $\varepsilon = 2.1 \times 10^4$ at 238 nm	46219001

B. MATERIALS AND METHODS

B.1. Data-Gathering Method

Not applicable to this submission. Refer to the DER for MRID 46578960 for the residue analytical method used for the rice field trial, storage stability, and processing studies submitted in conjunction with the orthosulfamuron tolerance petition.

B.2. Enforcement Method

The petitioner submitted a description and method validation data for an LC/MS/MS enforcement multiresidue method (QuEChERS European method) for the determination of residues of orthosulfamuron in a matrix with low water content. This study was conducted in Germany, and the method was used to determine residues of orthosulfamuron *per se* in/on rice grain.

B.2.1. Principle of the Method

The QuEChERS multiresidue method uses a single-step buffered ACN extraction and salting out liquid-liquid partitioning from the water in the sample with magnesium sulfate and sodium acetate. The sample is cleaned up by using dispersive solid phase extraction (SPE) to remove organic acids, excess water, and other components with primary secondary amine (PSA) and magnesium sulfate. An aliquot of the supernatant is diluted with 0.1% formic acid for HPLC/MS/MS analysis.

TABLE B.2.1. Summary Parameters for a European Analytical Enforcement Multiresidue Method for the Quantitation of Orthosulfamuron Residues in Rice Grain.

Method ID	QuEChERS multiresidue method
Analyte	Orthosulfamuron *per se*
Extraction solvent/technique	Samples of rice grain are soaked in warm water and allowed to swell. Samples are then extracted with ACN with 1% acetic acid. The internal standard spiking solution (d10-parathion in ACN) is added to the extraction solution and subsequently, magnesium sulfate and sodium acetate are added. The mixture is shaken by hand for about 1 minute, and then centrifuged.
Cleanup strategies	After complete separation of the above extraction solution, the upper ACN phase is cleaned up by dispersive SPE. Primary secondary amine (PSA) and magnesium sulfate are added to the upper ACN phase, and shaken by hand for 1 minute and centrifuged for 2 minutes. An aliquot of the supernatant is diluted with 0.1% formic acid for analysis.
Instrument/Detector	HPLC utilizing a C18 column with tandem mass spectrometry (MS/MS) detection using electrospray ionization operating in the positive ion mode. A gradient mobile phase of ACN and water each containing 0.1% formic acid is used. The ions monitored for orthosulfamuron are 425→199 amu (for quantitation) and 425→120 amu (for qualitative confirmation) and the ions monitored for the d10-parathion internal standard are 302→238 amu.
Standardization method	External and internal standards are used. The internal standard of d10-parathion is added to samples to consider the loss of the acidic ACN during the liquid-liquid partitioning step after salting out. Therefore a correction factor is included in the calculation formula. External standards of orthosulfamuron (calibration curve) are used for quantitation.

TABLE B.2.1. Summary Parameters for a European Analytical Enforcement Multiresidue Method for the Quantitation of Orthosulfamuron Residues in Rice Grain.	
Stability of std solutions	Fortification and external standard solutions were stored refrigerated (4 °C) in the dark; the stability of the standard solutions was not discussed.
Retention times	~5.1 minutes for orthosulfamuron

C. RESULTS AND DISCUSSION

C.1. Data-Gathering Method

Not applicable to this submission.

C.2. Enforcement Method

The petitioner has submitted validation data to determine the applicability of the European enforcement multiresidue method (LC/MS/MS; QuEChERS method) for the determination of orthosulfamuron residues in rice grain. These data were generated by Dr. Specht & Partner, Chemische Laboratorien GmbH. The samples used were obtained by the laboratory. Method validation data are presented in Table C.2.1; adequate recoveries of othosulfamuron were obtained. Following fortification of rice grain samples at 0.01 and 0.10 ppm, recoveries of orthosulfamuron averaged 87% with a standard deviation of 11%. The fortification levels used in method validation are adequate to bracket expected residue levels in rice grain; however, no validation data were provided for rice straw.

TABLE C.2.1. Recovery Results from Method Validation of Rice Grain using the European Enforcement Multiresidue Analytical Method. [1]			
Matrix	Spiking Level (ppm)	Recoveries Obtained	Mean Recovery ± SD [CV] (%)
Rice grain	0.01	70, 77, 82, 84, 105	87 ± 11 [13]
	0.1	78, 81, 94, 96, 99	

[1] Standards were prepared in acetonitrile.

TABLE C.2.2. Characteristics for the European Enforcement Multiresidue Analytical Method for the Quantitation of Orthosulfamuron Residues in Rice Grain.	
Analyte (s)	Orthosulfamuron *per se*
Equipment ID	Agilent 1100 HPLC: Phenomenex Luna C18 column; Applied Biosystems QTrap 4000 tandem mass spectrometer
Limit of quantitation (LOQ)	0.01 ppm
Limit of detection (LOD)	0.003 ppm
Accuracy/Precision	Percent recoveries and coefficients of variance (CVs) indicate acceptable accuracy/precision for residues of orthosulfamuron in rice grain at the LOQ and 10x LOQ. Recovery ranges (and CVs) were 70-105% (13) for orthosulfamuron in/on rice grain. See Table C.2.1 above.
Reliability of the Method [ILV]	No data have been submitted reflecting independent laboratory validation (ILV) of the method.
Linearity	The method/detector response was linear (coefficient of determination, r^2 = 0.9996) within the range of 0.04-2.0 ng/mL.

TABLE C.2.2. Characteristics for the European Enforcement Multiresidue Analytical Method for the Quantitation of Orthosulfamuron Residues in Rice Grain.	
Specificity	The method is capable of determining orthosulfamuron in the presence of the sample matrix. 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.

D. CONCLUSION

Adequate validation data have been submitted for the QuEChERS multiresidue method (QuEChERS method) using rice grain fortified with orthosulfamuron at 0.01 (LOQ) and 0.10 ppm. The fortification levels and samples used in method validation are adequate to bracket expected residue levels for rice grain. However, the QuEChERS method does not meet the FDA multiresidue method requirements under 860.1360.

E. REFERENCES

None.

F. DOCUMENT TRACKING

Petition Number: 5F6957
DP Barcode: D319614
PC Code: 108209

Primary Evaluator	Douglas Dotson, Chemist, RAB2 D. Dotson	Date: 2/14/2007
Peer Reviewer	Dennis McNeilly, Chemist, RAB2 Dennis McNeilly	Date: 2/14/2007

This DER was originally prepared under contract by Dynamac Corporation (2275 Research Boulevard, Suite 300; Rockville, MD 20850; submitted 06/12/2006). The DER has been reviewed by the Health Effects Division (HED) and revised to reflect current Office of Pesticide Programs (OPP) policies.

STUDY REPORTS:

46578960 Zini, G.; Crisippi, T. (2003) Set Up and Validation of a Method for Residue of IR5878 in Rice (Plant, Grain, Straw and Processed Commodities). Project Number: 2305. Unpublished study prepared by Isagro Ricerca S.R.L. 89 p.

46578983 Rose, J. (2004) Storage Stability of IR5878 in Rice Grain and Straw Stored in the Dark Below -20°C: (Final Report). Project Number: 1176W, A2_06.00/01. Unpublished study prepared by PTRL West, Inc. 68 p.

EXECUTIVE SUMMARY:

Isagro S.p.A. has submitted an LC/MS/MS method (referenced as Report ISA-0102V) and an LC/MS method (referenced as Report 2305) for the determination of residues of orthosulfamuron in/on rice grain and straw.

The LC/MS/MS method (Report ISA-0102V) was the data-collection method used in the analysis of samples collected from the rice field trial, storage stability, and processing studies. The method was based on the "Enforcement Method (including Validation) for the Determination of Residues of IR5878 in Rice Grain, Rice Green Plant and Rice Straw" as presented in Report ISA-0102V, Dr. Specht & Partner, 2002. Method descriptions along with validation data were included as an attachment to the storage stability study (MRID 46578983).

The petitioner has also submitted a similar LC/MS method (MRID 46578960) with validation data, for rice RAC and processed commodities, which includes an additional optional cleanup step. For both methods, the LOQ is 0.05 ppm for all rice matrices, and the reported LOD is 0.01 ppm for Method Report ISA-0102V and 0.03 ppm for Method Report 2305.

Using LC/MS/MS method (Report ISA-0102V), homogenized rice samples were extracted twice with acetonitrile (ACN):0.02 M triethylamine (4:1, v:v) and then filtered. Sodium chloride was added to the filtrate to induce separation of the aqueous and ACN phases. An aliquot of the ACN layer was brought to volume with additional ACN and partitioned with hexane. The resulting ACN phase was evaporated to dryness, and residues were redissolved in methanol and

water for LC/MS/MS analysis. The ions monitored for orthosulfamuron are 425→199 amu (for quantitation) and 425→227 amu (for qualitative confirmation).

Using LC/MS Method (Report 2305), the ACN extract (after the addition of sodium chloride) was partitioned with hexane pre-saturated with ACN. The concentrated ACN phase was then redissolved in dichloromethane, filtered, and dried again for further cleanup (if necessary) through an alumina/activated charcoal column. The eluate was dried, and residues were redissolved in ACN for LC/MS analysis. The ion monitored for orthosulfamuron is 425 amu (for quantitation), and the transition ions are 199 and 227 amu (for qualitative confirmation).

Both methods are adequate for data collection based on acceptable method recoveries. Using Method Report ISA-0102V, recoveries of orthosulfamuron averaged 70% with a standard deviation (s.d.) of 5% for green plants and 80% (s.d. 8%) for grain following fortification of rice samples at 0.05 and 0.50 ppm. Acceptable concurrent validation recoveries were also obtained with Method Report ISA-0102V for rice grain and straw fortified at 0.05 ppm from the field trials, and for rice grain, polished rice, bran and hulls fortified at 0.05 ppm from the processing study. Using Method Report 2305, recoveries of orthosulfamuron averaged 96% (s.d. 7%) for plants, 79% (s.d. 8%) for grain, 88% (s.d. 13%) for straw, 103% (s.d. 8%) for husked rice, and 83% (s.d. 15%) for hulls following fortification of rice samples at 0.05 and 0.50 ppm. The fortification levels used in method validation and concurrent validation are adequate to bracket expected residue levels in rice grain, straw, hulled rice, and hulls.

STUDY/WAIVER ACCEPTABILITY/DEFICIENCIES/CLARIFICATIONS:

Under the conditions and parameters used in the study, analytical method test data are classified as scientifically acceptable for data collection methods. However, if either method is proposed for enforcement purposes, the petitioner is required to submit additional data under this guideline topic including independent laboratory validation (ILV) data, radiovalidation data, and confirmatory data.

If Method Report ISA-0102V is proposed for tolerance enforcement, and analyte identification is confirmed by analyzing sample extracts using LC/MS/MS and comparing the ion ratio for the two MS/MS ion transitions acquired during analysis with the average ion ratio obtained for the calibration standards, then a confirmatory method would not be required.

The acceptability of this study for regulatory purposes is addressed in the U.S. EPA Residue Chemistry Summary Document, D332290, D. Dotson, 2/14/2007.

COMPLIANCE:

Signed and dated Good Laboratory Practice (GLP), Quality Assurance and Data Confidentiality statements were provided.

For MRID 46578960, the petitioner stated that the study was not conducted in accordance with GLP standards as defined in 40 CFR 160, and is at variance with GLP standards as defined by the US EPA in the following manner: (a) The study was conducted in compliance with the OECD principles of Good Laboratory Practice as defined by the Republic of Italy, as enunciated in the "Legislative Decree 27.01.1992, No. 120: activation of directive No. 88/320/CE and No. 90/18/CEE with regard to inspection and verification of good laboratory practice" and subsequent updating.; and (b) The signed compliance statement of the Study Director does not conform with US EPA standards which signify GLP compliance.

A. BACKGROUND INFORMATION

Orthosulfamuron is a postemergence herbicide that Isagro S.p.A. is proposing for use on rice grown in the United States for the control of annual and perennial broadleaf weeds, sedges, and barnyard grass. Orthosulfamuron belongs to the sulfamoylurea class of herbicides. It reportedly acts by inhibiting the plant enzyme acetolactate synthase, which is active in the biosynthesis of valine, leucine, and isoleucine.

TABLE A.1. Test Compound Nomenclature.

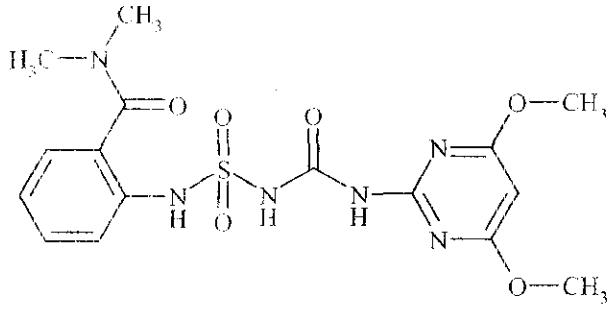
Compound	
Common name	Orthosulfamuron
Company experimental name	IR5878
IUPAC name	1-(4,6-dimethoxypyrimidin-2-yl)-3-[2-(dimethylcarbamoyl)phenylsulfamoyl]urea
CAS name	2-[[[[[(4,6-dimethoxy-2-pyrimidinyl)-amino]carbonyl]amino]sulfonyl]amino]-N,N-dimethylbenzamide
CAS registry number	213464-77-8
End-use product (EP)	0.51% G formulation (IR5878 0.5 GR; EPA Co. No. 80289) 51.5% WG formulation (IR5878 50 WG; EPA Co. No. 80289)

Table 2. Physicochemical Properties of the Technical Grade of Orthosulfamuron.		
Parameter	Value	Reference (MRID)
Color	White	46219004
Physical State	Fine Powder at 20°C	46219005
Odor	Odorless	46219006
pH	4.35 at 25°C (1% aqueous dispersion)	46219013
Density	1.45 g/mL at 20°C	46219008
Water solubility at 20°C	pH 4 buffer: 0.062 g/L pH 7 buffer: 0.63 g/L pH 8.5 buffer: 39 g/L	46219009
Solvent solubility at 20°C	n-heptane: 0.23 mg/L xylene: 130 mg/L acetone: 20 g/L ethyl acetate: 3.3 g/L dichloromethane: 56 g/L methanol: 8.3 g/L	Electronic communication, J. Messina to E. Kraft, 9/6/2006
Vapor pressure	1.1×10^{-4} at 20°C	46219010
Dissociation constant, pK_a	The test material becomes increasingly less soluble in water as the pH is lowered and undergoes degradation (hydrolysis) at neutral to acidic pHs. The test material is predicted to have 5 overlapping dissociation constants.	46219011
Octanol/water partition coefficient, $Log(K_{OW})$	pH 4: 2.0 pH 7: 1.3	46219012
UV/visible absorption spectrum	at pH 6.9, A=0.49 and $\varepsilon = 2.1 \times 10^4$ at 238 nm	46219001

B. MATERIALS AND METHODS

B.1. Data-Gathering Method

An LC/MS/MS method was used for the quantitation of orthosulfamuron residues in the rice field trial, storage stability, and processing studies associated with the orthosulfamuron tolerance petition. The studies reference that the method used was based on the "Enforcement Method (including Validation) for the Determination of Residues of IR5878 in Rice Grain, Rice Green Plant and Rice Straw" as presented in Report ISA-0102V, Dr. Specht & Partner, 2002. This method with validation data was available only as an attachment to the storage stability study (MRID 46578983).

The petitioner also submitted a description and method validation data for a similar LC/MS method, which includes an additional optional cleanup step, for rice RAC and processed commodities (MRID 46578960).

B.1.1. Principle of the Method:

Under the Method Report ISA-0102V and as described for the rice field trial, storage stability, and processing studies, homogenized samples were extracted twice with acetonitrile (ACN):0.02 M triethylamine (4:1, v:v) and filtered. Sodium chloride was added to the filtrate to induce separation of the aqueous and ACN phases. An aliquot of the ACN layer was brought to volume with additional ACN and partitioned with hexane. The resulting ACN phase was evaporated to dryness and residues were redissolved in methanol and water for LC/MS/MS analysis.

Under LC/MS Method Report 2305, the ACN extract after the addition of sodium chloride was partitioned with hexane pre-saturated with ACN. The concentrated ACN phase was then redissolved in dichloromethane, filtered, and dried again for further cleanup (if necessary) through an alumina/activated charcoal column. The eluate was dried and residues were redissolved in ACN for LC/MS analysis.

TABLE B.1.1. Summary Parameters for the Analytical Method Used for the Quantitation of Orthosulfamuron Residues in Rice Commodities.

Method ID	LC/MS/MS Report ISA-0102V	LC/MS Report 2305
Analyte	Orthosulfamuron	
Extraction solvent/technique	Homogenized samples were extracted twice with acetonitrile (ACN):0.02 M triethylamine mixture (4:1, v:v) and filtered. The filtrate was transferred to a separatory funnel and salted with sodium chloride	
Cleanup strategies	After shaking, the upper organic phase was transferred to a second separatory funnel, brought to volume with additional ACN, and partitioned with n-hexane. The lower ACN phase was collected and dried at 40°C by rotary evaporation. The residue was redissolved in methanol and water was added.	After shaking, the upper organic phase was transferred to a second separatory funnel and partitioned with n-hexane pre-saturated with ACN. After shaking, the lower ACN phase was collected and dried at reduced pressure by rotary evaporation. The residue was redissolved in dichloromethane (DCM), filtered, and dried again. If necessary, the residue was then redissolved in n-hexane:acetone (8:2, v:v) for further cleanup through an alumina:activated charcoal (2:1) column with a sodium sulfate layer; residues were eluted with DCM:methyl alcohol:acetic acid (94.5:1, v:v:v). The eluate was dried by rotary evaporation to remove the acetic acid and residues were redissolved in ACN.
Instrument/Detector	HPLC with a C18 column and a gradient mobile phase of water and methanol each containing 0.05% acetic acid. MS detector using electrospray ionization in the positive ion mode. The ions monitored for orthosulfamuron are 425→199 amu (for quantitation) and 425→227 amu (for qualitative confirmation).	HPLC with a C18 column and a gradient mobile phase of water plus 10 mM ammonium acetate and ACN, each containing 0.1% formic acid. MS detector using electrospray ionization in the positive ion mode. The ion monitored for orthosulfamuron is 425 amu (for quantitation) and the transition ions are 199 and 227 amu (for qualitative confirmation).
Standardization method	Calibration curve of external standards of orthosulfamuron, using linear regression analysis.	
Stability of std solutions	Stability of standard solutions was not discussed.	
Retention times	~7.1 minutes (as per method write-up)	~12 minutes (representative chromatograms)

B.2. Enforcement Method

Not applicable to this submission. We note that an analytical method has not been proposed for enforcement of tolerances for plant commodities.

C. RESULTS AND DISCUSSION

The petitioner has submitted method validation data for the LC/MS/MS method under Report ISA-0102V and the LC/MS method under Report 2305. These data were generated by Dr. Specht & Partner, Chemische Laboratorien GmbH (Germany; Report ISA-0102V) or Isagro Ricerca S.r.l. (Italy; Report 2305); the samples used were obtained by the laboratory. Method validation data are presented in Table C.1.1; overall adequate recoveries of othosulfamuron were obtained with rice commodities. Following fortification of rice samples at 0.05 and 0.50 ppm, method recoveries of orthosulfamuron averaged 70% (s.d. 5%) for green plants and 80% (s.d. 8%) for grain using Method Report ISA-0102V. Acceptable concurrent validation recoveries were also obtained for rice grain and straw fortified at 0.05 ppm from the field trials, and for rice grain, polished rice, bran, and hulls fortified at 0.05 ppm from the processing study. Following fortification of rice samples at 0.05 and 0.50 ppm, recoveries of orthosulfamuron averaged 96% (s.d. 7%) for plants, 79% (s.d. 8%) for grain, 88% (s.d. 13%) for straw, 103% (s.d. 8%) for husked rice, and 83% (s.d. 15%) for hulls using Method Report 2305. The fortification levels used in method and concurrent validation are adequate to bracket expected residue levels in rice grain, straw, hulled rice, and hulls.

C.1. Data-Gathering Method

TABLE C.1.1. Recovery Results from Method Validation of Rice Commodities using the Data-Gathering Analytical Method. [1]

Matrix	Spiking Level (ppm)	Recoveries Obtained (%)	Mean Recovery ± SD [CV] (%)
LC/MS/MS Report ISA-0102V [1]			
Rice green plants	0.0502	62, 67, 73, 74, 78	70 ± 4.9 [7.0]
	0.502	64, 69, 70, 73, 73	
Rice grain	0.0502	60, 78, 81, 81, 85	80 ± 8.1 [10]
	0.502	78, 79, 80, 83, 92	
Rice straw	0.0504	Not reported	
	0.504		
LC/MS Report 2305 [2]			
Rice plants	0.0520	94.23, 95.92, 98.15, 104.77, 110.04	96.39 ± 6.94 [7.2]
	0.5200	87.47, 89.77, 91.94, 92.27, 99.35	
Paddy rice (grain)	0.0500	67.65, 73.49, 73.75, 75.08, 94.67	79.39 ± 8.34 [10.5]
	0.4995	74.35, 79.33, 80.67, 85.04, 89.87	
Rice straw	0.0520	85.3, 92.6, 93.3, 95.5, 105.9	88.37 ± 12.68 [14.4]
	0.5200	69.74, 72.59, 79.35, 82.83, 106.49	
Husked rice	0.0500	97.16, 97.26, 116.32	103.37 ± 8.05 [7.8]
	0.4995	97.86, 101.45, 110.19	
Rice hulls	0.0500	80.82, 97.62, 103.68	82.60 ± 14.80 [17.9]
	0.4995	67.53, 70.74, 75.24	

[1] Standards were prepared in methanol/water.
[2] Standards were prepared in acetonitrile.

TABLE C.1.2. Characteristics for the Data-Gathering Analytical Method Used for the Quantitation of Orthosulfamuron Residues in Rice Commodities.

Method ID	LC/MS/MS Report ISA-0102V	LC/MS Report 2305
Analyte(s)	Orthosulfamuron	
Equipment ID	HP 1100 HPLC; Phenomenex LUNA C18 column; PE-Sciex API 2000 mass spectrometer	HP 1100 HPLC, Agilent Technologies; INERTISIL ODS-3 column; quadrapole mass detector
Limit of quantitation (LOQ)	0.05 ppm; lowest level of method validation.	
Limit of detection (LOD)	0.01 ppm; 3x background noise.	0.03 ppm; evaluation of background noise at the retention time of orthosulfamuron.
Accuracy/Precision	Percent recoveries and coefficients of variance (CVs) indicate acceptable accuracy/precision for residues of orthosulfamuron in rice green plants and grain at the LOQ and 10x LOQ. Recovery ranges (and CVs) were 60-92% (7-10) for orthosulfamuron in/on rice green plants and grain. See Table C.1.1 above.	Percent recoveries and coefficients of variance (CVs) indicate acceptable accuracy/precision for residues of orthosulfamuron in rice plants, paddy rice (grain), rice straw, husked rice, and rice hulls at the LOQ and 10x LOQ. Recovery ranges (and CVs) were 68-116% (7-18) for orthosulfamuron in/on rice matrices. See Table C.1.1 above.
Reliability of the Method [ILV]	No data have been submitted reflecting independent laboratory validation (ILV) of the method.	

TABLE C.1.2. Characteristics for the Data-Gathering Analytical Method Used for the Quantitation of Orthosulfamuron Residues in Rice Commodities.		
Method ID	LC/MS/MS Report ISA-0102V	LC/MS Report 2305
Linearity	The method states the detector response is linear within the range of 0.503-67.0 ng/mL (a representative calibration curve was not included)	The method/detector response was linear (coefficient of determination, r^2 = 0.99785) within the range of 0.022-1.110 mg/L.
Specificity	The method is capable of determining orthosulfamuron in the presence of the sample matrix. 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.	

C.2. Enforcement Method

The petitioner has stated that for the purpose of tolerance enforcement, the parent orthosulfamuron can be determined using HPLC with a mass spectrometer (MS) detector. The proposed LOD and LOQ for the method are 0.03 and 0.05 ppm, respectively.

D. CONCLUSION

The LC/MS/MS and LC/MS methods are adequate as data collection methods for the determination of residues of orthosulfamuron *per se* in/on rice commodities. Adequate validation data have been submitted for the data collection methods using rice matrices (rice plant, grain, straw, husked rice, and rice hulls) fortified with orthosulfamuron at 0.05 (LOQ) and 0.5 ppm.

E. REFERENCES

None.

F. DOCUMENT TRACKING

Petition Number: 5F6957
DP Barcode: D319614
PC Code: 108209

Primary Evaluator Douglas Dotson, Chemist, RAB2 D. Dotson Date: 2/14/2007

Peer Reviewer Dennis McNeilly, Chemist, RAB2 Date: 2/14/2007

This DER was originally prepared under contract by Dynamac Corporation (2275 Research Boulevard, Suite 300; Rockville, MD 20850; submitted 06/12/2006). The DER has been reviewed by the Health Effects Division (HED) and revised to reflect current Office of Pesticide Programs (OPP) policies.

STUDY REPORTS:

46578962 Kidd, G.; Gedik, L. (2003) The Distribution and Metabolism of (^{14}C)-IR5878 in the Lactating Goat: In Life Phase. Project Number: 204061, A2_06.02/01. Unpublished study prepared by Inveresk Research International. 57 p.

46578963 Rizzo, F.; Mainolfi, K.; Pizzingrilli, G. (2003) Profiling of (^{14}C)-IR5878 Metabolites in Milk and Edible Tissues from Lactating Goat. Project Number: MEF.03.01, A2_06.02/02. Unpublished study prepared by Isagro S.R.L. (Formerly Agrimont). 168 p.

EXECUTIVE SUMMARY:

Isagro S.p.A. submitted a study investigating the metabolism of [^{14}C-5-pyrimidinyl] orthosulfamuron (PY label; specific activity 22.13 µCi/mg) and [^{14}C-U-phenyl]orthosulfamuron (PH label; specific activity 18.35 µCi/mg) in lactating goats. The radiolabeled test substances were administered orally to separate goats at 18.09 mg/day (PY label) or 22.66 mg/day (PH label) for five consecutive days. Based on the daily dietary intake, the dosage corresponded to 10.26 ppm (PY label) and 13.11 ppm (PH label) in the feed. Milk was collected twice daily during the dosing period, and the test goats were sacrificed 23 hours after the last dose administration. Tissues collected at animal sacrifice include both kidneys, liver, omental fat, renal fat, and skeletal muscle (hind and fore quarters). The in-life phase was conducted by Inveresk Research (Tranent, Scotland) whereas the analytical phase was performed by Isagro Ricerca Srl (Novara, Italy).

Total radioactive residues (TRR) were determined at the in-life facility. TRR were 0.005-0.014 ppm in milk, <0.002 ppm in muscle, 0.003 ppm in omental and renal fat, 0.125 ppm in liver, and 0.090 ppm in kidney from the goat orally dosed with PY-labeled orthosulfamuron. TRR were 0.004-0.016 ppm in milk, 0.007 ppm in muscle, 0.003 ppm in omental and renal fat, 0.131 ppm in liver, and 0.144 ppm in kidney from the goat orally dosed with PH-labeled orthosulfamuron. TRR were highest in liver and kidney, and <0.01 ppm in muscle and fat. The TRR were consistently low in milk but appear to plateau at 72 hours for the PY label goat and 48 hours for the PH label goat. The majority (90-97%) of the administered dose was excreted: ~50% in the feces, ~32-40% in the urine, and ~8% in the cage washes.

Muscle and fat were not extracted for metabolite identification because the TRR was <0.01 ppm. The majority of the radioactivity was released with solvent extraction; ~95-103% TRR from milk with methanol, and ~93% TRR from kidney and ~48-69% TRR from liver with ACN/ammonium bicarbonate. Additional radioactivity was released from the nonextractable residues of liver with chemical or enzyme hydrolyses. Protease hydrolysis released the largest amount (45% TRR from PY label liver and 23% from PH label liver). Nonextractable residues were nondetectable in milk and ≤5% TRR (≤0.005 ppm) in kidney (both labels). Nonextractable residues remaining in liver after enzyme hydrolysis were not determined; however, based on the total radioactivity released, the nonextractable residues would be <10% TRR in liver (both labels). These procedures adequately extracted and characterized the majority of residues from goat milk and tissues. Accountabilities ranged from 92 to 100%.

Residues were quantitated by TLC, and identified with goat urine metabolites using TLC, HPLC, and/or LC/MS. Sample integrity was maintained by appropriate freezer storage prior to residue analysis; however, actual extraction and analysis dates were not provided. Goat milk, kidney, and liver samples may have been stored for up to 5.5 months based on the initial dosing date (11/14/02) and a statement by the petitioner that analysis was completed April 2003. No storage stability data are available to support the storage intervals and conditions of samples from the goat metabolism study. The petitioner should submit the dates of extraction, initial TLC analysis, and metabolite identification analyses.

The metabolite profile differed somewhat between the PY and PH labels, demonstrating possible cleavage of the molecule between the two rings. Parent, orthosulfamuron, was identified in all goat milk and tissue samples. The parent was present at low levels in milk, accounting for 8.9-11.2% TRR (0.001 ppm) in PY label milk, and 7.9-16.6% TRR (0.001-0.002 ppm) in PH label milk. The parent was identified as a major residue in tissues, accounting for 26.4% TRR (0.024 ppm) in PY label kidney, 27.8% TRR (0.037 ppm) in PH label kidney, 12.7% TRR (0.016 ppm) in PY label liver, and 20.5% TRR (0.030 ppm) in PH label liver.

Pyr-O-Sulf DOP urea was identified as the major metabolite in PY label milk and kidney accounting for 59.1-76.4% TRR (0.005-0.011 ppm) in milk and 25.7% TRR (0.023 ppm) in kidney; Pyr-O-Sulf DOP urea was also identified in PY label liver as a minor metabolite (3.3% TRR, 0.004 ppm). Pyr-O-Sulf DOP urea only contains the parent pyrimidinyl ring and, therefore, was not identified in any PH label goat matrices. DOP urea, another pyrimidinyl ring metabolite, was only identified in liver at 3.8% TRR (0.005 ppm).

Metabolites N-desm-O-desm IR5878, N-desm IR5878, and O-desm IR5878, each with the molecule bridge between the two rings intact, were identified in both PY and PH label goat matrices. N-desm IR5878 accounted for 9.4-17.0% TRR (0.001-0.002 ppm) in PY label milk, 13.3% TRR (0.012 ppm) in PY label kidney, and 7.3% TRR (0.009 ppm) in PY label liver; and 21.9-34.9% TRR (0.004-0.005 ppm) in PH label milk, 17.7% TRR (0.023 ppm) in PH label kidney, and 10.9% TRR (0.016 ppm) in PH label liver. O-desm IR5878 was identified as a minor residue in all milk and tissue samples; 2.7-8.2% TRR (≤0.001 ppm) in milk, 3.4-4.0% TRR (≤0.005 ppm) in kidney, and 4.1-7.0% TRR (≤0.010 ppm) in liver (both labels). N-desm-

O-desm IR5878 was also identified as a minor residue, accounting for 2.5-7.2% TRR (≤0.001 ppm) in PH label milk, 1.8-2.5% TRR (0.002 ppm) in PY and PH label kidney, and 1.4% TRR (0.002 ppm) in PY label liver. N-desm-O-desm IR5878 was not detected in PY label milk or PH label liver.

Metabolites DBS acid, N-desm DB amine, and DBS amide were only identified in PY label milk and tissues, because each contain only the parent phenyl ring. DBS acid accounted for 8.6-15.2% TRR (≤0.002 ppm) in milk, 11.4% TRR (0.015 ppm) in kidney, and 4.6% TRR (0.007 ppm) in liver. N-desm DB amine accounted for 9.4-16.3% TRR (≤0.002 ppm) in milk, 13.8% TRR (0.018 ppm) in kidney, and 8.0% TRR (0.012 ppm) in liver; and DBS amide accounted for 10.6-14.1% TRR (≤0.002 ppm) in milk, 4.5% TRR (0.006 ppm) in kidney, and 3.9% TRR (0.006 ppm) in liver.

A significant amount of radioactivity in goat liver was characterized as protein bound residues (45.5% TRR, 0.057 ppm in PY label liver and 22.6% TRR, 0.032 ppm in PH label liver). The remaining residues in goat milk and tissues were characterized as unknowns totaling 2.7-15.9% TRR in milk and 11.8-20.4% TRR in kidney and liver, with individual peaks accounting for <0.01 ppm.

In the goat metabolism study, the radioactivity in goat milk, muscle, and fat was negligible, and very low in the liver and kidneys. The parent was metabolized to metabolites both with the molecule bridge intact or broken between the pyrimidinyl and phenyl rings, and a significant amount of the residue in liver was eventually bound to protein. The metabolic pathway for orthosulfamuron in goats is similar to that in rats.

STUDY/WAIVER ACCEPTABILITY/DEFICIENCIES/CLARIFICATIONS:

Under the conditions and parameters used in the study, the livestock metabolism data are tentatively classified as scientifically unacceptable, but upgradeable. The dates of sample extraction, initial TLC analysis, and metabolite identification analyses are required to determine the storage intervals of samples from the goat metabolism study. If the initial quantitative TLC analyses were conducted within 6 months of sample collection, then supporting storage stability data will not be required to support the additional analyses for metabolite identification in goat matrices.

The acceptability of this study for regulatory purposes is addressed in the U.S. EPA Residue Chemistry Summary Document, D332290, D. Dotson, 2/14/2007.

COMPLIANCE:

Signed and dated Good Laboratory Practice (GLP), Quality Assurance and Data Confidentiality statements were provided. The GLP Compliance Statements cite that the study was not conducted in accordance with GLP Standards as defined by the USA EPA, but in compliance with OECD Principles of GLP as defined by the Republic of Italy, as enunciated in the "Legislative Decree 27.01.1992, No. 120: activation of directive No. 88/320/CEE and No. 90/18/CEE with regard to inspection and verification of GLP (analytical study) or OECD Principles of GLP as set forth by the United Kingdom Department of Health (Schedule 1, Good Laboratory Regulations 1999; SI 1999/3106) (in-life study).

A. BACKGROUND INFORMATION

Orthosulfamuron is a postemergence herbicide that Isagro S.p.A. is proposing for use on rice grown in the United States for the control of annual and perennial broadleaf weeds, sedges, and barnyard grass. Orthosulfamuron belongs to the sulfamoylurea class of herbicides. It reportedly acts by inhibiting the plant enzyme acetolactate synthase, which is active in the biosynthesis of valine, leucine, and isoleucine.

TABLE A.1. Test Compound Nomenclature.

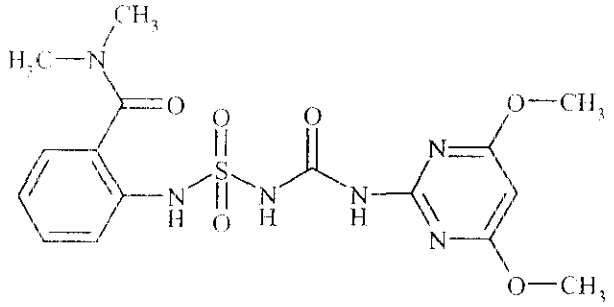
Compound	
Common name	Orthosulfamuron
Company experimental name	IR5878
IUPAC name	1-(4,6-dimethoxypyrimidin-2-yl)-3-[2-(dimethylcarbamoyl)phenylsulfamoyl]urea
CAS name	2-[[[[[(4,6-dimethoxy-2-pyrimidinyl)-amino]carbonyl]amino]sulfonyl]amino]-N,N-dimethylbenzamide
CAS registry number	213464-77-8
End-use product (EP)	0.51% G formulation (IR5878 0.5 GR; EPA Co. No. 80289) 51.5% WG formulation (IR5878 50 WG; EPA Co. No. 80289)

Table 2. Physicochemical Properties of the Technical Grade of Orthosulfamuron.

Parameter	Value	Reference (MRID)
Color	White	46219004
Physical State	Fine Powder at 20°C	46219005
Odor	Odorless	46219006
pH	4.35 at 25°C (1% aqueous dispersion)	46219013

Table 2. Physicochemical Properties of the Technical Grade of Orthosulfamuron.		
Parameter	Value	Reference (MRID)
Density	1.45 g/mL at 20°C	46219008
Water solubility at 20°C	pH 4 buffer: 0.062 g/L pH 7 buffer: 0.63 g/L pH 8.5 buffer: 39 g/L	46219009
Solvent solubility at 20°C	n-heptane: 0.23 mg/L xylene: 130 mg/L acetone: 20 g/L ethyl acetate: 3.3 g/l dichloromethane 56 g/L methanol: 8.3 g/L	Electronic communication, J. Messina to E. Kraft, 9/6/2006
Vapor pressure	1.1×10^{-4} at 20°C	46219010
Dissociation constant, pK_a	The test material becomes increasingly less soluble in water as the pH is lowered and undergoes degradation (hydrolysis) at neutral to acidic pHs. The test material is predicted to have 5 overlapping dissociation constants.	46219011
Octanol/water partition coefficient, $Log(K_{OW})$	pH 4: 2.0 pH 7: 1.3	46219012
UV/visible absorption spectrum	at pH 6.9, A= 0.49 and $\varepsilon = 2.1 \times 10^4$ at 238 nm	46219001

B. EXPERIMENTAL DESIGN

Two lactating goats were orally dosed with radiolabeled orthosulfamuron at a nominal rate of 10 ppm in the diet for five consecutive days. One goat was dosed with [^{14}C-5-pyrimidinyl]orthosulfamuron (PY label) and another was dosed with [^{14}C-U-phenyl]orthosulfamuron (PH label). Each radiolabeled test substance was diluted with nonlabeled orthosulfamuron and prepared in a gelatin capsule for dosing. A third goat received gelatin capsules without orthosulfamuron as a control.

The goats were maintained in individual metabolism cages and fed a commercial protein ration twice daily at milking and hay *ad libitum*. The PY label goat received 18.09 mg/day and the PH label goat received 22.66 mg/day of the test substance. Actual dosing rates in the diet were calculated using the feed consumption for that day.

The goats were milked twice daily and just prior to sacrifice. Excreta were collected once daily. Goats were sacrificed 23 hours after the last dosing after which tissues, blood and bile were collected.

B.1. Livestock

TABLE B.1.1. General Test Animal Information.

Species	Breed	Age	Weight at study initiation (kg)	Health Status	Description of housing/holding area
Goat	Saanen	3.5-6 yrs.	57.5-69.0	Good general health	Individually housed in loose pens (pre-trial) and metabolism crates suitable for separate collection of urine and feces; maintained on a 12 hour light/dark cycle with temperatures ranging 17-21 °C and a relative humidity of 27-74%.

TABLE B.1.2. Test Animal Dietary Regime.

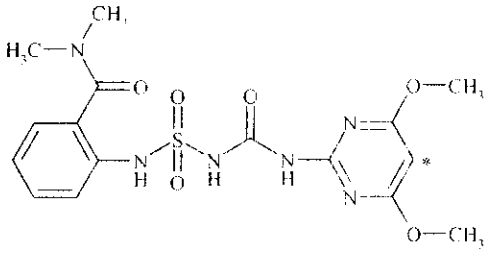
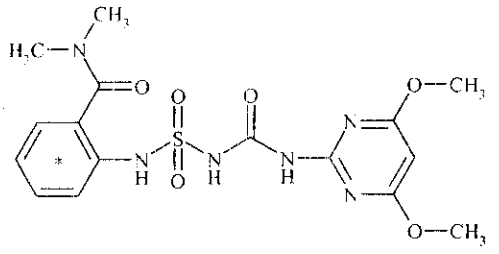
Composition of Diet	Feed consumption (kg/day)	Water	Acclimation period	Predosing
Protein concentrate ration fed twice daily (300 g) at milking; hay offered *ad libitum*	Daily consumption of 0.602-0.625 kg feed and 0.701-1.507 kg hay.	*Ad libitum*	7 days	None

TABLE B.1.3. Test Animal Dosing Regime.

Treatment Type	Treatment Level (ppm test material in vehicle)	Vehicle	Parameters	Timing/Duration
Oral	Target dose equivalent to 10 ppm in the diet; mean dose of 10.26 ppm for the PY label and 13.11 ppm for the PH label.	Capsule using a bolus gun.	Test material in ACN was added to the capsule and the solvent evaporated.	Dosed twice daily, after the morning and afternoon milkings, for five consecutive days.

B.2. Test Materials

TABLE B.2.1. Test Material Characteristics.

Chemical structure		
Radiolabel position	[^{14}C-5-pyrimidinyl]orthosulfamuron (PY-label)	[^{14}C-U-phenyl]orthosulfamuron (PH-label)
Lot No.	208	209
Radiochemical Purity	>98% (TLC and HPLC)	>98% (TLC and HPLC)
Specific activity	Test substance: 4.452 MBq/mg (120.323 µCi/mg) Dosing solution: 0.82 MBq/mg (22.13 µCi/mg)	Test substance: 7.357 MBq/mg (198.850 µCi/mg) Dosing solution: 0.68 MBq/mg (18.35 µCi/mg)

B.3. Sampling Information

TABLE B.3.1. Sample Collection Information.

Milk collected	Urine, feces and cage wash collected	Interval from last dose to sacrifice	Tissues harvested and analyzed
Twice daily (ca. 0830 and 1630 hour); 1231-1354 g/goat prior to first dosing and 350-1384 g/goat from each milking during the dosing period.	Urine, feces, and cage washes separately collected at 24-hour intervals.	23 hours	Kidneys, liver, omental fat, renal fat, skeletal muscle (hind and fore quarter)

B.4. Identification/Characterization of Residues

B.4.1. Sample Handling and Preparation

Tissue samples were processed immediately after collection. kidney and liver samples were homogenized in a blender, and frozen muscle and fat samples were homogenized with dry ice to a fine powder. All samples not analyzed immediately were stored frozen (ca. -20°C) until analysis. Samples were analyzed for total radioactivity at the in-life facility (Inveresk Research, Tranent, Scotland) and frozen samples were shipped within 2.3 months of collection to Isagro Ricerca Srl (Novara, Italy) for extraction and metabolite analysis. Samples were stored frozen (ca. -20°C) at the analytical laboratory until extraction and analysis.

Samples (both labels) of liver and kidney, as well as milk collected at 24, 72, 104, and 120 hours, were extracted for metabolite identification/characterization. Muscle and fat samples were not extracted because of low TRR (≤0.007 ppm). Urine and feces samples were extracted to aid in identification of milk and tissue metabolites.

Aliquots of milk were lyophilized for 24 hours, and the resulting powder was extracted twice with hexane and then twice with methanol. The hexane and methanol extracts were collected after centrifugation; the methanol extract was concentrated and reserved for TLC analysis. Nonextractable residues were dissolved in 0.2 M NaOH for LSC analysis.

Aliquots of homogenized liver and kidney samples were extracted (3x) with acetonitrile (ACN):50 mM ammonium bicarbonate (NH_4HCO_3) pH 8.5 (1:1, v:v) and centrifuged. Liver samples were further extracted with 50 mM NH_4HCO_3 at pH 11 and centrifuged. Individual extracts were radioassayed and all extracts were combined for TLC analysis.

Subsamples of liver nonextractable residues were further treated to release bound radioactivity. Separate aliquots were further extracted with: (i) hexane; (ii) acetone:0.5 N HCl (1:1, v:v); (iii) surfactant; (iv) 6 N HCl at reflux (120 °C for 2 hours) and the hydrolysate was partitioned with ethyl acetate; or (v) protease in pH 7 buffer at 37°C for 48 hours.

The extraction flowcharts are presented in Figures B.4.1.1 (milk), B.4.1.2 (liver and kidney), and B.4.1.3 (liver nonextractable residues). These were copied from MRID 46578963 without alteration.

Figure B.4.1.1. Extraction Scheme for Goat Milk.

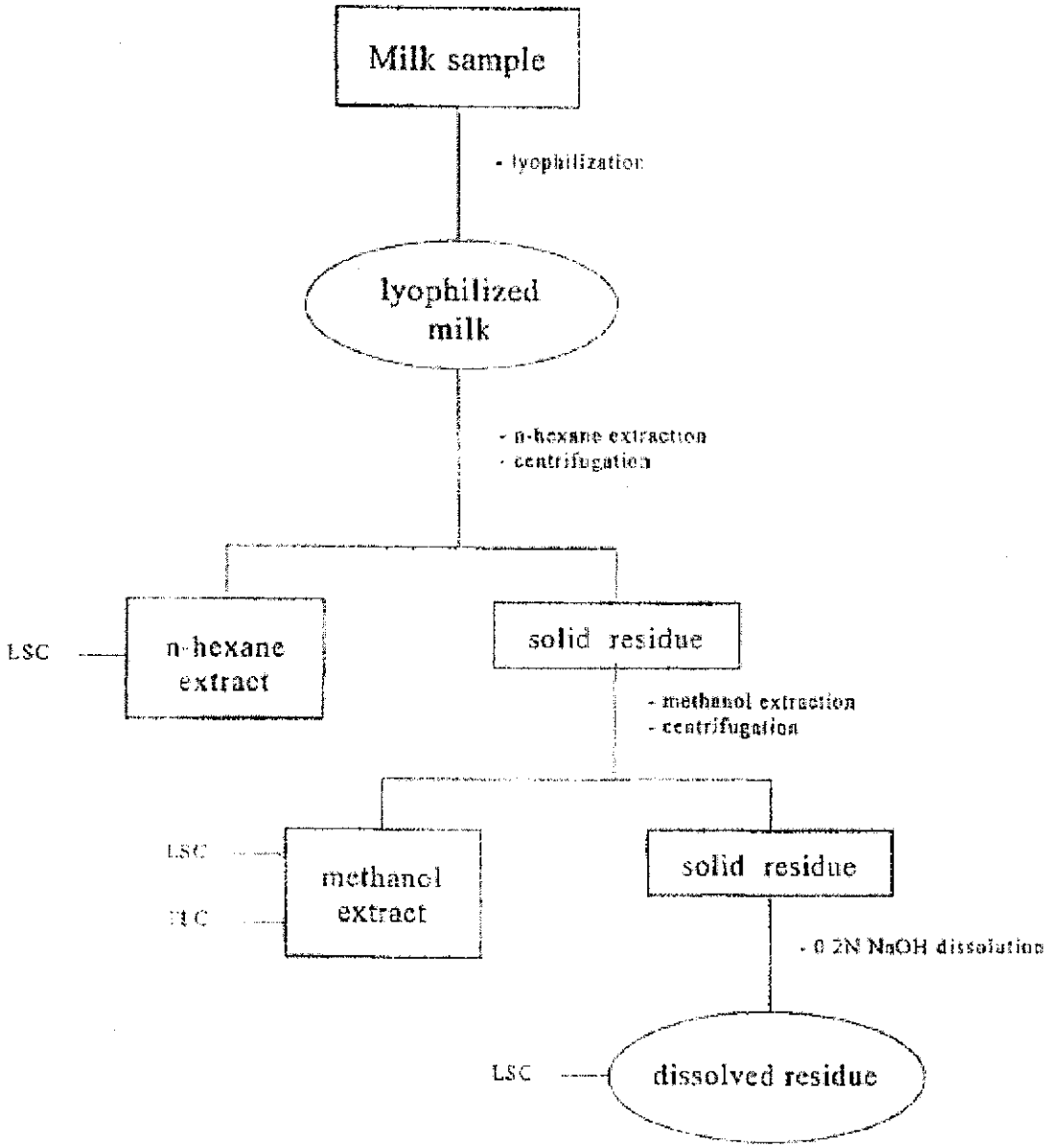


Figure B.4.1.2. Extraction Scheme for Goat Kidney and Liver.

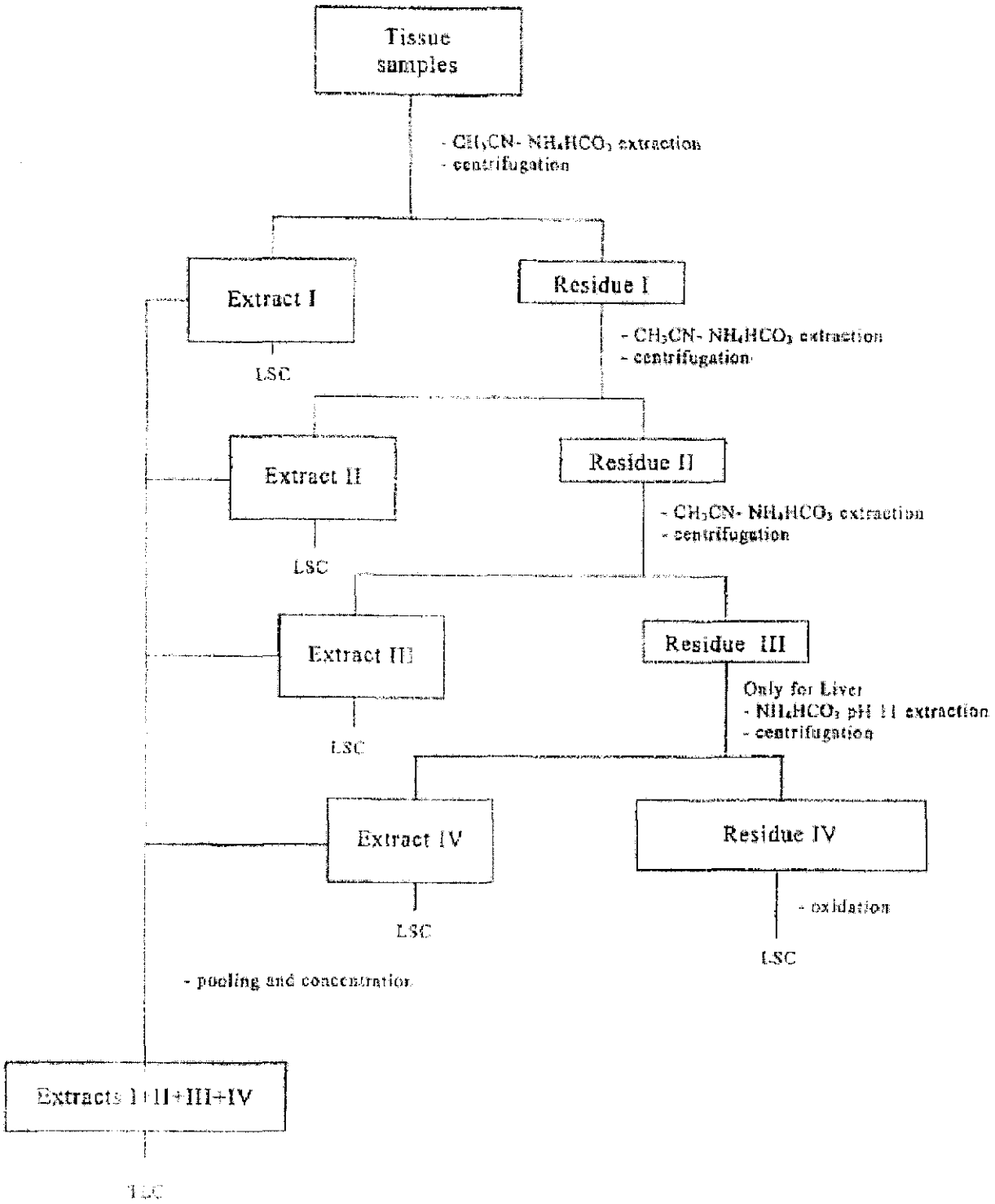
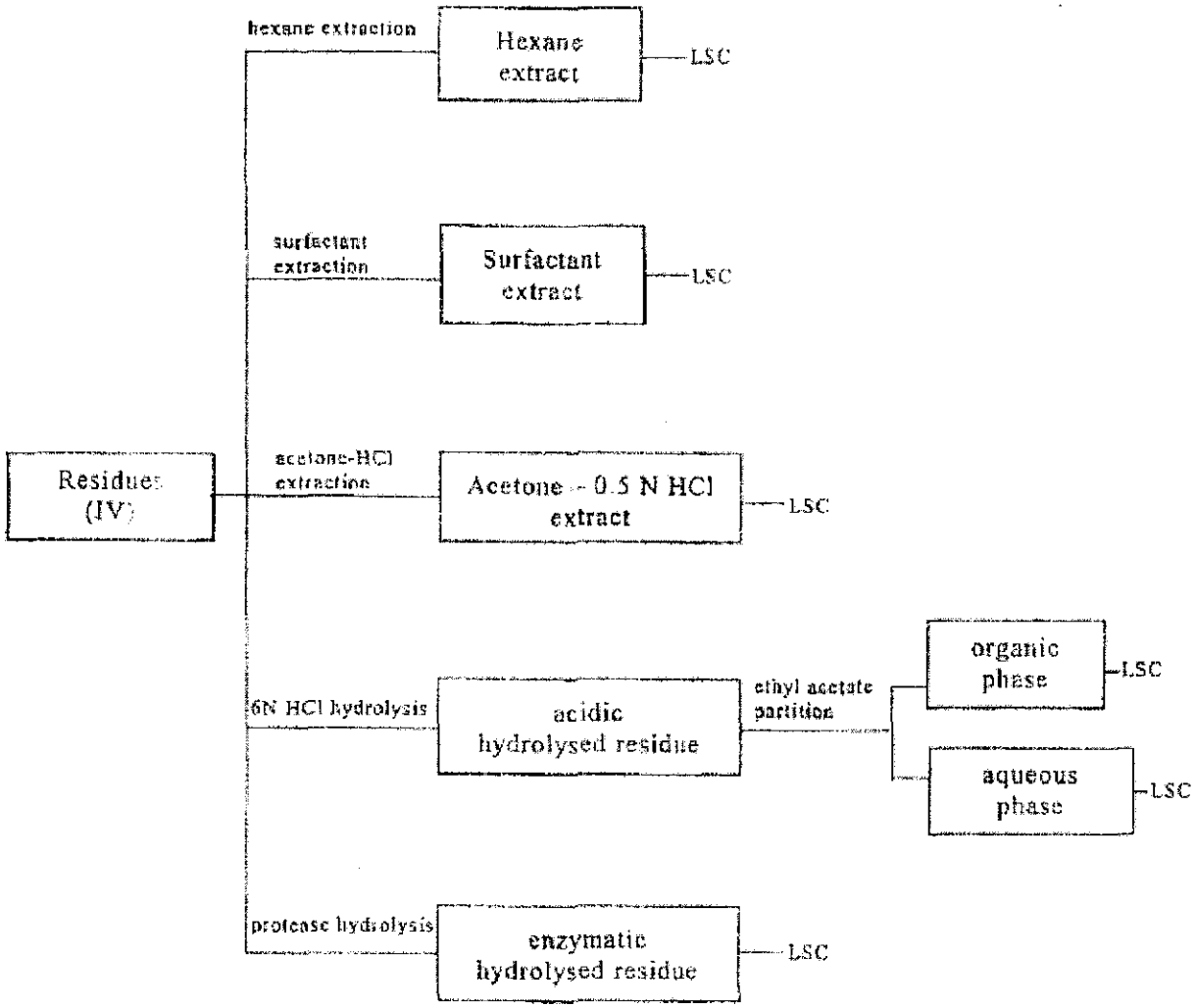


Figure B.4.1.3. Extraction Scheme for Goat Liver Nonextractable Residues.



B.4.2. Analytical Methodology

TRR in milk, urine, cage wash, and bile were determined by LSC. TRR in tissues and feces were determined by combustion/LSC; fat was sonicated with scintillation fluid prior to LSC analysis. Extracts and hydrolysates were radioassayed by LSC, and nonextractable residues were radioassayed by combustion/LSC. The reported limits of quantitation (LOQ) for TRR determinations were 0.002 ppm for muscle, liver, and kidney, and 0.001 ppm for fat (twice the background). The LOQ for milk was not reported.

Extracts of milk, liver, and kidney were subjected to TLC analysis for quantitation and identification of metabolites. Normal phase TLC analyses were conducted using silica gel 60 F_{254} plates and a solvent system of chloroform:methanol:ammonium hydroxide (75:22:3, v:v:v). TLC plates were evaluated using radioimaging; luminescence was detected by a photo multiplier.

Unresolved radioactivity (origin) was isolated from liver and kidney using preparative TLC. The plate zones were scraped off and extracted with ACN:water (1:1, v:v) and analyzed by normal phase TLC using a solvent system of acetone:ethyl acetate:water:ammonium hydroxide (50:25:24:1, v:v:v:v).

Metabolites in goat milk and tissues were identified by comparison with urine metabolites by TLC. Representative milk extracts (24 and 120 hours) were co-chromatographed to establish that the same metabolites were present in milk at different collection times. Liver and kidney extracts were also co-chromatographed and contained the same metabolites. The milk and kidney extracts were then co-chromatographed with goat urine, demonstrating that milk, kidney, and liver metabolites corresponded to the urine metabolites.

Metabolites were isolated from urine (120 hour ethyl acetate extract) using preparative TLC (described above) and the isolated residue was analyzed by TLC and HPLC. HPLC analyses were conducted using a C18 column, a gradient mobile phase of ammonium acetate pH 4.5 and ACN, and a radiodetector. Isolated urine metabolites were co-chromatographed with radiolabeled reference standards or radiolabeled metabolites isolated from rat urine (Metabolism Studies MEF.02.15 and MEF.01.13). Feces and milk extracts were also co-chromatographed with radiolabeled reference standards to identify metabolites at low levels in urine. Chemical names and structures for the reference standards are presented in Appendix I.

LC/MS analyses for confirmation of metabolite identifications were conducted using the same LC conditions discussed above, and mass spectrophotometer detection with electrospray ionization (ESI) in the positive mode.

C. RESULTS AND DISCUSSION

Sample storage intervals and conditions are reported in Table C.1. Actual extraction and analysis dates were not provided. Goat milk, kidney, and liver samples may have been stored for up to ~10 months based on the dosing date and experimental end date. No storage stability data are available to support the storage intervals and conditions of samples from the goat metabolism study. The petitioner should submit the dates of extraction, initial TLC analysis, and metabolite identification analyses. If the initial quantitative TLC analyses were conducted within 6 months of sample collection, then supporting storage stability data will not be required to support the additional analyses for metabolite identification in goat matrices.

Total radioactive residues (TRR) in goat milk and tissues are presented in Table C.2.1. TRR were determined at the in-life facility. TRR were 0.005-0.014 ppm in milk, <0.002 ppm in muscle, 0.003 ppm in omental and renal fat, 0.125 ppm in liver, and 0.090 ppm in kidney from a goat orally dosed with [^{14}C-5-pyrimidinyl]orthosulfamuron (PY label) at 10.26 ppm in the diet for 5 consecutive days. TRR were 0.004-0.016 ppm in milk, 0.007 ppm in muscle, 0.003 ppm in omental and renal fat, 0.131 ppm in liver, and 0.144 ppm in kidney from a goat orally dosed with [^{14}C-U-phenyl]orthosulfamuron (PH label) at 13.11 ppm in the diet for 5 consecutive days. TRR

were highest in liver and kidney, and <0.01 ppm in muscle and fat. The TRR were consistently low in milk but appear to plateau at 72 hours for the PY label goat and 48 hours for the PH label goat. The majority (90-97%) of the administered dose was excreted: ~50% in the feces, ~32-40% in the urine, and ~8% in the cage washes. Refer to Figures C.2.1.1. (PY label cumulative excreta), C.2.1.2. (PY label milk), C.2.1.3. (PH label cumulative excreta), and C.2.1.4 (PH label milk) for the percent of the administered dose in excreta and the TRR in goat milk over time. These figures were copied without alteration from MRID 46578962.

The distribution of the radioactivity in goat milk, kidney, and liver is summarized in Tables C.2.2.1. (PY label) and C.2.2.2. (PH label). Muscle and fat were not extracted because the TRR were <0.01 ppm. The distribution of radioactivity was similar between the two labels. The majority of the radioactivity was released with solvent extraction; ~95-103% TRR from milk with methanol, and ~93% TRR from kidney and ~48-69% TRR from liver with ACN/ammonium bicarbonate. Additional radioactivity was released from the nonextractable residues of liver with chemical or enzyme hydrolyses. Protease hydrolysis released the largest amount (45% TRR from PY label liver and 23% from PH label liver). Nonextractable residues were nondetectable in milk and ≤5% TRR (≤0.005 ppm) in kidney (both labels). Nonextractable residues remaining in liver after enzyme hydrolysis were not determined; however, based on the total radioactivity released, the nonextractable residues would be <10% TRR in liver (both labels). These procedures adequately extracted and characterized the majority of residues from goat milk and tissues. Accountabilities ranged from 92 to 100%. Residues were quantitated by TLC and were identified with goat urine metabolites using TLC, HPLC, and/or LC/MS.

The characterization and identification of residues in goat milk, kidney, and liver are summarized in Tables C.2.3.1. (PY label) and C.2.3.2. (PH label). The metabolite profile differed somewhat between the PY and PH labels, demonstrating possible cleavage of the molecule between the two rings. Parent orthosulfamuron was identified in all goat milk and tissue samples. The parent was present at low levels in milk, accounting for 8.9-11.2% TRR (0.001 ppm) in PY label milk, and 7.9-16.6% TRR (0.001-0.002 ppm) in PH label milk. The parent was identified as a major residue in tissues, accounting for 26.4% TRR (0.024 ppm) in PY label kidney, 27.8% TRR (0.037 ppm) in PH label kidney, 12.7% TRR (0.016 ppm) in PY label liver, and 20.5% TRR (0.030 ppm) in PH label liver.

Pyr-O-Sulf DOP urea was identified as the major metabolite in PY label milk and kidney accounting for 59.1-76.4% TRR (0.005-0.011 ppm) in milk and 25.7% TRR (0.023 ppm) in kidney; Pyr-O-Sulf DOP urea was also identified in liver as a minor metabolite (3.3% TRR, 0.004 ppm). Pyr-O-Sulf DOP urea only contains the parent pyrimidinyl ring and, therefore, was not identified in any PH label goat matrices. DOP urea, another pyrimidinyl ring metabolite, was only identified in liver at 3.8% TRR (0.005 ppm).

Metabolites N-desm-O-desm IR5878, N-desm IR5878, and O-desm IR5878, each with the molecule bridge between the two rings intact, were identified in both PY and PH label goat matrices. N-desm IR5878 was a significant residue accounting for 9.4-17.0% TRR (0.001-0.002 ppm) in PY label milk, 13.3% TRR (0.012 ppm) in PY label kidney, and 7.3% TRR (0.009 ppm) in PY label liver; and 21.9-34.9% TRR (0.003-0.005 ppm) in PH label milk, 17.7% TRR (0.023

ppm) in PH label kidney, and 10.9% TRR (0.016 ppm) in PH label liver. O-desm IR5878 was identified as a minor residue in all milk and tissue samples; 2.7-8.2% TRR (≤0.001 ppm) in milk, 3.4-4.0% TRR (≤0.005 ppm) in kidney, and 4.1-7.0% TRR (≤0.010 ppm) in liver (both labels). N-desm-O-desm IR5878 was also identified as a minor residue, accounting for 2.5-7.2% TRR (≤0.001 ppm) in PH label milk, 1.8-2.5% TRR (0.002 ppm) in PY and PH label kidney, and 1.4% TRR (0.002 ppm) in PY label liver. N-desm-O-desm IR5878 was not detected in PY label milk or PH label liver.

Metabolites DBS acid, N-desm DB amine, and DBS amide were only identified in PY label milk and tissues because each contains only the parent phenyl ring. DBS acid accounted for 8.6-15.2% TRR (≤0.002 ppm) in milk, 11.4% TRR (0.015 ppm) in kidney, and 4.6% TRR (0.007 ppm) in liver; N-desm DB amine accounted for 9.4-16.3% TRR (≤0.002 ppm) in milk, 13.8% TRR (0.018 ppm) in kidney, and 8.0% TRR (0.012 ppm) in liver; and DBS amide accounted for 10.6-14.1% TRR (≤0.002 ppm) in milk, 4.5% TRR (0.006 ppm) in kidney, and 3.9% TRR (0.006 ppm) in liver.

A significant amount of radioactivity in goat liver was characterized as protein-bound residues (45.5% TRR, 0.057 ppm in PY label liver and 22.6% TRR, 0.032 ppm in PH label liver). The remaining residues in goat milk and tissues were characterized as unknowns totaling 2.7-15.9% TRR in milk and 11.8-20.4% TRR in kidney and liver, with individual peaks accounting for <0.01 ppm.

Identifications of orthosulfamuron, N-desm-O-desm IR5878, N-desm IR5878, O-desm IR5878, N-desm DBS amine, and Pyr-O-Sulf DOP urea were confirmed by LC/MS analysis of the metabolites in goat urine.

There were no apparent adverse health effects of the test substance to the test goats as determined by clinical observations, measurements of body weight, feed consumption, and milk production.

C.1. Storage Stability

Samples were analyzed for total radioactivity at the in-life facility. Frozen samples were shipped within 2.3 months of collection to the analytical laboratory where samples were stored frozen (ca. -20°C) until extraction and analysis. Extraction and analysis dates were not provided. The petitioner indicated that analyses were performed by April 2003. The maximum storage interval from collection to analysis was based on the dosing date and an analysis date of April 2003. Assuming analysis was conducted on the last day of the month; milk, kidney, and liver samples may have been stored for up to 5.5 months. No data were submitted to support the storage intervals and conditions of samples from the goat metabolism study.

TABLE C.1. Summary of Storage Conditions.

Matrix	Storage Temperature (°C)	Actual Storage Duration [1]	Interval of Demonstrated Storage Stability
Goat milk, kidney, and liver	-20	≤167 days (5.5 months)	None provided

[1] Actual extraction and analysis dates were not provided. The maximum storage interval is based on the petitioner's statement that analysis was completed by April 2003; duration from harvest to analysis was calculated assuming analysis on the last day of the month.

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

TABLE C.2.1. Total Radioactive Residues (TRR) in Milk, Tissue and Excreta.

Matrix	Collection Timing	PY Label – Goat F1		PH Label – Goat F2	
		% AD	ppm	% AD	ppm
Urine	Dose Day 1-5 (0-120 hour)	39.74		32.09	
Feces	Dose Day 1-5 (0-120 hour)	50.01		49.82	
Cage Wash	Dose Day 1-5 (0-120 hour)	7.51		8.35	
Milk	8 hour	0.0	0.005	0.0	0.004
	24 hour	0.02	0.009	0.01	0.013
	32 hour	0.02	0.010	0.02	0.013
	48 hour	0.04	0.011	0.03	0.013
	56 hour	0.05	0.010	0.03	0.013
	72 hour	0.07	0.012	0.05	0.013
	80 hour	0.07	0.012	0.05	0.012
	96 hour	0.09	0.014	0.06	0.011
	104 hour	0.10	0.012	0.07	0.013
	120 hour	0.12	0.014	0.09	0.016
Muscle	At sacrifice		<0.002 (LOQ)		0.007
Fat, omental	At sacrifice		0.003		0.003
Fat, renal	At sacrifice		0.003		0.003
Kidneys	At sacrifice	0.02	0.090	0.02	0.131
Liver	At sacrifice	0.13	0.125	0.13	0.144
Bile	At sacrifice	0.05	1.811	0.06	1.337
Sum of Administered Dose (%)		97.58		90.56	

FIGURE C.2.1.1. Pharmacokinetics of Orthosulfamuron in Excreta of PY Label Goat

Animal 1F (dosed with [^{14}C-5-Pyrimidinyl]-IR5878)

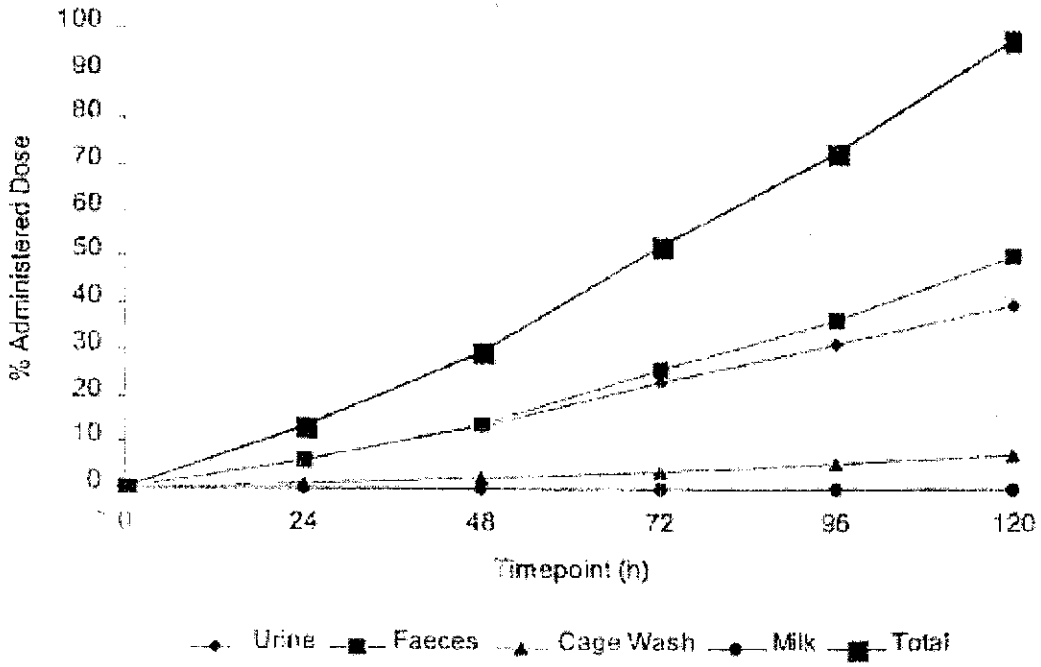


FIGURE C.2.1.2. Pharmacokinetics of Orthosulfamuron in Milk of PY Label Goat

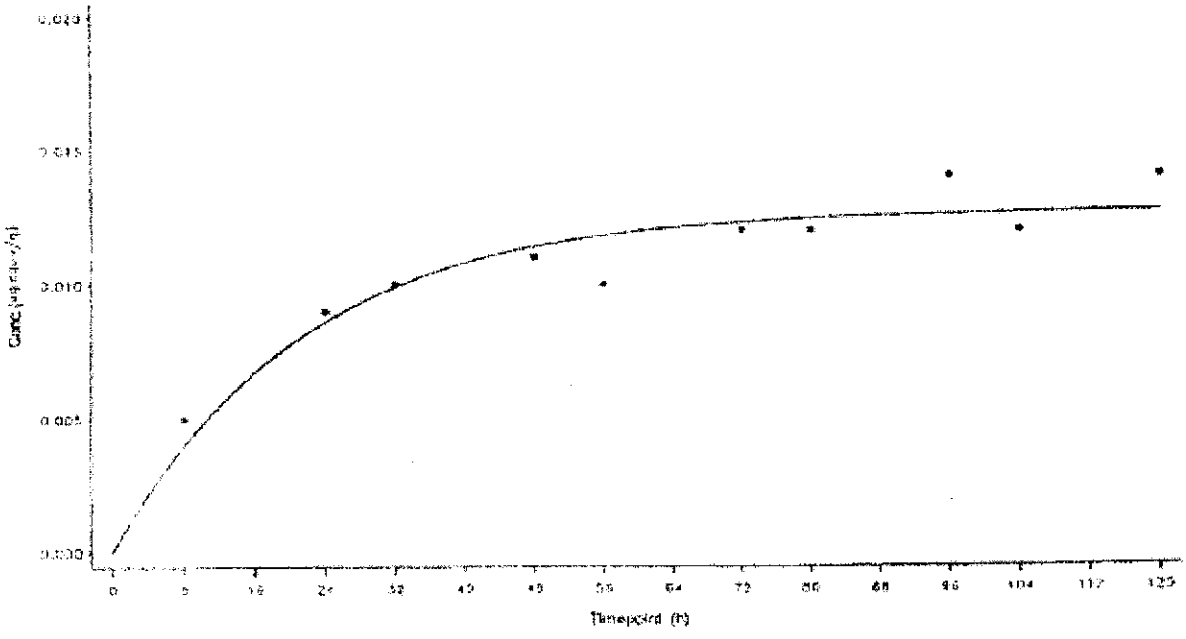


FIGURE C.2.1.3. Pharmacokinetics of Orthosulfamuron in Excreta of PH Label Goat *Animal 2F (dosed with [^{14}C-U-Phenyl]-IR5878)*

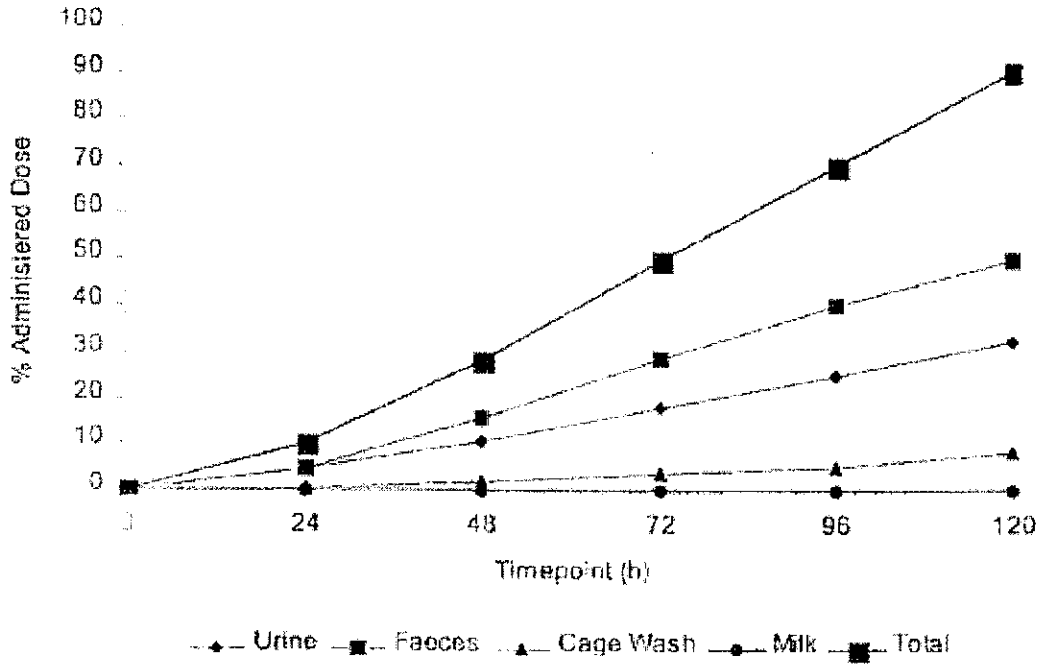


FIGURE C.2.1.4. Pharmacokinetics of Orthosulfamuron in Milk of PH Label Goat

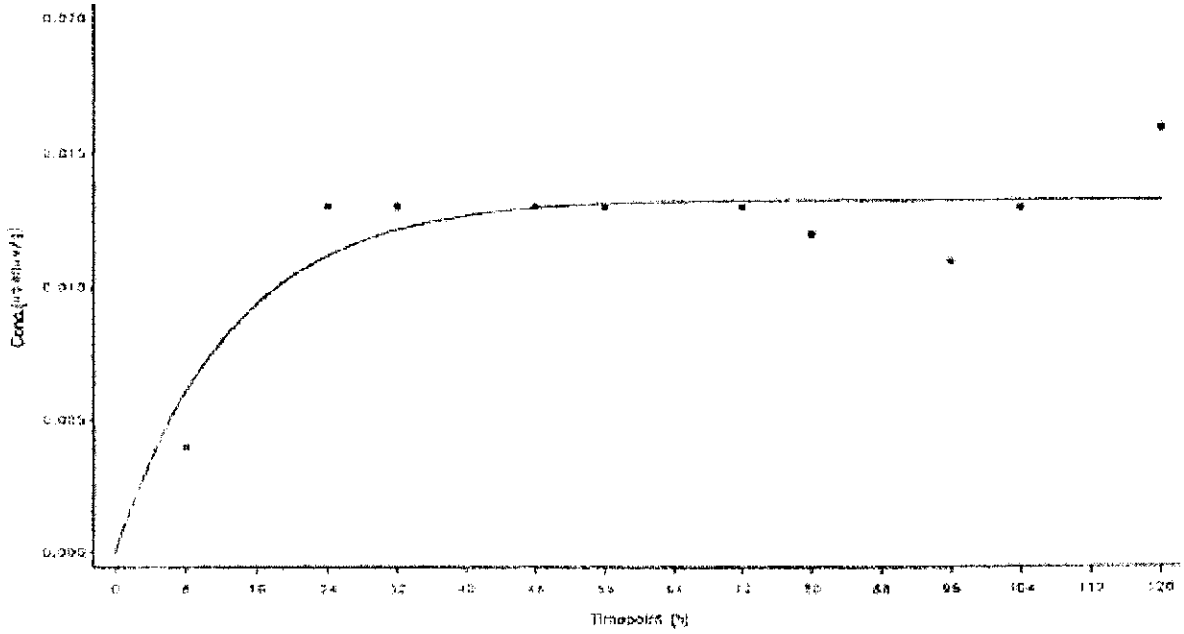


TABLE C.2.2.1. Distribution of the Parent and the Metabolites in Lactating Goat Matrices when Dosed with [Pyrimidinyl-^{14}C]Orthosulfamuron at 10.26 ppm. [1]

Metabolite Fraction	Milk, 24 hr		Milk, 72 hr		Milk, 104 hr		Milk, 120 hr		Kidney		Liver	
	TRR= 0.009 ppm		TRR= 0.012 ppm		TRR= 0.012 ppm		TRR= 0.014 ppm		TRR= 0.090 ppm		TRR= 0.125 ppm	
	%TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm
Solvent extract [2]	102.6	0.009	97.28	0.012	97.45	0.012	96.25	0.013	92.57	0.083	48.28	0.060
Orthosulfamuron	11.23	0.0010	9.23	0.0011	9.83	0.0012	8.91	0.0012	26.41	0.0238	12.72	0.0159
Pyr-O-Sulf DOP urea	59.15	0.0053	67.96	0.0082	69.23	0.0083	76.36	0.0107	25.66	0.0231	3.34	0.0042
N-desm-O-desm IR5878	--	--	--	--	--	--	--	--	2.46	0.0022	1.40	0.0017
N-desm IR5878	16.96	0.0015	13.08	0.0016	15.27	0.0018	9.37	0.0013	13.34	0.0120	7.30	0.0091
O-desm IR5878	6.71	0.0006	5.30	0.0006	2.94	0.0004	2.68	0.0004	3.95	0.0036	4.07	0.0051
DOP urea	--	--	--	--	--	--	--	--	--	--	3.77	0.0047
Unknown (pyr-1) [3]	--	--	--	--	--	--	--	--	11.50	0.0104	7.08	0.0089
Unknown (pyr-3)	5.95	0.0005	4.42	0.0005	2.71	0.0003	2.69	0.0004	4.01	0.0036	2.43	0.0030
Unknown (pyr-8)	--	--	--	--	--	--	--	--	4.90	0.0044	5.89	0.0074
Nonextractable	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	5.07	0.005	47.21	0.059
-Hexane soluble											--	--
-Surfactant soluble											8.11	0.010
-Acidic acetone soluble											5.42	0.007
-Acid (reflux) hydrolysate											31.72	0.040
-Protease hydrolysate											45.46	0.057

[1] Shading indicates that the extraction step and/or characterization analysis was not conducted for the matrix in question.
[2] Milk was extracted with methanol (the initial hexane extraction did not release any radioactivity); tissues were extracted with ACN/NH_4HCO_3.
[3] Pyr-1 was resolved as 8 peaks each present at ≤3.48% TRR (≤0.0031 ppm) in kidney and ≤2.66% TRR (≤0.0033 ppm) in liver.

TABLE C.2.2.2. Distribution of the Parent and the Metabolites in Lactating Goat Matrices when Dosed with [Phenyl-^{14}C]Orthosulfamuron at 13.11 ppm. [1]

Metabolite Fraction	Milk, 24 hr		Milk, 72 hr		Milk, 104 hr		Milk, 120 hr		Kidney		Liver	
	TRR= 0.013 ppm		TRR= 0.013 ppm		TRR= 0.013 ppm		TRR= 0.016 ppm		TRR= 0.131 ppm		TRR= 0.144 ppm	
	%TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm
Solvent extract [2]	99.42	0.013	99.70	0.013	97.81	0.013	95.43	0.015	92.73	0.121	69.23	0.100
Orthosulfamuron	16.64	0.0022	8.48	0.0011	9.31	0.0012	7.89	0.0013	27.83	0.0365	20.49	0.0295
N-desm-O-desm IR5878	2.48	0.0003	5.47	0.0007	4.96	0.0006	7.19	0.0012	1.81	0.0024	--	--
N-desm IR5878	30.15	0.0039	24.25	0.0032	34.94	0.0045	21.86	0.0035	17.72	0.0232	10.88	0.0157
O-desm IR5878	8.17	0.0011	7.10	0.0009	5.97	0.0008	8.08	0.0013	3.44	0.0045	7.00	0.0101
DBS acid	15.21	0.0020	9.71	0.0013	8.60	0.0011	12.76	0.0020	11.41	0.0149	4.62	0.0067
N-desm DB amine	9.40	0.0012	16.25	0.0021	13.85	0.0018	14.23	0.0023	13.79	0.0181	8.04	0.0116
DBS amide	10.56	0.0014	12.94	0.0017	14.09	0.0018	12.10	0.0019	4.54	0.0060	3.92	0.0057
Unknown (phe-1) [3]	--	--	6.87	0.0009	2.60	0.0003	7.44	0.0012	6.96	0.0091	9.41	0.0136
Unknown (phe-2)	7.40	0.0010	8.66	0.0011	5.94	0.0008	8.44	0.0014	4.87	0.0064	5.08	0.0073
Nonextractable	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.63	0.002	24.29	0.035
-Hexane soluble											--	--
-Surfactant soluble											3.52	0.005
-Acidic acetone soluble											3.52	0.005
-Acid (reflux) hydrolysate											19.23	0.028

TABLE C.2.2.2. Distribution of the Parent and the Metabolites in Lactating Goat Matrices when Dosed with [Phenyl-^{14}C]Orthosulfamuron at 13.11 ppm. [1]

Metabolite Fraction	Milk, 24 hr		Milk, 72 hr		Milk, 104 hr		Milk, 120 hr		Kidney		Liver	
	TRR= 0.013 ppm		TRR= 0.013 ppm		TRR= 0.013 ppm		TRR= 0.016 ppm		TRR= 0.131 ppm		TRR= 0.144 ppm	
	%TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm
-Protease hydrolysate											22.57	0.032

[1] Shading indicates that the extraction step and/or characterization analysis was not conducted for the matrix in question.
[2] Milk was extracted with methanol (the initial hexane extraction did not release any radioactivity); tissues were extracted with ACN/NH_4HCO_3.
[3] Phe-1 was resolved as 8 peaks each present at ≤1.21% TRR (≤0.0016 ppm) in kidney and ≤2.61% TRR (≤0.0038 ppm) in liver.

TABLE C.2.3.1. Summary of Characterization and Identification of Radioactive Residues in Lactating Goat Matrices when Dosed with [Pyrimidinyl-^{14}C]Orthosulfamuron at 10.26 ppm.

Compound	Milk, 24 hr		Milk, 72 hr		Milk, 104 hr		Milk, 120 hr		Kidney		Liver	
	TRR= 0.009 ppm		TRR= 0.012 ppm		TRR= 0.012 ppm		TRR= 0.014 ppm		TRR= 0.090 ppm		TRR= 0.125 ppm	
	%TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm
Orthosulfamuron	11.23	0.0010	9.23	0.0011	9.83	0.0012	8.91	0.0012	26.41	0.0238	12.72	0.0159
Pyr-O-Sulf DOP urea	59.15	0.0053	67.96	0.0082	69.23	0.0083	76.36	0.0107	25.66	0.0231	3.34	0.0042
N-desm-O-desm IR5878	--	--	--	--	--	--	--	--	2.46	0.0022	1.40	0.0017
N-desm IR5878	16.96	0.0015	13.08	0.0016	15.27	0.0018	9.37	0.0013	13.34	0.0120	7.30	0.0091
O-desm IR5878	6.71	0.0006	5.30	0.0006	2.94	0.0004	2.68	0.0004	3.95	0.0036	4.07	0.0051
DOP urea	--	--	--	--	--	--	--	--	--	--	3.77	0.0047
Unknowns	5.95	0.0005	4.42	0.0005	2.71	0.0003	2.69	0.0004	20.41	0.0184	15.40	0.0193
Protein bound	--	--	--	--	--	--	--	--	--	--	45.46	0.057
Total identified	94.05	0.0084	95.57	0.0115	97.27	0.0117	97.32	0.0136	71.82	0.0647	32.6	0.0407
Total characterized	5.95	0.0005	4.42	0.0005	2.71	0.0003	2.69	0.0004	20.41	0.0184	60.86	0.0763
Total extractable	102.6	0.009	97.28	0.012	97.45	0.012	96.25	0.013	92.57	0.083	93.74	0.117
Unextractable (PES)[1]	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	5.07	0.005	NR [3]	NR
Accountability[2]	100		100		100		93		98		>94	

[1] Residues remaining after exhaustive extractions.
[2] Accountability = (Total extractable + Total unextractable)/(TRR from combustion analysis; see TABLE C.2.1) * 100.
[3] NR = Not reported. Nonextractable residues after enzyme hydrolysis were not analyzed; however, based on the extractable and enzyme hydrolyzed residues, the remaining bound residues would be <10% TRR.

TABLE C.2.3.2. Summary of Characterization and Identification of Radioactive Residues in Lactating Goat Matrices when Dosed with [Phenyl-^{14}C]Orthosulfamuron at 13.11 ppm.

Compound	Milk, 24 hr		Milk, 72 hr		Milk, 104 hr		Milk, 120 hr		Kidney		Liver	
	TRR= 0.013 ppm		TRR= 0.013 ppm		TRR= 0.013 ppm		TRR= 0.016 ppm		TRR= 0.131 ppm		TRR= 0.144 ppm	
	%TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm
Orthosulfamuron	16.64	0.0022	8.48	0.0011	9.31	0.0012	7.89	0.0013	27.83	0.0365	20.49	0.0295
N-desm-O-desm IR5878	2.48	0.0003	5.47	0.0007	4.96	0.0006	7.19	0.0012	1.81	0.0024	--	--
N-desm IR5878	30.15	0.0039	24.25	0.0032	34.94	0.0045	21.86	0.0035	17.72	0.0232	10.88	0.0157
O-desm IR5878	8.17	0.0011	7.10	0.0009	5.97	0.0008	8.08	0.0013	3.44	0.0045	7.00	0.0101
DBS acid	15.21	0.0020	9.71	0.0013	8.60	0.0011	12.76	0.0020	11.41	0.0149	4.62	0.0067
N-desm DB amine	9.40	0.0012	16.25	0.0021	13.85	0.0018	14.23	0.0023	13.79	0.0181	8.04	0.0116
DBS amide	10.56	0.0014	12.94	0.0017	14.09	0.0018	12.10	0.0019	4.54	0.0060	3.92	0.0057

TABLE C.2.3.2. Summary of Characterization and Identification of Radioactive Residues in Lactating Goat Matrices when Dosed with [Phenyl-^{14}C]Orthosulfamuron at 13.11 ppm.

Compound	Milk, 24 hr		Milk, 72 hr		Milk, 104 hr		Milk, 120 hr		Kidney		Liver	
	TRR= 0.013 ppm		TRR= 0.013 ppm		TRR= 0.013 ppm		TRR= 0.016 ppm		TRR= 0.131 ppm		TRR= 0.144 ppm	
	%TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm
Unknowns	7.40	0.0010	15.53	0.0020	8.54	0.0011	15.88	0.0026	11.83	0.0155	14.49	0.0209
Protein bound	--	--	--	--	--	--	--	--	--	--	22.57	0.032
Total identified	92.61	0.0121	84.2	0.0110	91.72	0.0118	84.11	0.0135	80.54	0.1056	54.95	0.0793
Total characterized	7.40	0.0010	15.53	0.0020	8.54	0.0011	15.88	0.0026	11.83	0.0155	37.06	0.0529
Total extractable	99.42	0.013	99.70	0.013	97.81	0.013	95.43	0.015	92.73	0.121	91.8	0.132
Unextractable (PES)[1]	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.63	0.002	NR[3]	NR
Accountability[2]	100		100		100		94		94		>92	

[1] Residues remaining after exhaustive extractions.
[2] Accountability = (Total extractable + Total unextractable)/(TRR from combustion analysis; see TABLE C.2.1) * 100.
[3] NR = Not reported. Nonextractable residues after enzyme hydrolysis were not analyzed; however, based on the extractable and enzyme hydrolyzed residues, the remaining bound residues would be <10% TRR.

C.3. Proposed Metabolic Profile

In the goat metabolism study, the radioactivity in goat milk, muscle, and fat was negligible, and very low in the liver and kidneys. The parent was metabolized to metabolites both with the molecule bridge intact or broken between the pyrimidinyl and phenyl rings, and a significant amount of the residue in liver was eventually bound to protein. The metabolic pathway for orthosulfamuron in goats is similar to that in rats.

FIGURE C.3.1. Proposed Metabolic Profile of Orthosulfamuron in Lactating Goat.

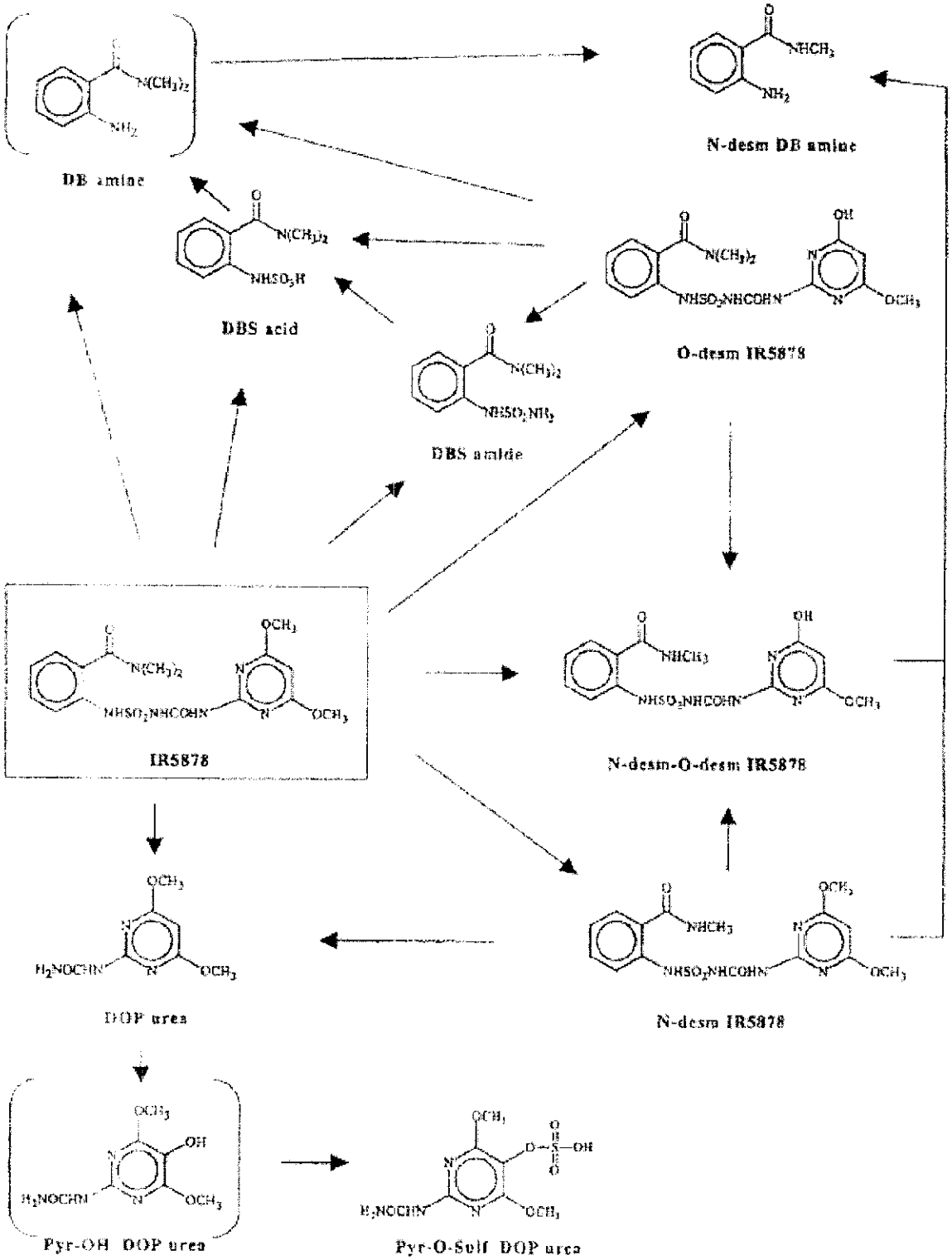


TABLE C.3.1. Identification of Compounds from Metabolism Study.

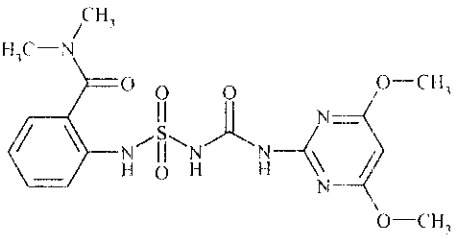
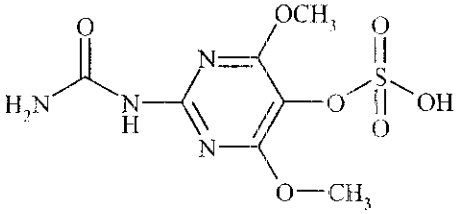
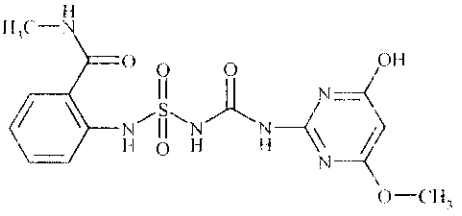
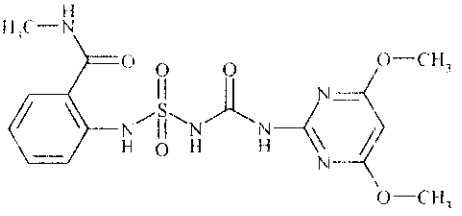
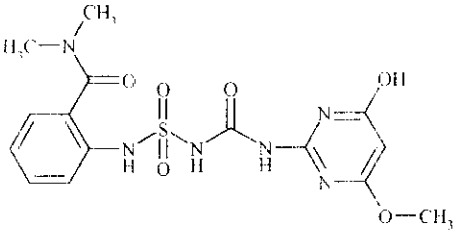
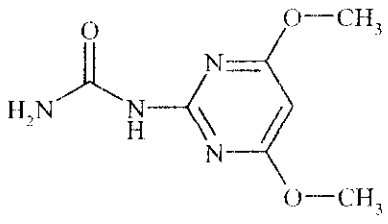
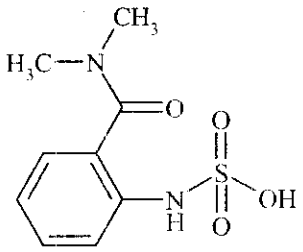
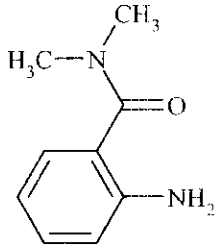
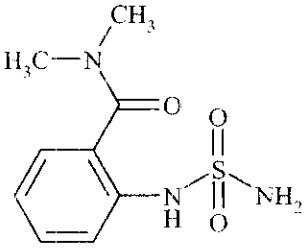
Common name/code Figure C.3.1 ID No.	Chemical name	Chemical structure
Orthosulfamuron	2-[[[[[(4,6-dimethoxy-2-pyrimidinyl)-amino]carbonyl]amino]sulfonyl]amino]-N,N-dimethylbenzamide	
Pyr-O-Sulf DOP urea	(4,6-dimethoxy-5-sulfate pyrimidin-2-yl)urea	
N-desm-O-desm IR5878	1-(4-methoxy-6-hydroxypyrimidin-2-yl)-3-[2-(methylcarbamoyl) phenylsulfamoyl]urea	
N-desm IR5878	1-(4,6-dimethoxypyrimidin-2-yl)-3-[2-(methylcarbamoyl)phenylsulfamoyl]urea	
O-desm IR5878	1-(4-methoxy-6-hydroxypyrimidin-2-yl)-3-[2-(dimethylcarbamoyl) phenylsulfamoyl]urea	
DOP urea	(4,6-dimethoxypyrimidin-2-yl)urea	

TABLE C.3.1. Identification of Compounds from Metabolism Study.		
Common name/code Figure C.3.1 ID No.	Chemical name	Chemical structure
DBS acid	2-sulfoamino-N,N-dimethylbenzamide	
N-desm DB amine	2-amino-N-methylbenzamide	
DBS amide	2-sulfamoylamino-N,N-dimethylbenzamide	

D. CONCLUSION

Total radioactive residues (TRR) ranged from 0.005 to 0.014 ppm in milk, <0.002 ppm in muscle, 0.003 ppm in omental and renal fat, 0.125 ppm in liver, and 0.090 ppm in kidney from a goat orally dosed with [^{14}C-5-pyrimidinyl]orthosulfamuron (PY label) at 10.26 ppm in the diet for 5 consecutive days. TRR were 0.004-0.016 ppm in milk, 0.007 ppm in muscle, 0.003 ppm in omental and renal fat, 0.131 ppm in liver, and 0.144 ppm in kidney from a goat orally dosed with [^{14}C-U-phenyl]orthosulfamuron (PH label) at 13.11 ppm in the diet for 5 consecutive days. Residue characterization was conducted only for representative milk, kidney, and liver as these goat matrices contained radioactivity greater than 0.01 ppm, and the distribution of the radioactivity was similar between the two labels. Following solvent extraction of residues with methanol or ACN/ammonium bicarbonate, and enzyme (protease) hydrolysis (liver only), ~92-103% of the TRR was extractable from milk, kidney, and liver.

The metabolite profile differed somewhat between the PY and PH labels, demonstrating cleavage of the molecule between the two rings. Parent, orthosulfamuron, was identified in all goat milk and tissue samples. The parent was present at low levels in milk (both labels), accounting for 7.9-16.6% TRR (≤0.002 ppm). The parent was identified as a major residue in

tissues, accounting for 26.4-27.8% TRR in kidney (both labels), 12.7% TRR in PY label liver, and 20.5% TRR in PH label liver.

Pyr-O-Sulf DOP urea was identified as the major metabolite in PY label milk and kidney accounting for 59.1-76.4% TRR in milk and 25.7% TRR in kidney. Pyr-O-Sulf DOP urea was also identified in PY label liver as a minor metabolite (3.3% TRR). Pyr-O-Sulf DOP urea only contains the parent pyrimidinyl ring and, therefore, was not identified in any PH label goat matrices. DOP urea, another pyrimidinyl ring metabolite, was only identified in liver at 3.8% TRR.

Metabolites N-desm-O-desm IR5878, N-desm IR5878, and O-desm IR5878, each with the molecule bridge between the two rings intact, were identified in both PY and PH label goat matrices. N-desm IR5878 was a significant residue accounting for 9.4-17.0% TRR in PY label milk, 13.3% TRR in PY label kidney, and 7.3% TRR in PY label liver; and 21.9-34.9% TRR in PH label milk, 17.7% TRR in PH label kidney, and 10.9% TRR in PH label liver. O-desm IR5878 was identified as a minor residue (<9% TRR) in all milk and tissue samples. N-desm-O-desm IR5878 was also identified as a minor residue (<8% TRR) in PH label milk, PY and PH label kidney, and PY label liver; N-desm-O-desm IR5878 was not detected in PY label milk or PH label liver.

Metabolites DBS acid, N-desm DB amine, and DBS amide were only identified in PY label milk and tissues because each contains only the parent phenyl ring. DBS acid accounted for 8.6-15.2% TRR in milk, 11.4% TRR in kidney, and 4.6% TRR in liver; N-desm DB amine accounted for 9.4-16.3% TRR in milk, 13.8% TRR in kidney, and 8.0% TRR in liver; and DBS amide accounted for 10.6-14.1% TRR in milk, 4.5% TRR in kidney, and 3.9% TRR in liver.

A significant amount of radioactivity in goat liver was characterized as protein bound residues (45.5% TRR in PY label liver and 22.6% TRR in PH label liver).

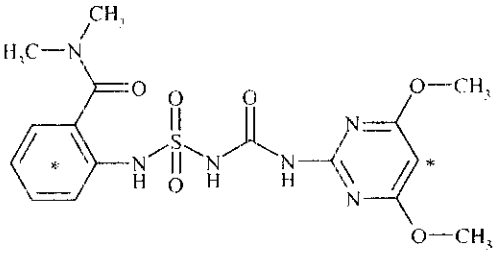
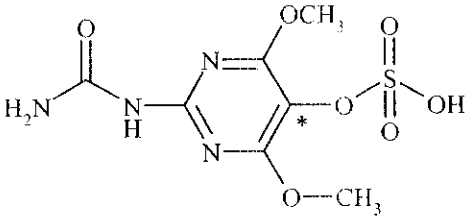
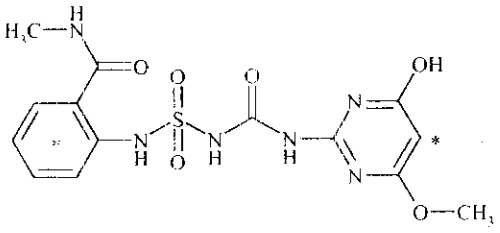
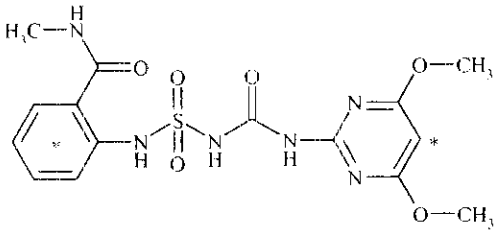
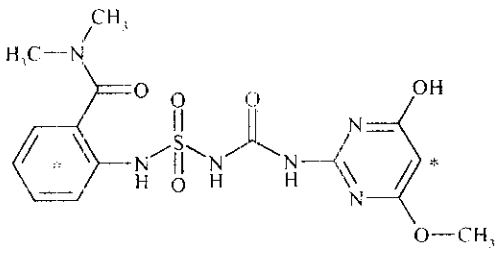
In the goat metabolism study, the radioactivity in goat milk, muscle, and fat was negligible, and very low in the liver and kidneys. The parent was metabolized to metabolites both with the molecule bridge intact or broken between the pyrimidinyl and phenyl rings, and a significant amount of the residue in liver was eventually bound to protein. The metabolic pathway for orthosulfamuron in goats is similar to that in rats.

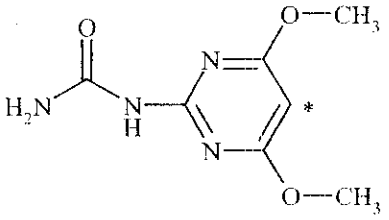
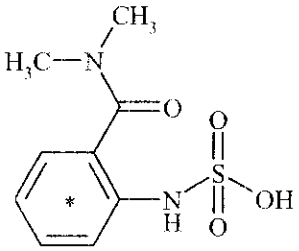
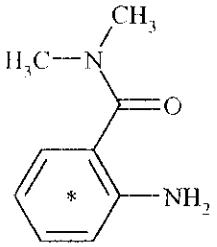
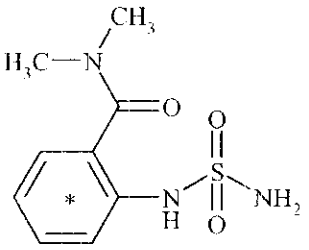
E. REFERENCES

None.

F. DOCUMENT TRACKING

Petition Number: 5F6967
DP Barcode: D319614
PC Code: 108209

APPENDIX I. Chemical Names and Structures of Reference Standards Used in Goat Metabolism Study.		
Common name; Company code	Chemical name	Chemical structure
[^{14}C]Orthosulfamuron; IR5878 (PY and PH label)	2-[[[[[(4,6-dimethoxy-2-pyrimidinyl)-amino]carbonyl]amino]sulfonyl]amino]-N,N-dimethylbenzamide	
[^{14}C]Pyr-O-SulH DOP urea (PY-label in rat urine)	(4,6-dimethoxy-5-sulfate pyrimidin-2-yl)urea	
[^{14}C]N-desm-O-desm IR5878 (PY and PH label in rat urine)	1-(4-methoxy-6-hydroxypyrimidin-2-yl)-3-[2-(methylcarbamoyl) phenylsulfamoyl]urea	
[^{14}C]N-desm IR5878 (PY and PH label in rat urine)	1-(4,6-dimethoxypyrimidin-2-yl)-3-[2-(methylcarbamoyl)phenylsulfamoyl]urea	
[^{14}C]O-desm IR5878 (PY and PH label in rat urine)	1-(4-methoxy-6-hydroxypyrimidin-2-yl)-3-[2-(dimethylcarbamoyl) phenylsulfamoyl]urea	

APPENDIX I. Chemical Names and Structures of Reference Standards Used in Goat Metabolism Study.		
Common name; Company code	Chemical name	Chemical structure
[^{14}C-5-pyrimidinyl]DOP urea	(4,6-dimethoxypyrimidin-2-yl)urea	
[^{14}C-U-phenyl]DBS acid	2-sulfoamino-N,N-dimethylbenzamide	
N-desm DB amine	2-amino-N-methylbenzamide	
[^{14}C-U-phenyl]DBS amide	2-sulfamoylamino-N,N-dimethylbenzamide	

Orthosulfamuron/IR5878/PC Code 108209/Isagro S.p.A.
DACO 7.4.3/OPPTS 860.1850/OECD IIA 6.6.3, 6.8.7 and IIIA 8.6
Confined Accumulation in Rotational Crops - Carrot, Lettuce & Wheat

Primary Evaluator	Douglas Dotson, Chemist, RAB2 D. Dotson	Date: 2/14/2007
Peer Reviewer	Dennis McNeilly, Chemist, RAB2 Dm McNeilly	Date: 2/14/2007

This DER was originally prepared under contract by Dynamac Corporation (2275 Research Boulevard, Suite 300; Rockville, MD 20850; submitted 06/12/2006). The DER has been reviewed by the Health Effects Division (HED) and revised to reflect current Office of Pesticide Programs (OPP) policies.

STUDY REPORT:

46578966 Rizzo, F.; Pizzingrilli, G. (2004) Uptake, Translocation and Metabolism of [(^{14}C)-pyrimidinyl] IR5878 in Rotated Crops of Wheat, Carrots and Lettuce. Project Number: MEF.02.06. A2_06.06/01. Unpublished study prepared by Isagro S.R.L. (Formerly Agrimont). 204 p.

EXECUTIVE SUMMARY:

Isagro S.p.A. has submitted a confined rotational crop study with [^{14}C-5-pyrimidinyl] orthosulfamuron (PY label; specific activity 112.2 µCi/mg). The radiolabeled test substance was mixed with acetonitrile/water and applied micro-dropwise to sandy loam soil in plastic pots, maintained outdoors, at a nominal rate of 0.067 lb ai/A. Carrot (root vegetable), lettuce (leafy vegetable), and wheat (small grain) were planted in the treated soil as representative rotational crops at plantback intervals (PBIs) of ca. 30, 120, and 365 days. The in-life and analytical phases of the study were conducted at Isagro Ricerca (Novara, Italy).

Total radioactive residues (TRR) accumulated at ≥0.01 ppm in carrot tops planted 42, 127, or 376 days after treatment of the soil. TRR also accumulated at ≥0.01 ppm in wheat straw rotated crops planted 29, 121, or 365 days after treatment of the soil. TRR were below 0.01 ppm in carrot root, lettuce, wheat forage, and wheat grain rotated crop matrices at all plantback intervals (PBIs). In carrot tops and wheat straw, TRR were highest at the ~120-day PBI. Residues were 0.0229 ppm at the 42-day PBI, 0.0401 ppm at the 127-day PBI, and 0.0299 ppm at the 376-day PBI in carrot tops. Residues were 0.1090 ppm at the 29-day PBI, 0.1224 ppm at the 121-day PBI, and 0.0951 ppm at the 365-day PBI in wheat straw. In the other crop matrices with very low TRR, the TRR appear to decline with later PBIs.

Rotated crop matrices with TRR >0.005 ppm were subjected to residue characterization. The majority of the radioactivity (57-89% TRR) was extracted from the rotated crop matrices using ACN/ammonium bicarbonate. Extracts containing >0.01 ppm (carrot tops and wheat straw, all PBIs), were partitioned into DCM, ethyl acetate, and aqueous soluble phases for metabolite analysis. The majority of the extractable residue in carrot tops was organosoluble, while the majority of the extractable residue in wheat straw was aqueous soluble. Nonextractable residues

in the extracted crop matrices were <0.01 ppm in carrot tops, lettuce, wheat forage, and wheat grain, and <0.04 ppm in wheat straw. Accountabilities ranged from 95% to 114%. The extraction procedures extracted sufficient residues from all PBIs. Metabolites (and the absence of the parent) were quantitated and identified by co-chromatography with the respective radiolabeled standard using normal and reverse phase TLC.

Only carrot top and wheat straw extracts contained sufficient radioactivity for metabolite analysis. Total identified residues ranged from 58% to 86% TRR. The parent, orthosulfamuron, was not identified in rotated carrot tops or wheat straw at any PBI. The major and only metabolite identified in rotated crop matrices was DOP urea. DOP urea accounted for 80.4%, 75.1%, and 86.3% TRR (0.018-0.030 ppm) in carrot tops from the 42-, 127, and 376-day PBIs, respectively. DOP urea accounted for 75.1%, 63.1%, and 58.0% TRR (0.055-0.082 ppm) in wheat straw from the 29-, 121, and 365-day PBIs, respectively. The remaining extractable residues in carrot tops were characterized as two unknowns in the ethyl acetate phase, each present at <0.01 ppm.

Nonextractable residues were further characterized as cellulose- and lignin-bound residues accounting for ~5-9% TRR in carrot tops (all PBIs), ~13-18% TRR in 29- and 121-day PBI wheat forage, ~8-13% TRR in wheat straw (all PBIs), and ~21-26% TRR in wheat grain (all PBIs). The uncharacterized nonextractable residues represented <0.03 ppm in these rotated crop matrices.

STUDY/WAIVER ACCEPTABILITY/DEFICIENCIES/CLARIFICATIONS:

Under the conditions and parameters used in the study, the confined rotational crop residue data are classified as scientifically acceptable. A confined rotational crop study with [^{14}C-U-phenyl]orthosulfamuron was separately submitted, refer to the DER for MRID 46578988. The acceptability of this study for regulatory purposes is addressed in the U.S. EPA Residue Chemistry Summary Document, D319614, D. Dotson, 2/14/2007.

COMPLIANCE:

Signed and dated Good Laboratory Practice (GLP), Quality Assurance and Data Confidentiality statements were provided. The GLP Compliance Statements cite that the study was not conducted in accordance with GLP Standards as defined by the USA EPA, but in compliance with OECD principles of GLP as defined by the Republic of Italy, as enunciated in the "Legislative Decree 27.01.1992, No. 120: activation of directive No. 88/320/CEE and No. 90/18/CEE with regard to inspection and verification of GLP.

A. BACKGROUND INFORMATION

Orthosulfamuron is a postemergence herbicide that Isagro S.p.A. is proposing for use on rice grown in the United States for the control of annual and perennial broadleaf weeds, sedges, and barnyard grass. Orthosulfamuron belongs to the sulfamoylurea class of herbicides. It acts by inhibiting the plant enzyme acetolactate synthase which is active in the biosynthesis of valine, leucine, and isoleucine.

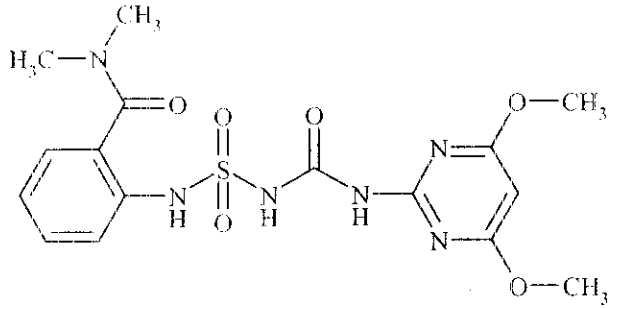
TABLE A.1. Test Compound Nomenclature.	
Compound	
Common name	Orthosulfamuron
Company experimental name	IR5878
IUPAC name	1-(4,6-dimethoxypyrimidin-2-yl)-3-[2-(dimethylcarbamoyl)phenylsulfamoyl]urea
CAS name	2-[[[[[(4,6-dimethoxy-2-pyrimidinyl)-amino]carbonyl]amino]sulfonyl]amino]-N,N-dimethylbenzamide
CAS registry number	213464-77-8
End-use product (EP)	0.51% G formulation (IR5878 0.5 GR; EPA Co. No. 80289) 51.5% WG formulation (IR5878 50 WG; EPA Co. No. 80289)

Table 2. Physicochemical Properties of the Technical Grade of Orthosulfamuron.		
Parameter	Value	Reference (MRID)
Color	White	46219004
Physical State	Fine Powder at 20°C	46219005
Odor	Odorless	46219006
pH	4.35 at 25°C (1% aqueous dispersion)	46219013
Density	1.45 g/mL at 20°C	46219008
Water solubility at 20°C	pH 4 buffer: 0.062 g/L pH 7 buffer: 0.63 g/L pH 8.5 buffer: 39 g/L	46219009
Solvent solubility at 20°C	n-heptane: 0.23 mg/L xylene: 130 mg/L acetone: 20 g/L ethyl acetate: 3.3 g/L dichloromethane: 56 g/L methanol: 8.3 g/L	Electronic communication, J. Messina to E. Kraft, 9/6/2006
Vapor pressure	1.1×10^{-4} at 20°C	46219010

Table 2. Physicochemical Properties of the Technical Grade of Orthosulfamuron.		
Parameter	Value	Reference (MRID)
Dissociation constant, pK_a	The test material becomes increasingly less soluble in water as the pH is lowered and undergoes degradation (hydrolysis) at neutral to acidic pHs. The test material is predicted to have 5 overlapping dissociation constants.	46219011
Octanol/water partition coefficient, $Log(K_{OW})$	pH 4: 2.0 pH 7: 1.3	46219012
UV/visible absorption spectrum	at pH 6.9, A=0.49 and $\varepsilon = 2.1 \times 10^4$ at 238 nm	46219001

B. EXPERIMENTAL DESIGN

Orthosulfamuron radiolabeled on the fifth carbon of the pyrimidinyl ring was prepared in acetonitrile (ACN)/water. The radiolabeled test solution was applied micro-dropwise to sandy loam soil in 18 plastic pots (36 cm wide x 78 cm long x 30 cm high) at a nominal rate of 0.067 lb ai/A. After aging for ca. 30, 120, and 365 days, representative rotated crops of carrot (root vegetable), lettuce (leafy vegetable), and wheat (small grain) were planted in the treated soil. A single pot was used for planting of carrot and lettuce at each plantback interval (PBI), and four pots were used for planting of wheat at each PBI; additional pots were treated for soil analyses. Four pots of soil were treated with ACN/water for controls: one pot each for planting of carrot and lettuce, and two pots for planting of wheat.

Plants were grown to maturity outdoors in accordance with usual and customary agricultural practices for each crop in that location. Plants were treated with chemicals and fertilizers, and irrigated as needed. Temperature, relative humidity, and sunlight exposure were recorded daily.

Immature wheat forage was collected at the tillering growth stage. Carrot roots and tops, lettuce leaves, as well as wheat grain and straw were collected at maturity.

B.1. Test Site and Crop Information

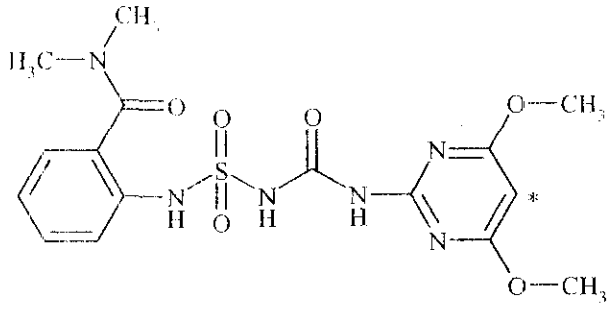
TABLE B.1.1. Test Site Information.							
Testing Environment and location	Soil characteristics						
	Type	% Sand	% Silt	% Clay	%OM	pH	CEC (meq/100 g)
Pots maintained outdoors at Isagro Ricerca Srl (Novara, Italy)	loamy sand	75.5	21.5	3	2.20	5.34	12.69

TABLE B.1.2. Crop Information.

Crop; crop group	Variety	Plantback intervals (days)	Growth stage at harvest	Harvested Matrix	Harvesting procedure
Carrot; Vegetable, root and tuber, group 1, and Vegetable, leaves of root and tuber, group 2	Mezzalunga Nantese 3	42, 127, 376	Maturity: 124 DAP	root and tops	Whole plant pulled from the pot; excess soil was brushed off; root and top were separated and the root washed with water.
Lettuce; Vegetable, leafy, except brassica, group 4	Kagraner Sommer 2	42, 127, 376	Maturity: 44 DAP	leaves	Plants cut close to the soil surface and rinsed with water.
Wheat; Grain, cereal, group 15, and Grain, cereal, forage, fodder, and straw, group 16	Gemini	29, 121, 365	Immature (tillering stage): 66 DAP Maturity: 118 DAP	forage grain and straw	Plants cut close to the soil surface; mature plants were separated into grain and straw.

B.2. Test Materials

TABLE B.2.1. Test Material Characteristics.

Chemical structure	
Radiolabel position	[^{14}C-5-pyrimidinyl]orthosulfamuron (PY-label)
Lot No.	180
Purity	>97% (TLC and HPLC)
Specific activity	4.152 MBq/mg (112.217 µCi/mg)

B.3. Study Use Pattern

TABLE B.3.1. Use Pattern Information.

Chemical name	[^{14}C-5-pyrimidinyl]orthosulfamuron
Application method	The radiolabeled test material was prepared in ACN:water (3:7, v:v) on the day of application and applied micro-dropwise to the bare soil.
Application rate	75.13-75.47 g ai/ha Mean of applications made to treated pots = 0.067 lb ai/A (75.25 g ai/ha)
Number of application (s)	One
Timing of application (s)	Preplant to bare soil
DAT[1] (days)	Target plantback intervals of 30, 120, and 365 days; refer to Table B.1.2. for actual plantback intervals

[1] DAT = Days after treatment

B.4. Identification/Characterization of Residues

B.4.1. Sample Handling and Preparation

All rotated crop samples were immediately stored frozen (-20°C) until they were processed. For processing, whole crop samples were cut into pieces and finely ground with dry ice. The homogenized samples were frozen until extraction and analysis.

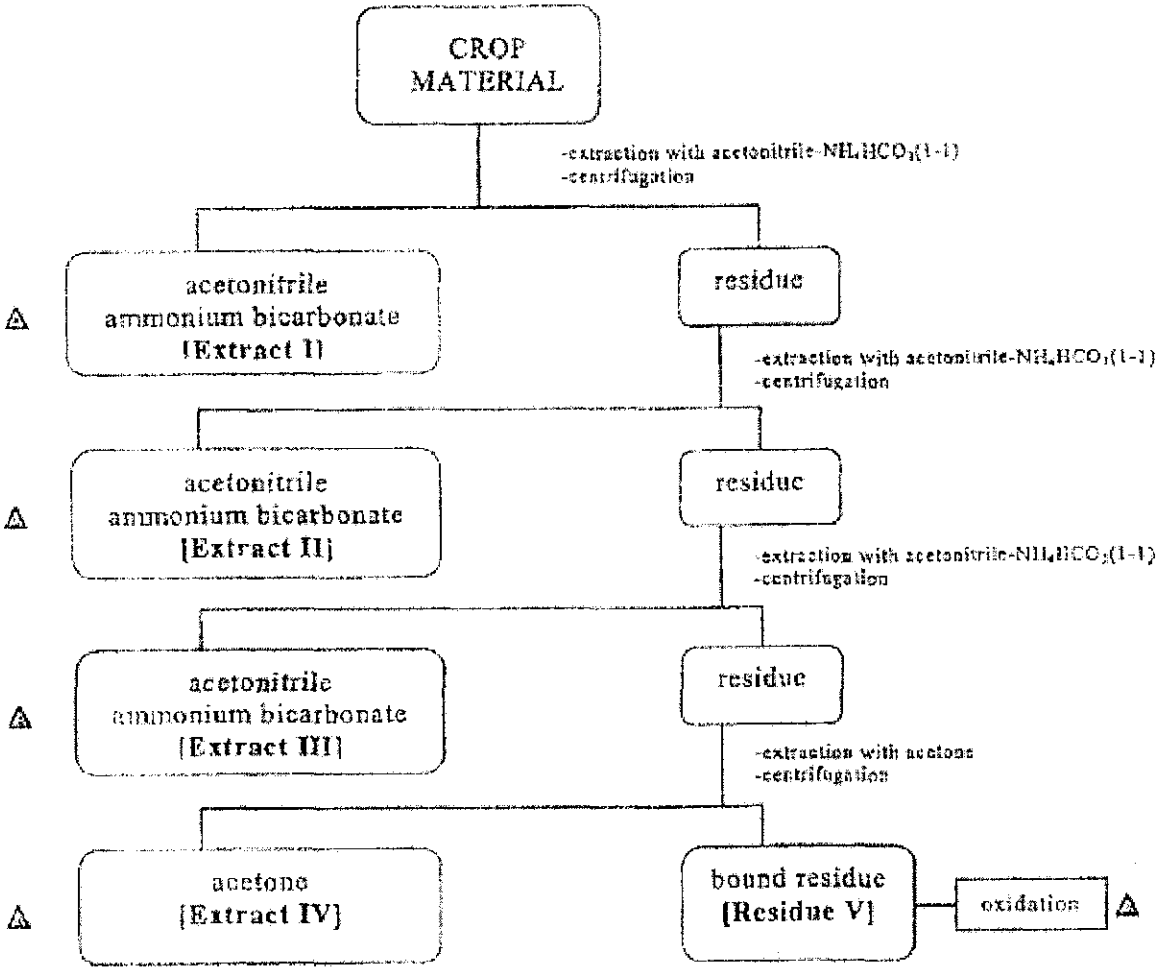
Rotational crop commodities with a TRR >0.005 ppm were subjected to extraction procedures. Carrot root from all PBIs, 127- and 376-PBI lettuce, and 365-PBI wheat forage were not extracted. A brief discussion of the extraction processes follows.

An aliquot of the rotated crop matrix was extracted three times with ACN:50mM ammonium bicarbonate (NH_4HCO_3; 5:5, v:v), then once with acetone, and centrifuged. Each supernatant was separately collected and brought to volume with additional extraction solvent for radioassay. The ACN extracts of carrot leaves and wheat straw (from all PBIs, with >0.01 ppm extracted residues), were sequentially partitioned with dichloromethane (DCM) and ethyl acetate, after concentration to aqueous. The resulting DCM, ethyl acetate, and aqueous phases were brought to volume with the respective solvent and reserved for TLC analysis.

Nonextractable residues (>10% TRR) of carrot leaves, wheat straw, and wheat grain (all PBIs), and 29-PBI and 121-PBI wheat forage were fractionated to determine incorporation into natural components. The bound residue was refluxed with 5% sodium hydroxide (110°C for 3 hours) and centrifuged to isolate the cellulose fraction (solids) from the soluble fraction. The soluble fraction was then acidified with 6 N HCl (pH 2) to precipitate the lignin fraction.

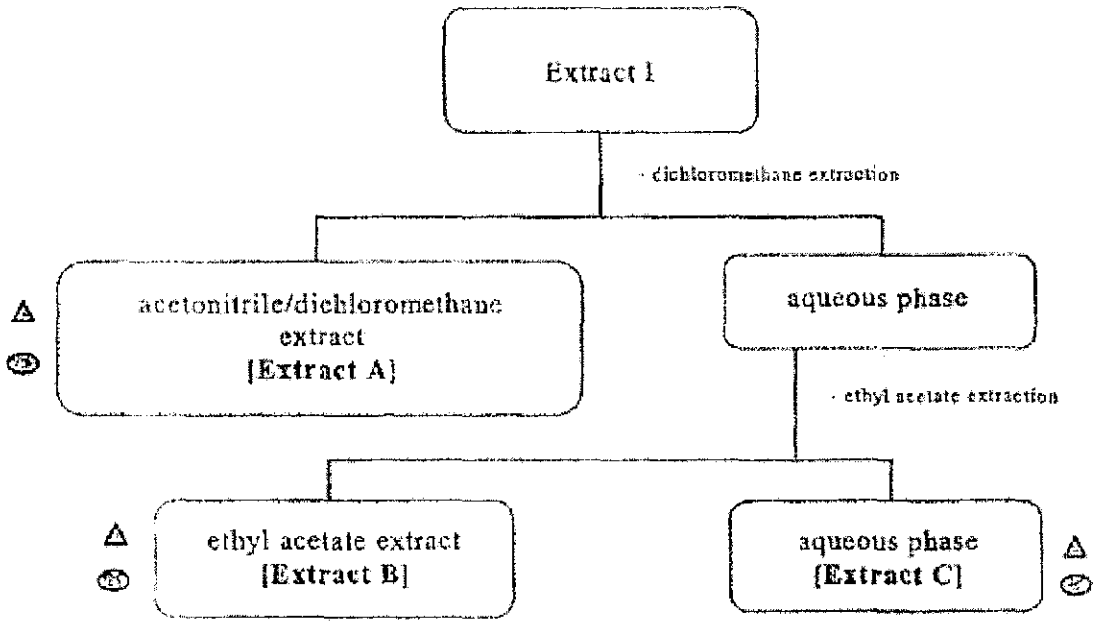
The extraction flowcharts are presented in Figures B.4.1.1 (crops), B.4.1.2 (ACN extract), and B.4.1.3 (nonextractable residues). These flowcharts were copied from MRID 46578966 without alteration.

Figure B.4.1.1. Extraction Scheme for Rotated Crop Matrices.



LSC analysis

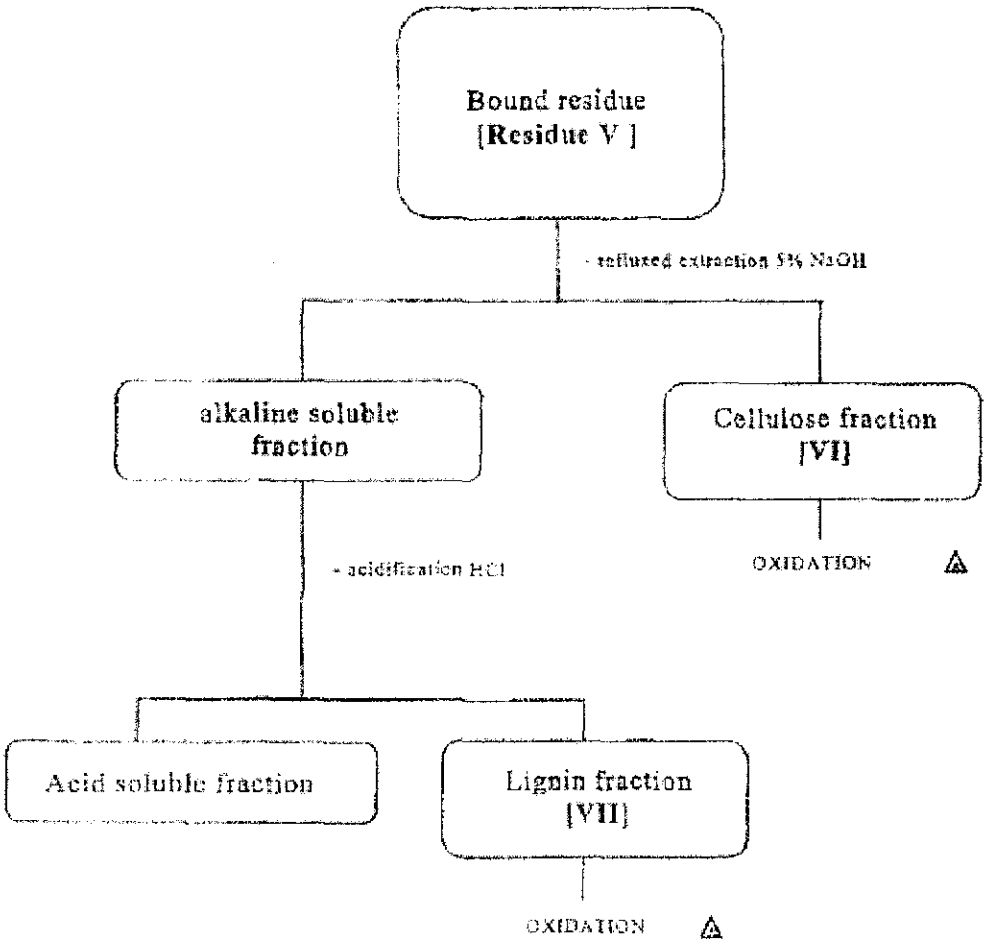
Figure B.4.1.2. Partitioning Scheme for the ACN Extract I.



Δ LSC analysis

TLC analysis

Figure B.4.1.3. Fractionation Scheme for Nonextractable Residues.



B.4.2. Analytical Methodology

Total radioactive residues (TRR) in rotational crop matrices were determined by combustion/LSC of the finely ground sample. Aliquots of ground carrot root and leaves, lettuce, and wheat forage were freeze-dried to remove water prior to combustion. Extracts and hydrolysates were radioassayed by LSC and nonextractable residues were radioassayed by combustion/LSC. The limit of determination for LSC determinations was twice the background counting rate.

The DCM, ethyl acetate, and aqueous phases of the ACN:NH_4HCO_3 extract of carrot leaves and wheat straw were each subjected to TLC analysis for identification and quantitation of metabolites. Normal phase TLC analyses were conducted using silica gel 60 F_{254} plates and a solvent system of chloroform:methanol:ammonium hydroxide (70:27:3, v:v:v). Reverse phase TLC analyses were conducted using RP-18 F_{254S} plates and a solvent system of ACN:water (92:8, v:v). Samples were analyzed by both normal and reverse phase TLC systems, but normal

phase TLC was used for quantitation of residues because of better separation of the ^{14}C-compounds. TLC plates were evaluated using radioimaging. Luminescence was detected by a photo multiplier. Metabolites were identified by co-chromatography with radiolabeled reference standards (parent and DOP urea) using both TLC systems. Chemical names and structures for the reference standards are presented in Appendix I

C. RESULTS AND DISCUSSION

The storage conditions and intervals for rotational crop samples are presented in Table C.1. All samples were analyzed for TRR within 2 months of harvest. Samples which were extracted and/or analyzed by TLC were done so within 6 months of harvest, therefore, no storage stability data are required to support the sample storage intervals and conditions of the confined rotational crop study.

TRR in rotational crops are reported in Table C.2.1. Total radioactive residues (TRR) accumulated at ≥0.01 ppm in carrot tops planted 42, 127, or 376 days after treatment of the soil. TRR also accumulated at ≥0.01 ppm in wheat straw rotated crops planted 29, 121, or 365 days after treatment of the soil. TRR were below 0.01 ppm in carrot root, lettuce, wheat forage, and wheat grain rotated crop matrices at all plantback intervals (PBIs). In carrot tops and wheat straw, TRR were highest at the ~120-day PBI. Residues were 0.0229 ppm at the 42-day PBI, 0.0401 ppm at the 127-day PBI, and 0.0299 ppm at the 376-day PBI in carrot tops. Residues were 0.1090 ppm at the 29-day PBI, 0.1224 ppm at the 121-day PBI, and 0.0951 ppm at the 365-day PBI in wheat straw. In the other crop matrices with very low TRR, the TRR appear to decline with later PBIs.

Rotated crop matrices with TRR >0.005 ppm were extracted; the extraction profiles and distribution of the radioactivity in these rotational crops are presented in Tables C.2.2.1 (carrot tops, all PBIs), C.2.2.2. (lettuce, 42-day PBI and wheat forage, 29- and 121-day PBIs), C.2.2.3. (wheat straw, all PBIs) and C.2.2.4. (wheat grain, all PBIs). The majority of the radioactivity (57-89% TRR) was extracted from the rotated crop matrices using ACN/ammonium bicarbonate. Extracts containing >0.01 ppm (carrot tops and wheat straw, all PBIs) were partitioned into DCM, ethyl acetate, and aqueous soluble phases for metabolite analysis. The majority of the extractable residue in carrot tops was organosoluble, while the majority of the extractable residue in wheat straw was aqueous soluble. Nonextractable residues in the extracted crop matrices were <0.01 ppm in carrot tops, lettuce, wheat forage, and wheat grain, and <0.04 ppm in wheat straw; Accountabilities ranged from 95% to 114%. The extraction procedures extracted sufficient residues from all PBIs.

The characterization and identification of residues in rotational crop matrices which were chromatographically analyzed are summarized in Tables C.2.3.1. (carrot tops, all PBIs) and C.2.3.2. (wheat straw, all PBIs). Total identified residues ranged from 58% to 86% TRR in rotated carrot tops and wheat straw. The parent, orthosulfamuron, was not identified in rotated carrot tops or wheat straw at any PBI.

The major and only metabolite identified was DOP urea. The identification of DOP urea and the absence of the parent were confirmed by co-chromatography with the respective radiolabeled standard using normal and reverse phase TLC. DOP urea accounted for 80.4%, 75.1%, and 86.3% TRR (0.018-0.030 ppm) in carrot tops from the 42-, 127, and 376-day PBIs, respectively. DOP urea accounted for 75.1%, 63.1%, and 58.0% TRR (0.055-0.082 ppm) in wheat straw from the 29-, 121, and 365-day PBIs, respectively. The remaining extractable residues in carrot tops were characterized as two unknowns in the ethyl acetate phase, each present at <0.01 ppm.

All extracted crop matrices with nonextractable residues >10% TRR were treated with base and acid to precipitate cellulose- and lignin-bound residues. Cellulose and lignin bound residues accounted for ~5-9% TRR in carrot tops (all PBIs), ~13-18% TRR in 29- and 121-day PBI wheat forage, ~8-13% TRR in wheat straw (all PBIs), ~21-26% TRR in wheat grain (all PBIs), and the uncharacterized nonextractable residues represented <0.03 ppm in these rotated crop matrices.

C.1. Storage Stability

Samples of rotated crop matrices were stored frozen (-20°C) after harvest. All samples were ground within 4-36 days of harvest, and TRR were determined within 9-28 days after grinding. Only the carrot top and wheat straw samples were chromatographically analyzed, and analysis occurred within ~6 months of harvest. Extracts were stored at 4°C until analysis.

TABLE C.1. Summary of Storage Conditions.

Matrix	Plantback interval (days)	Storage Temp. (°C)	Actual Storage Duration [1]	Interval of Demonstrated Storage Stability
Carrot root	42, 127, 376	-20	18 days (TRR analysis)	None required.
Carrot tops	42, 127, 376	-20	45-56 days (<2 months to TLC analysis)	
Lettuce	42, 127, 376	-20	147-151 days (<5 months to extraction)	
Wheat, forage	29, 121, 376	-20	179-183 days (~6 months to extraction)	
Wheat, straw	29, 121, 376	-20	117-119 days (<4 months to TLC analysis)	
Wheat, grain	29, 121, 376	-20	127-131 days (<4.5 months to extraction)	

[1] Only the carrot tops and wheat straw extracts were analyzed; these samples were analyzed within 17-96 days of extraction

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

TABLE C.2.1. Total Radioactive Residues (TRR) in Rotated Crop Matrices and the Treated Soil.

Matrix	Plantback interval (days)	PY Label (ppm)
Carrot, root	42	0.0010
	127	0.0007
	376	0.0008
Carrot, tops	42	0.0229
	127	0.0401
	376	0.0299
Lettuce, leaves	42	0.0056
	127	0.0035

TABLE C.2.1. Total Radioactive Residues (TRR) in Rotated Crop Matrices and the Treated Soil.		
Matrix	Plantback interval (days)	PY Label (ppm)
	376	0.0006
Wheat, forage	29	0.0063
	121	0.0060
	365	0.0039
Wheat, straw	29	0.1090
	121	0.1224
	365	0.0951
Wheat, grain	29	0.0079
	121	0.0076
	365	0.0054
Soil [1]	0	0.080
	29 (wheat planting)	0.024
	42 (carrot and lettuce planting)	0.029
	121 (wheat planting)	0.022
	127 (carrot and lettuce planting)	0.037
	365 (wheat planting)	0.010
	376 (carrot and lettuce planting)	0.036

[1] The petitioner reported the mean values obtained from all analyses on soils with the same aging period. TRR were calculated by the summation of extractable (acetone) and nonextractable residues in soil

TABLE C.2.2.1. Distribution of the Parent and the Metabolites in Rotational Carrot Tops Following Application of [Pyrimidinyl-^{14}C]Orthosulfamuron at 0.067 lb ai/A. [1]						
Metabolite Fraction	Carrot, tops 42-day PBI		Carrot, tops 127-day PBI		Carrot, tops 376-day PBI	
	TRR = 0.0229 ppm		TRR = 0.0401 ppm		TRR = 0.0299 ppm	
	%TRR	ppm	%TRR	ppm	%TRR	ppm
ACN/NH_4HCO_3 Extract I	81.22	0.0186	76.06	0.0305	*87.29*	0.0261
-DCM phase	54.15	0.0124	49.62	0.0199	52.51	0.0157
DOP urea	54.15	0.0124	49.62	0.0199	52.51	0.0157
-Ethyl acetate phase	9.17	0.0021	8.73	0.0035	11.71	0.0035
DOP urea	*8.30*	0.0019	*7.73*	0.0031	*10.37*	0.0031
Unknown 1	*0.87*	0.0002	*0.75*	0.0003	*0.67*	0.0002
Unknown 2	*0.17*	0.00004	*0.25*	0.0001	*0.33*	0.0001
-Aqueous phase	17.90	0.0041	17.71	0.0071	23.41	0.0070
DOP urea	17.90	0.0041	17.71	0.0071	23.41	0.0070
Nonextractable	32.75	0.0075	22.69	0.0091	15.38	0.0046
-Cellulose solic	2.62	0.0006	2.74	0.0011	5.69	0.0017
-Lignin precipitate	2.62	0.0006	2.74	0.0011	3.34	0.0010

[1] Percent TRR values in *italics* were calculated by the study reviewer from the reported ppm value.

TABLE C.2.2.2. Distribution of the Parent and the Metabolites in Rotational Lettuce and Wheat Forage Following Application of [Pyrimidinyl-^{14}C]Orthosulfamuron at 0.067 lb ai/A. [1]

Metabolite Fraction	Lettuce 42-day PBI		Wheat, forage 29-day PBI		Wheat, forage 121-day PBI	
	TRR = 0.0056 ppm		TRR = 0.0063 ppm		TRR = 0.0060 ppm	
	%TRR	ppm	%TRR	ppm	%TRR	ppm
ACN/NH_4HCO_3 Extract I	89.29	0.0050	71.43	0.0045	56.67	0.0034
Nonextractable	3.57	0.0002	33.33	0.0021	41.67	0.0025
-Cellulose solid			7.94	0.0005	11.67	0.0007
-Lignin precipitate			4.76	0.0003	6.67	0.0004

[1] Shading indicates that the extraction step and/or characterization analysis was not conducted for the matrix in question.

TABLE C.2.2.3. Distribution of the Parent and the Metabolites in Rotational Wheat Straw Following *Application of [Pyrimidinyl-^{14}C]Orthosulfamuron at 0.067 lb ai/A.*

Metabolite Fraction	Wheat, straw 29-day PBI		Wheat, straw 121-day PBI		Wheat, straw 365-day PBI	
	TRR = 0.1090 ppm		TRR = 0.1224 ppm		TRR = 0.0951 ppm	
	%TRR	ppm	%TRR	ppm	%TRR	ppm
ACN/NH_4HCO_3 Extracts I-III	75.14	0.0819	63.07	0.0772	58.04	0.0552
-DCM phase	31.56	0.0344	22.38	0.0274	22.50	0.0214
DOP urea	31.56	0.0344	22.38	0.0274	22.50	0.0214
-Ethyl acetate phase	--	<0.0022	--	<0.0013	--	<0.0011
-Aqueous phase	43.58	0.0475	40.69	0.0498	35.54	0.0338
DOP urea	43.58	0.0475	40.69	0.0498	35.54	0.0338
Nonextractable	20.18	0.0220	31.45	0.0385	40.38	0.0384
-Cellulose solid	7.34	0.0080	3.51	0.0043	9.46	0.0090
-Lignin precipitate	5.78	0.0063	4.82	0.0059	1.26	0.0012

TABLE C.2.2.4. Distribution of the Parent and the Metabolites in Rotational Wheat Grain Following Application of [Pyrimidinyl-^{14}C]Orthosulfamuron at 0.067 lb ai/A.

Metabolite Fraction	Wheat, grain 29-day PBI		Wheat, grain 121-day PBI		Wheat, grain 365-day PBI	
	TRR = 0.0079 ppm		TRR = 0.0076 ppm		TRR = 0.0054 ppm	
	%TRR	ppm	%TRR	ppm	%TRR	ppm
ACN/NH_4HCO_3 Extract I	60.76	0.0048	63.16	0.0048	61.11	0.0033
Nonextractable	44.30	0.0035	46.05	0.0035	46.30	0.0025
-Cellulose solid	6.33	0.0005	6.58	0.0005	7.41	0.0004
-Lignin precipitate	17.72	0.0014	14.47	0.0011	18.52	0.0010

TABLE C.2.3.1. Summary of Characterization and Identification of Radioactive Residues in Rotational Carrot Tops Following Application of [Pyrimidinyl-^{14}C]Orthosulfamuron at 0.067 lb ai/A.

Compound	Carrot, tops 42-day PBI		Carrot, tops 127-day PBI		Carrot, tops 376-day PBI	
	TRR = 0.0229 ppm		TRR = 0.0401 ppm		TRR = 0.0299 ppm	
	% TRR	ppm	% TRR	ppm	% TRR	ppm
DOP urea	80.35	0.0184	75.06	0.0301	86.29	0.0258
Unknowns	1.04	0.0002	1.00	0.0004	1.00	0.0003
Cellulose	2.62	0.0006	2.74	0.0011	5.69	0.0017
Lignin	2.62	0.0006	2.74	0.0011	3.34	0.0010
Total identified	80.35	0.0184	75.06	0.0301	86.29	0.0258
Total characterized	6.28	0.0014	6.48	0.0026	10.03	0.0030
Total extractable	81.22	0.0186	76.06	0.0305	87.29	0.0261
Unextractable (PES)[1]	32.75	0.0075	22.69	0.0091	15.38	0.0046
Accountability[2]	114		98.8		103	

[1] Residues remaining after exhaustive extractions. Nonextractable residues were further characterized as cellulose and lignin bound residues precipitated under basic and acidic conditions (as reported in the table). Uncharacterized nonextractable residues were <0.01 ppm in all matrices.
[2] Accountability = (Total extractable + Total unextractable)/(TRR from combustion analysis; see TABLE C.2.1) * 100.

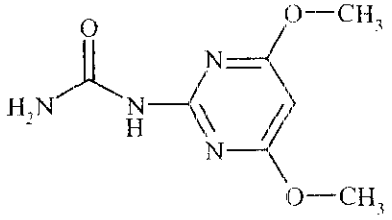
TABLE C.2.3.2. Summary of Characterization and Identification of Radioactive Residues in Rotational Wheat Straw Following Application of [Pyrimidinyl-^{14}C]Orthosulfamuron at 0.067 lb ai/A.

Compound	Wheat, straw 29-day PBI		Wheat, straw 121-day PBI		Wheat, straw 365-day PBI	
	TRR = 0.1090 ppm		TRR = 0.1224 ppm		TRR = 0.0951 ppm	
	% TRR	ppm	% TRR	ppm	% TRR	ppm
DOP urea	75.14	0.0819	63.07	0.0772	58.04	0.0552
Cellulose	7.34	0.0080	3.51	0.0043	9.46	0.0090
Lignin	5.78	0.0063	4.82	0.0059	1.26	0.0012
Total identified	75.14	0.0819	63.07	0.0772	58.04	0.0552
Total characterized	13.12	0.0143	8.33	0.0102	10.72	0.0102
Total extractable	75.14	0.0819	63.07	0.0772	58.04	0.0552
Unextractable (PES)[1]	20.18	0.0220	31.45	0.0385	40.38	0.0384
Accountability[2]	95.3		94.5		98.4	

[1] Residues remaining after exhaustive extractions. Nonextractable residues were further characterized as cellulose and lignin bound residues precipitated under basic and acidic conditions (as reported in the table); uncharacterized nonextractable residues were <0.03 ppm in all matrices.
[2] Accountability = (Total extractable + Total unextractable)/(TRR from combustion analysis; see TABLE C.2.1) * 100.

C.3. Proposed Metabolic Profile

The petitioner submitted a metabolic pathway for orthosulfamuron residues in rotational crops based on the [^{14}C-pyrimidinyl] and [^{14}C-phenyl]orthosulfamuron confined rotational crop studies. Refer to the DER for MRID 46578988 for a schematic of this pathway.

TABLE C.3.1. Identification of Compounds from the Confined Rotational Crop Study.		
Common name/code Figure C.3.1 ID No.	Chemical name	Chemical structure
DOP urea	(4,6-dimethoxypyrimidin-2-yl)urea	

D. CONCLUSION

TRR accumulated at ≥0.01 ppm in carrot tops planted 42, 127, or 376 days after treatment of the soil. TRR also accumulated at ≥0.01 ppm in wheat straw rotated crops planted 29, 121, or 365 days after treatment of the soil. TRR were below 0.01 ppm in carrot root, lettuce, wheat forage, and wheat grain rotated crop matrices at all plantback intervals (PBIs). In carrot tops and wheat straw, TRR were highest at the ~120-day PBI. Residues were 0.0229 ppm at the 42-day PBI, 0.0401 ppm at the 127-day PBI, and 0.0299 ppm at the 376-day PBI in carrot tops. Residues were 0.1090 ppm at the 29-day PBI, 0.1224 ppm at the 121-day PBI, and 0.0951 ppm at the 365-day PBI in wheat straw. In the other crop matrices with very low TRR, the TRR appear to decline with later PBIs.

The majority of the radioactivity (57-89% TRR) was extracted from the rotated crop matrices (with TRR >0.005 ppm) using ACN/ammonium bicarbonate. Extracts containing >0.01 ppm (carrot tops and wheat straw, all PBIs), were partitioned into DCM, ethyl acetate and aqueous soluble phases for metabolite analysis. The majority of the extractable residue in carrot tops was organosoluble, while the majority of the extractable residue in wheat straw was aqueous soluble. Nonextractable residues in the extracted crop matrices were <0.01 ppm in carrot tops, lettuce, wheat forage, and wheat grain, and <0.04 ppm in wheat straw.

Only carrot top and wheat straw extracts contained sufficient radioactivity for metabolite analysis. Total identified residues ranged from 58% to 86% TRR and the parent, orthosulfamuron, was not identified in rotated carrot tops or wheat straw at any PBI.

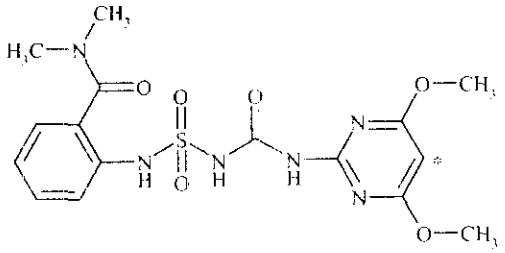
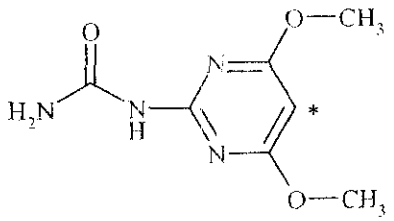
The major and only metabolite identified was DOP urea. DOP urea accounted for 75.1-86.3% TRR in carrot tops, and 58.0-75.1% TRR in wheat straw from all plantback intervals. Nonextractable residues were characterized as cellulose- and lignin-bound residues (~5-26% TRR) in carrot tops (all PBIs), 29- and 121-day PBI wheat forage, wheat straw (all PBIs), and wheat grain (all PBIs). Uncharacterized nonextractable residues represented <0.03 ppm in these rotated crop matrices.

E. REFERENCES

None.

F. DOCUMENT TRACKING

Petition Number: 5F6967
DP Barcode: D319614
PC Code: 108209

APPENDIX I. Chemical Names and Structures of Reference Standards Used in Confined Rotational Crop Study.		
Common name; Company code	Chemical name	Chemical structure
[^{14}C-pyrimidinyl]orthosulfamuron; IR5878	2-[[[[[(4,6-dimethoxy-2-pyrimidinyl)-amino]carbonyl]amino]sulfonyl]amino]-N,N-dimethylbenzamide	
[^{14}C-pyrimidinyl]DOP urea	(4,6-dimethoxypyrimidin-2-yl)urea	

Primary Evaluator Douglas Dotson, Chemist, RAB2 D. Dotson Date: 2/14/2007

Peer Reviewer Dennis McNeilly, Chemist, RAB2 Dennis McNeilly Date: 2/14/2007

This DER was originally prepared under contract by Dynamac Corporation (2275 Research Boulevard, Suite 300; Rockville, MD 20850; submitted 06/12/2006). The DER has been reviewed by the Health Effects Division (HED) and revised to reflect current Office of Pesticide Programs (OPP) policies.

STUDY REPORT:

46578961 Rizzo, F.; Pizzingrilli, G. (2004) Metabolism of ^{14}C-IR5878 in Rice. Project Number: MEF.03.08. Unpublished study prepared by Isagro Ricerca Srl. 206 p.

EXECUTIVE SUMMARY:

Isagro S.p.A. has submitted a study investigating the metabolism of [^{14}C-5-pyrimidinyl] orthosulfamuron (PY label; specific activity 120.3 µCi/mg) and [^{14}C-U-phenyl]orthosulfamuron (PH label; specific activity 198.9 µCi/mg) in rice. Each radiolabeled test substance was prepared as a WG formulation blank, suspended in water with a nonionic surfactant. It was applied as a single foliar application to rice plants at the 2-leaf growth stage (grown outdoors in minipaddies) at 0.070 lb ai/A (PY label) or 0.066 lb ai/A (PH label). Separate plots were treated at an exaggerated rate in case additional material was required for metabolite identification; these samples were not used in the study. Samples of mature whole grain (kernel plus hulls) and straw were collected 112 days following treatment. Subsamples of whole grain were separated into husked rice and hulls to investigate residue concentration. The in-life and analytical phases of the study were conducted by Isagro Ricerca Srl (Novara, Italy).

Total radioactive residues (TRR) in rice matrices, determined by combustion/LSC, were 0.0632 ppm and 0.0898 ppm in samples of rice whole grain and straw, respectively, treated with PY-label orthosulfamuron. TRR were 0.0368 ppm and 0.1101 ppm in samples of rice whole grain and straw, respectively, treated with PH-label orthosulfamuron. In both PY- and PH-label grain, TRR appear to be concentrated in the hull of rice grain. TRR were 0.0584 ppm and 0.1128 ppm in PY-label husked rice and hulls, and 0.0330 ppm and 0.0502 ppm in PH-label husked rice and hulls, respectively.

The distribution of radioactivity was similar between the two labels. Samples of rice grain were first extracted with hexane, which only released ~1-2% TRR. A relatively large amount of radioactivity was released from both rice grain (~21-29% TRR; 0.008-0.018 ppm) and straw (~48-49% TRR; 0.043-0.054 ppm) with solvent extraction (ACN/NH_4OH). Partitioning of the solvent extract separated the organic and aqueous soluble residues, respectively as 13.0% TRR (0.008 ppm) and 15.7% TRR (0.010 pm) in PY-label grain; 5.4% TRR (0.002 ppm) and 16.0%

TRR (0.006 ppm) in PH-label grain; 21.2% TRR (0.019 ppm) and 27.3% TRR (0.025 ppm) in PY-label straw; and 7.5% TRR (0.008 ppm) and 41.5% TRR (0.046 ppm) in PH-label straw. Additional extraction with aqueous sodium bicarbonate at reflux released only ≤6% TRR (<0.006 ppm) from rice grain and straw. The majority of the radioactivity (62-71% TRR, <0.04 ppm) in rice grain remained nonextractable, and was subjected to various enzyme hydrolyses, which released an additional 40-48% TRR. Nonextractable radioactivity (41-42% TRR, <0.05 ppm) in rice straw following solvent extraction was subjected to base and acid hydrolyses to precipitate cellulose and lignin (25% TRR).

Nonextractable residues were not reported following hydrolysis procedures; however, accountabilities were >68-95.1% for rice grain and straw, and uncharacterized nonextractable residues were ≤0.02 ppm in rice grain and straw. These procedures adequately extracted and characterized the majority of residues from rice grain and straw. Residues were identified and quantitated by normal and reverse phase TLC. Since all samples of rice grain and straw were stored frozen and analyzed within 124 days (4.1 months) of harvest, no supporting storage stability data are needed.

The metabolite profile differed between the PY and PH labels, demonstrating cleavage of the molecule between the two rings. Parent, orthosulfamuron, was not identified in PY-label grain and straw, and was only identified in PH-label grain and straw at trace levels (1.6% TRR, <0.001 ppm and 2.5% TRR, 0.003 ppm, respectively)

DOP urea was the only residue identified in PY-label grain and straw (13.0% TRR (0.008 ppm) and 21.2% TRR (0.019 ppm), respectively). DOP urea was not identified in PH-label matrices. The metabolite DBS acid was the major metabolite identified in PH-label straw at 34.8% TRR (0.038 ppm). DBS acid was identified in PH-label grain, but at lower levels (6.8% TRR, 0.003 ppm). Metabolites DB amine and DBS amide were also identified in both grain and straw at minor levels (<4% TRR, <0.005 ppm). The remaining unknowns, present at 15.7% TRR in PY-label grain, 27.3% TRR in PY-label straw, 9.2% TRR in PH-label grain and 6.8% TRR in PH-label straw, were characterized as organic or aqueous soluble and individually accounted for <0.01 ppm. Nonextractable residues were characterized as natural components accounting for: 22-25% TRR as cellulose, 6-9% TRR as starch, 3-4% TRR as pectin, 3-4% TRR as protein, and 1-2% TRR as lignin in grain (both labels); and 9-11% TRR as cellulose and 14-15% TRR as lignin in straw (both labels).

In the rice metabolism study, TRR levels were extremely low in rice matrices, and no significant degradation compounds were present in either grain or straw. The majority of the residue was bound, and the radioactivity was incorporated into natural components.

STUDY/WAIVER ACCEPTABILITY/DEFICIENCIES/CLARIFICATIONS:

Under the conditions and parameters used in the study, the rice metabolism data are classified as scientifically acceptable. The acceptability of this study for regulatory purposes is addressed in the U.S. EPA Residue Chemistry Summary Document, D332290, D. Dotson, 2/14/2007.

COMPLIANCE:

Signed and dated Good Laboratory Practice (GLP), Quality Assurance and Data Confidentiality statements were provided. The GLP Compliance Statement cites that the study was not conducted in accordance with GLP Standards as defined by the USA EPA, but in compliance with OECD principles of GLP as defined by Number 1 [ENV/MC/CHEM(98)17] on "The OECD principles of GLP;" Council Directives 88/320/EEC and 90/18/EEC as actuated by the Italian Decrees No. 120 of 1/27/92 and subsequent Ministerial Decree of 8/5/99; and EC directive 2004/9/EC of the European Parliament and of the Council.

A. BACKGROUND INFORMATION

Orthosulfamuron is a postemergence herbicide that Isagro S.p.A. is proposing for use on rice grown in the United States for the control of annual and perennial broadleaf weeds, sedges, and barnyard grass. Orthosulfamuron belongs to the sulfamoylurea class of herbicides. It reportedly acts by inhibiting the plant enzyme acetolactate synthase which is active in the biosynthesis of valine, leucine, and isoleucine.

TABLE A.1. Test Compound Nomenclature.

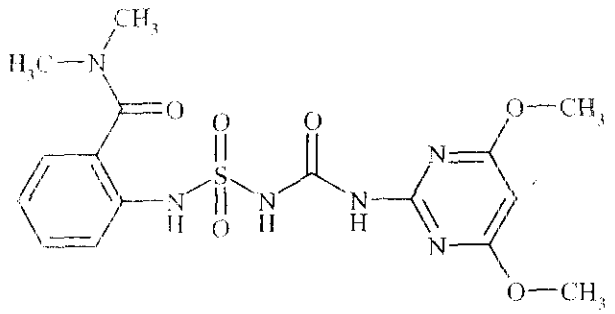
Compound	
Common name	Orthosulfamuron
Company experimental name	IR5878
IUPAC name	1-(4,6-dimethoxypyrimidin-2-yl)-3-[2-(dimethylcarbamoyl)phenylsulfamoyl]urea
CAS name	2-[[[[[(4,6-dimethoxy-2-pyrimidinyl)-amino]carbonyl]amino]sulfonyl]amino]-N,N-dimethylbenzamide
CAS registry number	213464-77-8
End-use product (EP)	0.51% G formulation (IR5878 0.5 GR; EPA Co. No. 80289) 51.5% WG formulation (IR5878 50 WG; EPA Co. No. 80289)

Table 2. Physicochemical Properties of the Technical Grade of Orthosulfamuron.		
Parameter	Value	Reference (MRID)
Color	White	46219004
Physical State	Fine Powder at 20°C	46219005
Odor	Odorless	46219006
pH	4.35 at 25°C (1% aqueous dispersion)	46219013
Density	1.45 g/mL at 20°C	46219008
Water solubility at 20°C	pH 4 buffer: 0.062 g/L pH 7 buffer: 0.63 g/L pH 8.5 buffer: 39 g/L	46219009
Solvent solubility at 20°C	n-heptane: 0.23 mg/L xylene: 130 mg/L acetone: 20 g/L ethyl acetate: 3.3 g/L dichloromethane: 56 g/L methanol: 8.3 g/L	Electronic communication. J. Messina to E. Kraft, 9/6/2006
Vapor pressure	1.1×10^{-4} at 20°C	46219010
Dissociation constant, pK_a	The test material becomes increasingly less soluble in water as the pH is lowered and undergoes degradation (hydrolysis) at neutral to acidic pHs. The test material is predicted to have 5 overlapping dissociation constants.	46219011
Octanol/water partition coefficient, $Log(K_{OW})$	pH 4: 2.0 pH 7: 1.3	46219012
UV/visible absorption spectrum	at pH 6.9, A=0.49 and $\varepsilon = 2.1 \times 10^4$ at 238 nm	46219001

B. EXPERIMENTAL DESIGN

B.1. Test Site and Crop Information

For each radiolabeled test solution, six minipaddy units of rice were treated. Each minipaddy unit consisted of a pot 73 cm long x 54 cm wide x 55 cm high, containing 20-25 cm of soil flooded with 8-10 cm of water. Four minipaddy units were treated at the nominal rate of ~75 g ai/ha (0.067 lb ai/A) and two minipaddy units were treated at an exaggerated rate of ~150 g ai/ha (0.134 lb ai/A). The exaggerated rate plots were conducted for metabolite identification, if necessary. In addition, two minipaddy units of rice were treated with formulation blank prepared as the test formulations (without orthosulfamuron) for controls.

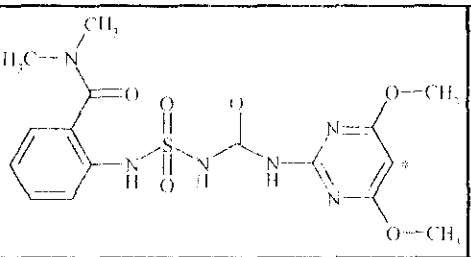
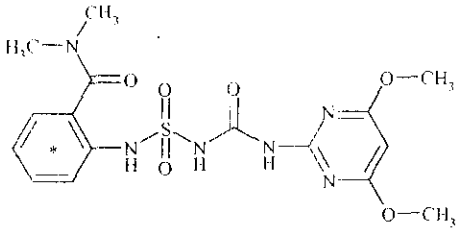
Minipaddy units were covered with a plastic tent during application and maintained in an outdoor green area during the study. Plants were grown using usual and customary agricultural practices for the area. Fertilizers, pesticides, and algae/insect/fungal chemical treatments were applied as required. Temperature, relative humidity, rainfall, and sunlight exposure were recorded daily throughout the study.

TABLE B.1.1. Test Site Information.					
Type	Method	Soil characteristics [1]			
		Type	%OM	pH	CEC
Foliar Treatment	Rice plants in minipaddy units maintained outdoors received a single application with a nitrogen assisted atomizer.	sandy loam	NA	NA	NA

[1] NA = Not applicable; only required for studies involving a soil treatment

TABLE B.1.2. Crop Information.				
Crop; crop group	Variety	Growth stage at application	Growth stage at harvest	Harvested Matrix
Rice: Grain, cereal, group 15, and Grain, cereal, forage, fodder, and straw, group 16	Loto	2-leaf (40 days after sowing)	mature	whole grain and straw; whole grain was also separated into husked rice and hulls

B.2. Test Materials

TABLE B.2.1. Test Material Characteristics.		
Chemical structure		
Radiolabel position	[^{14}C-5-pyrimidinyl]orthosulfamuron (PY-label)	[^{14}C-U-phenyl]orthosulfamuron (PH-label)
Lot No.	208	209
Radiochemical Purity	>97% (TLC and HPLC)	>96% (TLC and HPLC)
Specific activity	4.452 MBq/mg (120.323 µCi/mg)	7.357 MBq/mg (198.850 µCi/mg)

B.3. Study Use Pattern

TABLE B.3.1. Use Pattern Information.	
Chemical name	[^{14}C-5-pyrimidinyl]orthosulfamuron and [^{14}C-U-phenyl]orthosulfamuron
Application method	Each radiolabeled test substance was dissolved in acetonitrile and formulated as a WG by mixing with wettable powder. The solvent was evaporated and the remaining solid was suspended in water with a nonionic sufactant (0.02% v/v).
Application rate	PY-label: 0.070 lb ai/A (78 g ai/ha) or 0.141 lb ai/A (158 g ai/ha; exaggerated rate) PH-label: 0.066 lb ai/A (74 g ai/ha) or 0.134 lb ai/A (150 g ai/ha; exaggerated rate)
Number of applications	One
Timing of applications	Application was made at the 2-leaf growth stage
PHI	112 days after application

B.4. Identification/Characterization of Residues

B.4.1. Sample Handling and Preparation

At harvest, the water in each minipaddy was removed and mature rice plants were harvested by cutting the stem at the soil surface. The rice ears (whole grain; kernel plus hulls) were separated from the straw; a subsample of whole grain was separated into husked rice and hulls within a month of harvest. Samples of whole grain, husked rice, hulls, and straw were placed in frozen storage (-20°C) in the dark after collection.

Grain and husked rice samples were first homogenized (2x) with n-hexane to remove the seed oils. Samples were then centrifuged. Grain and husked rice after hexane extraction, and samples of hulls and straw were then extracted (2x) with acetonitrile (ACN):ammonium hydroxide (NH_4OH) pH 8.5 (1:1, v:v) and centrifuged. The respective supernatants were combined and brought to volume with ACN. The remaining residues were further extracted with 50 mM aqueous sodium bicarbonate ($NaHCO_3$) at reflux (80°C for 3 hours). The extract was centrifuged and the supernatant brought to volume with water.

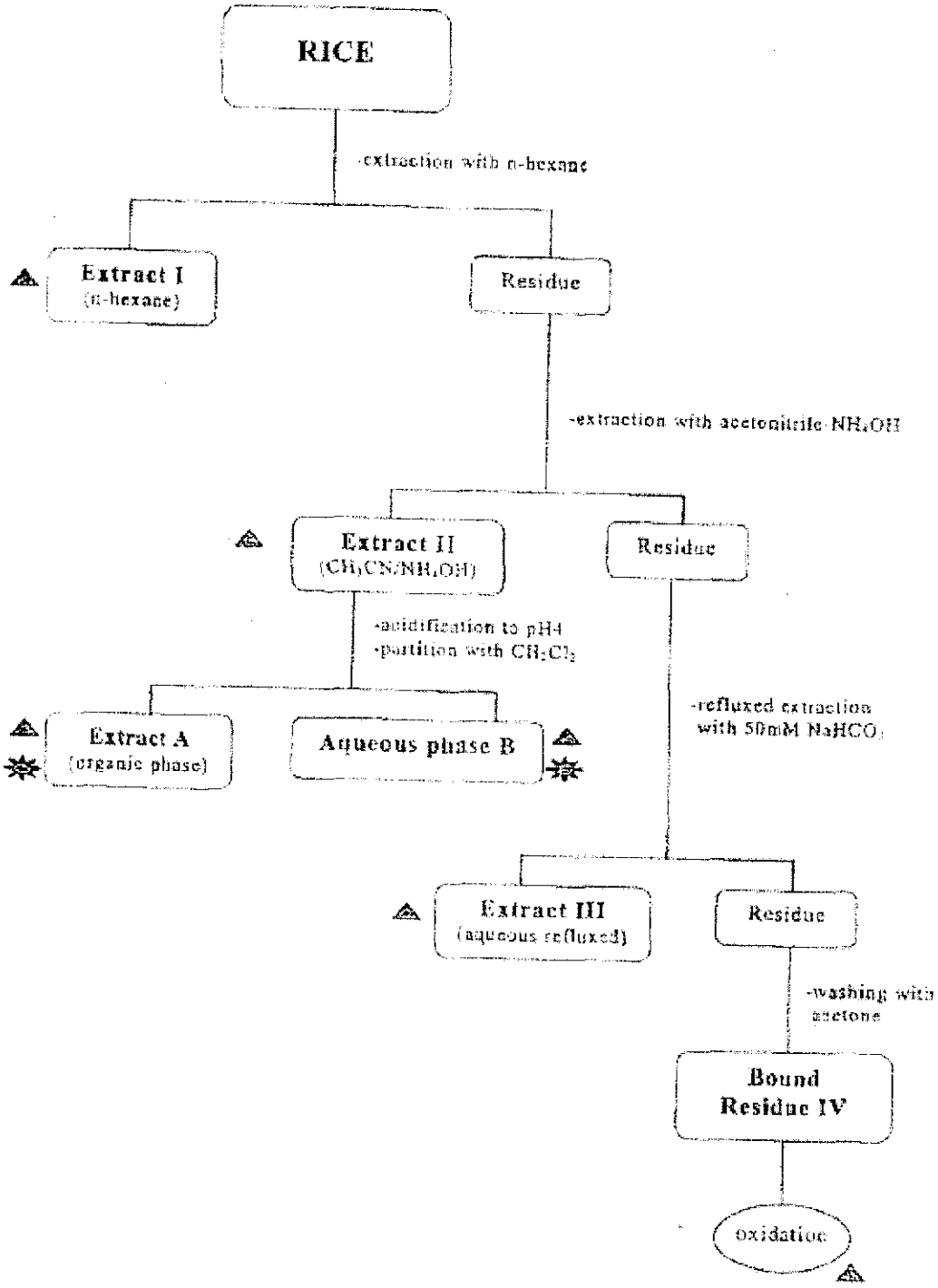
To characterize the residues, a subsample of the ACN/NH_4OH extract of each matrix was acidified to pH 4 with 0.1 N HCl and partitioned twice with dichloromethane. The aqueous and organic fractions were both reserved for TLC analysis.

Nonextractable residues of rice grain remaining after reflux extraction were washed with acetone, air-dried, and an aliquot was sequentially incubated with (i) 0.1 M acetate pH 4.8 buffer at 37 °C for 24 hours; (ii) cellulase in 0.1 M acetate pH 4.8 buffer at 37 °C for 24 hours (cellulose fraction); (iii) amylase in 0.002 M acetate pH 6.9 buffer at 37 °C for 24 hours (starch fraction); (iv) pectinase in 0.1 M acetate pH 4.0 buffer at 37 °C for 24 hours (pectin fraction); (v) protease in 0.05M phosphate pH 7.5 buffer at 37 °C for 24 hours (protein fraction); and (vi) dimethyl sulfoxide (DMSO) at 80 °C for 24 hours (lignin fraction). After each incubation step, the hydrolysate was collected for LSC analysis by centrifugation.

Nonextractable residues of rice straw, washed with acetone, were fractionated to determine incorporation into natural components. The bound residue was refluxed with 5% sodium hydroxide (110 °C for 3 hours) and centrifuged to isolate the cellulose fraction (solids) from the soluble fraction. The soluble fraction was then acidified with 6 N HCl (pH 2) to precipitate the lignin fraction.

The extraction flowcharts are presented in Figures B.4.1.1 (grain and husked rice), B.4.1.2 (hulls and straw), B.4.1.3 (grain nonextractable residues), and B.4.1.4. (straw nonextractable residues). These were copied from MRID 46578961 without alteration.

Figure B.4.1.1. Extraction Scheme for Whole Grain and Husked Rice.



LSC analysis

TLC analysis

Figure B.4.1.2. Extraction Scheme for Rice Hulls and Straw.

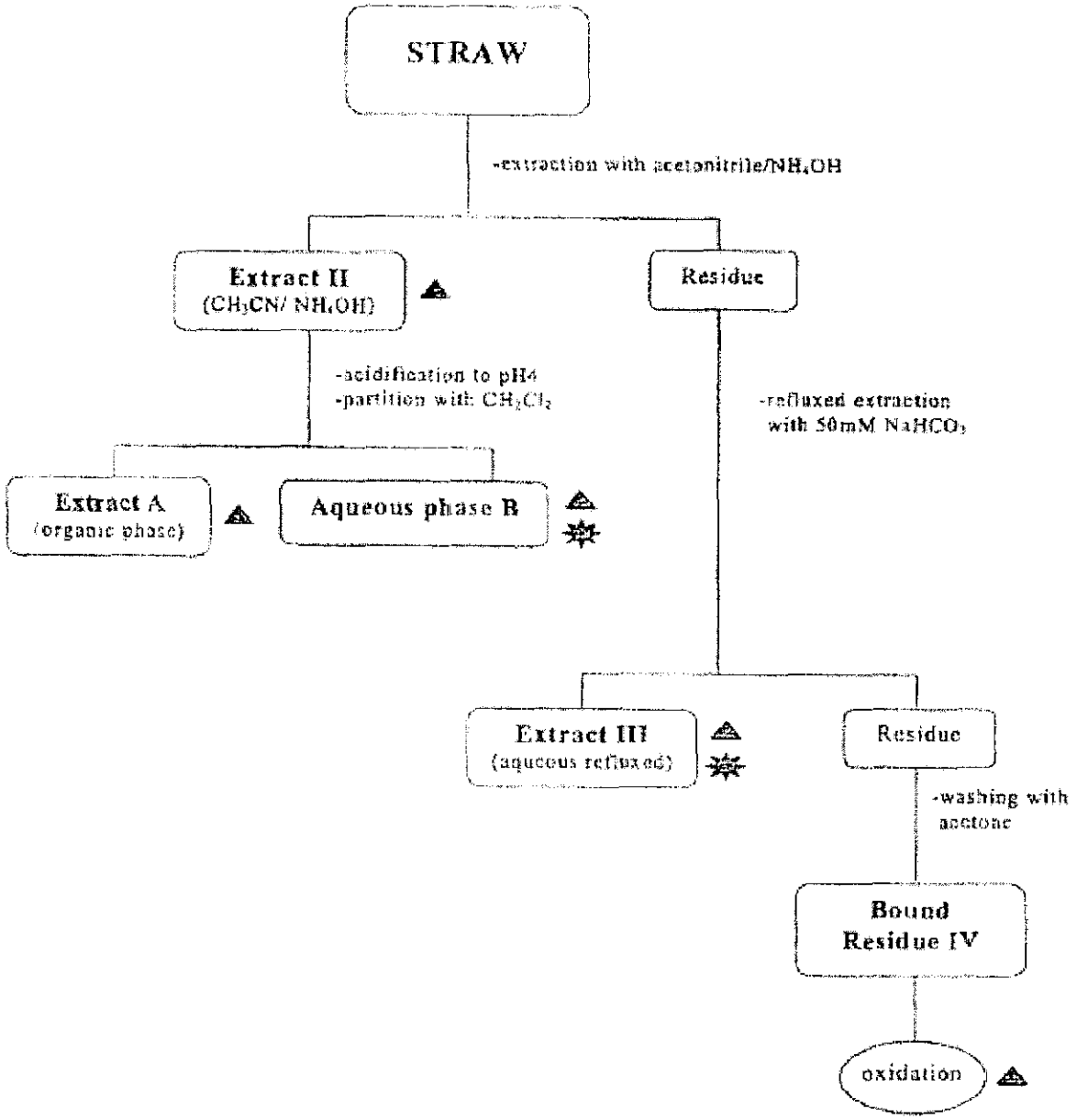


Figure B.4.1.3. Extraction Scheme for Nonextractable Residues in Grain.

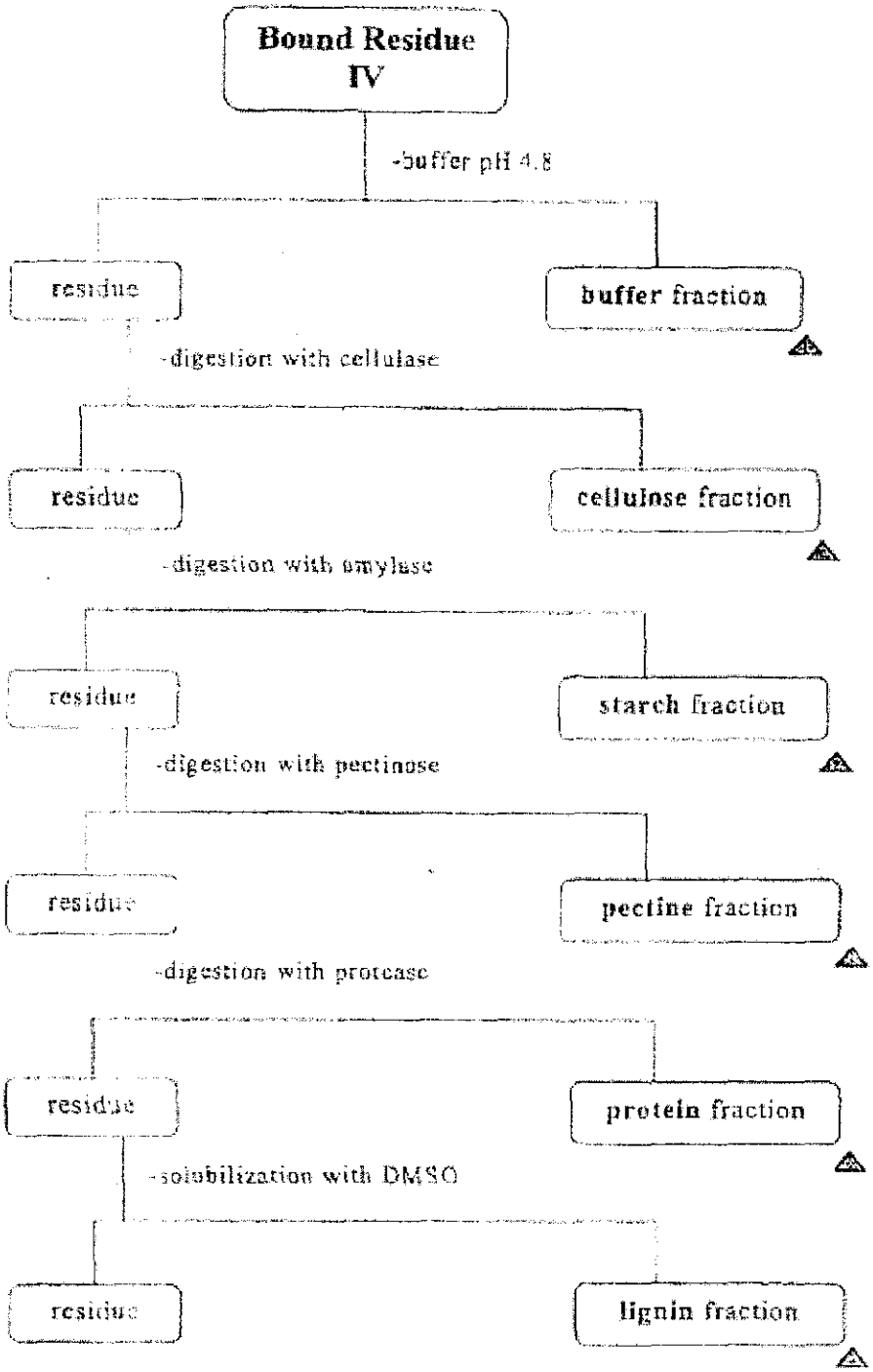
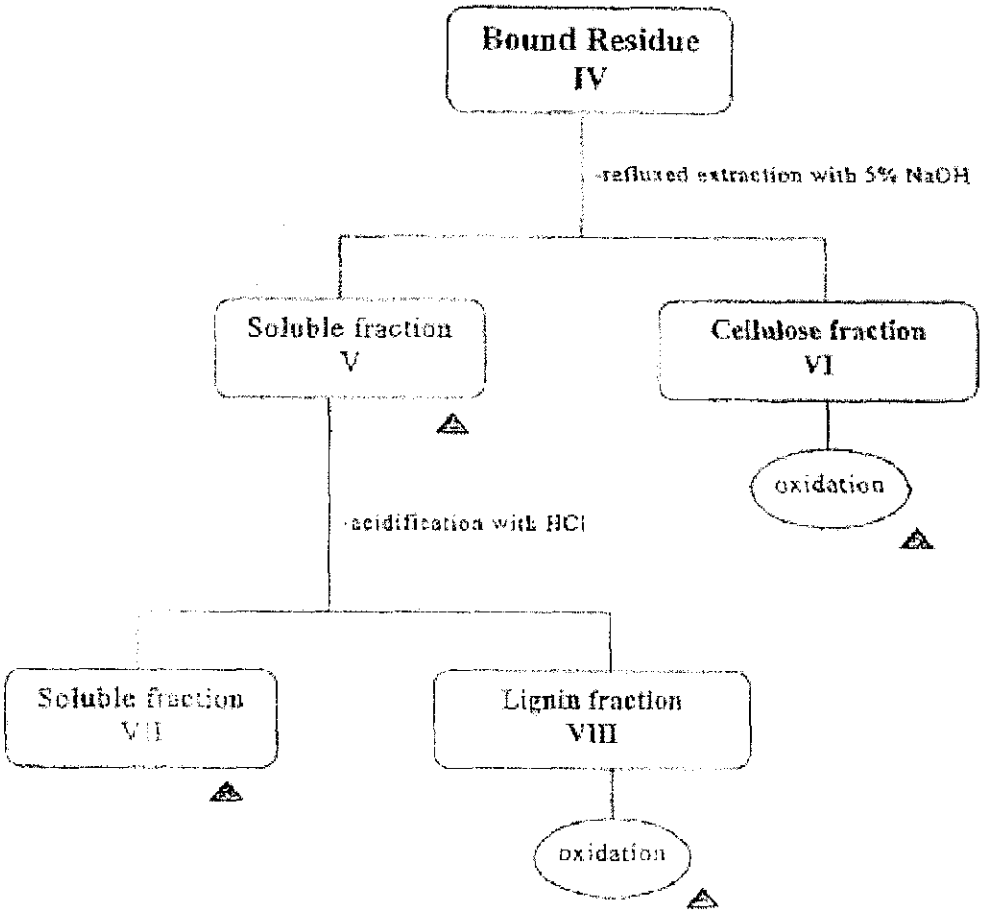


Figure B.4.1.4. Extraction Scheme for Nonextractable Residues in Straw.



B.4.2. Analytical Methodology

TRR in rice whole grain, husked rice, hulls, and straw were determined by combustion/LSC of ground subsamples. Extracts, hydrolysates, and soluble fractions were radioassayed in duplicate by LSC, and straw cellulose and lignin solids, and nonextractable residues were radioassayed in triplicate by combustion/LSC. The reported limit of detection for TRR determinations was twice the background.

The organic and aqueous phases of the ACN/NH_4OH extracts of whole grain, husked rice, hulls and straw were each subjected to TLC analysis for identification and quantitation of metabolites. Normal phase TLC analyses were conducted using silica gel plates and a solvent system of ACN:water (90:10, v:v; PY-label) or chloroform:methanol:ammonium hydroxide (75:22:3, v:v:v; PH-label). Reverse phase TLC analyses were conducted using RP-18 plates and a solvent system of ACN:water (92:8, v:v). Samples were analyzed by both normal and reverse phase TLC systems, but normal phase TLC was used for quantitation of residues in PY-labeled

samples and reverse phase TLC was used for quantitation of residues in PH-labeled samples. TLC plates were evaluated using radioimaging. Luminescence was detected by a photo multiplier. Metabolites were identified by co-chromatography with radiolabeled reference standards. Chemical names and structures for the reference standards are presented in Appendix I.

C. RESULTS AND DISCUSSION

The storage conditions and intervals for rice samples are presented in Table C.1. Actual extraction and analysis dates were not provided, but based on the reported month of analysis, all samples of rice were analyzed within 124 days (4.1 months) of harvest; therefore, no supporting storage stability data are needed.

Samples of whole grain and straw were collected 112 days after a single foliar application of [^{14}C-5-pyrimidinyl]orthosulfamuron (PY label) at 0.070 lb ai/A or [^{14}C-U-phenyl]orthosulfamuron (PH label) at 0.066 lb ai/A. Separate plots were treated at an exaggerated rate in case additional material was required for metabolite identification; these samples were not used in the study. Subsamples of whole grain were separated into husked rice and hulls.

Total radioactive residues (TRR) in rice matrices were determined by combustion/LSC, and are reported in Table C.2.1. TRR were 0.0632 ppm and 0.0898 ppm in samples of rice whole grain and straw, respectively, treated with PY-label orthosulfamuron. TRR were 0.0368 ppm and 0.1101 ppm in samples of rice whole grain and straw, respectively, treated with PH-label orthosulfamuron. In both PY- and PH-label grain, TRR appear to be concentrated in hulls of rice grain and husked rice contains lower radioactivity than the whole grain. TRR were 0.0584 ppm and 0.1128 ppm in PY-label husked rice and hulls, and 0.0330 ppm and 0.0502 ppm in PH-label husked rice and hulls, respectively.

The distribution of the radioactivity in rice matrices is presented in Tables C.2.2.1 (PY label) and C.2.2.2 (PH label); the distribution of residues in husked rice and hulls are presented for informational purposes and are not discussed further. The distribution of radioactivity was similar between the two labels. Samples of rice grain were first extracted with hexane, which only released ~1-2% TRR. A large amount of radioactivity was released from both rice grain (~21-29% TRR; 0.008-0.018 ppm) and straw (~48-49% TRR; 0.043-0.054 ppm) with solvent extraction (ACN/NH_4OH). Partitioning of the solvent extract separated the organic and aqueous soluble residues, respectively as 13.0% TRR (0.008 ppm) and 15.7% TRR (0.010 pm) in PY-label grain; 5.4% TRR (0.002 ppm) and 16.0% TRR (0.006 ppm) in PH-label grain; 21.2% TRR (0.019 ppm) and 27.3% TRR (0.025 ppm) in PY-label straw; and 7.5% TRR (0.008 ppm) and 41.5% TRR (0.046 ppm) in PH-label straw. Additional extraction with aqueous sodium bicarbonate at reflux released only 6% or less of the TRR (<0.006 ppm) from rice grain and straw.

The majority of the radioactivity (62-71% TRR, <0.04 ppm) in rice grain remained nonextractable, and was subjected to various enzyme hydrolyses, which released an additional

40-48% TRR. Nonextractable radioactivity (41-42% TRR, <0.05 ppm) in rice straw following solvent extraction was subjected to base and acid hydrolyses to precipitate cellulose and lignin (25% TRR). Nonextractable residues were not reported following hydrolysis procedures; however, accountabilities were >68-95.1% for rice grain and straw and uncharacterized nonextractable residues were ≤0.02 ppm in rice grain and straw. These procedures adequately extracted and characterized the majority of residues from rice grain and straw. Residues were identified and quantitated by normal and reverse phase TLC.

The characterization and identification of residues in rice matrices are summarized in Table C.2.3. Summaries of the residues in husked rice and hulls are not included. The metabolite profile differed between the PY and PH labels, demonstrating cleavage of the molecule between the two rings. Parent, orthosulfamuron, was not identified in PY-label grain and straw, and was only identified in PH-label grain and straw at trace levels (1.6% TRR, <0.001 ppm and 2.5% TRR, 0.003 ppm, respectively).

DOP urea was the only residue identified in PY-label grain and straw (13.0% TRR (0.008 ppm) and 21.2% TRR (0.019 ppm), respectively). DOP urea was not identified in PH-label matrices. The metabolite DBS acid was the major metabolite identified in PH-label straw at 34.8% TRR (0.038 ppm). DBS acid was identified in PH-label grain, but at lower levels (6.8% TRR, 0.003 ppm). Metabolites DB amine and DBS amide were also identified in both grain and straw at minor levels (<4% TRR, <0.005 ppm). The remaining unknowns, present at 15.7% TRR in PY-label grain, 27.3% TRR in PY-label straw, 9.2% TRR in PH-label grain and 6.8% TRR in PH-label straw, were characterized as organic or aqueous soluble and individually accounted for <0.01 ppm. Nonextractable residues were characterized as natural components accounting for: 22-25% TRR as cellulose, 6-9% TRR as starch, 4% TRR as pectin, 3-4% TRR as protein, and 1-2% TRR as lignin in grain (both labels); and 9-11% TRR as cellulose and 14-15% TRR as lignin in straw (both labels).

The identities of the orthosulfamuron, DOP-urea, DBS acid, DB amine and DBS amide were confirmed in respective extracts of rice grain and straw by co-chromatography with radiolabeled standards using two different TLC systems.

C.1. Storage Stability

Samples of rice whole grain and straw were stored frozen (-20°C) for ~2 months from harvest to extraction, and appear to have been analyzed within a month of extraction. Actual extraction and analysis dates were not provided. Extracts were immediately analyzed or stored at -4°C or -20°C until analysis.

TABLE C.1. Summary of Storage Conditions.			
Matrix	Storage Temperature (°C)	Actual Storage Duration [1]	Interval of Demonstrated Storage Stability
Rice, whole grain	-4 or -20	≤124 days (≤4.1 months)	None required
Rice, husked		≤93 days (≤3.1 months)	
Rice, hulls		≤93 days (≤3.1 months)	
Rice, straw		≤93 days (≤3.1 months)	

[1] Only the month/year of extraction and analysis was provided; duration from harvest to analysis was calculated assuming analysis on the last day of the month.

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

TABLE C.2.1. Total Radioactive Residues (TRR) in Rice Matrices.				
Matrix	Timing and Applic. No	PHI (days)	PY Label	PH Label
			ppm	ppm
Rice, whole grain	One foliar application at ~0.067 lb ai/A	112	0.0632	0.0368
-husked rice			0.0584	0.0330
-hulls			0.1128	0.0502
Rice, straw			0.0898	0.1101

TABLE C.2.2.1. Distribution of the Parent and the Metabolites in Rice Matrices Following Application of [Pyrimidinyl-^{14}C]Orthosulfamuron at 0.070 lb ai/A. [1]								
Metabolite Fraction	Whole Grain		Husked Rice		Hulls		Straw	
	TRR = 0.0632 ppm		TRR = 0.0584 ppm		TRR = 0.1128 ppm		TRR = 0.0898 ppm	
	% TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm
Hexane extract	2.22	0.0014	3.25	0.0019				
ACN/NH_4OH extract	*28.64*	0.0181	*21.06*	0.0123	*40.07*	0.0452	*48.33*	0.0434
-Organosoluble	12.97	0.0082	4.62	0.0027	25.62	0.0289	21.16	0.0190
DOP urea	12.97	0.0082	4.62	0.0027	25.62	0.0289	21.16	0.0190
-Aqueous soluble [2]	15.66	0.0099	16.61	0.0097	14.45	0.0163	27.28	0.0245
Aqueous reflux extract	6.49	0.0041	5.99	0.0035	7.45	0.0084	5.12	0.0046
Nonextractable	62.34	0.0394	67.12	0.0392	46.72	0.0527	41.65	0.0374
-pH 4.8 Buffer extract	*4.59*	0.0029						
-Cellulase hydrolysate (cellulose)	25.32	0.0160						
-Amylase hydrolysate (starch)	8.70	0.0055						
-Pectinase hydrolysate (pectin)	3.80	0.0024						
-Protease hydrolysate (protein)	3.48	0.0022						
-DMSO hydrolysate (lignin)	1.74	0.0011						
-Solids	NR	NR						
-Cellulose solid							11.02	0.0099
-Lignin precipitate							13.81	0.0124

[1] Shading indicates that the extraction step and/or characterization analysis was not conducted for the matrix in question. Percent TRR values in *italics* were calculated by the study reviewer from the reported ppm value in raw data. NR = Not reported. Metabolites for husked rice and straw were only reported as % of the TLC chromatogram; the study reviewer has estimated these values as %TRR and ppm for informational purposes.

[2] TLC analysis characterized four unknown peaks, each present at <0.005 ppm in grain and husked rice, and <0.008 ppm in straw and hulls.

TABLE C.2.2.2. Distribution of the Parent and the Metabolites in Rice Matrices Following Application of [Phenyl-^{14}C]Orthosulfamuron at 0.066 lb ai/A. [1]

Metabolite Fraction	Whole Grain		Husked Rice		Hulls		Straw	
	TRR = 0.0368 ppm		TRR = 0.0330 ppm		TRR = 0.0502 ppm		TRR = 0.1101 ppm	
	% TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm
Hexane extract	0.82	0.0003	1.21	0.0004				
ACN/NH_4OH extract	*21.47*	0.0079	*20.30*	0.0067	*29.28*	0.0147	*48.96*	0.0539
-Organosoluble	5.43	0.0020	2.42	0.0008	11.15	0.0056	7.45	0.0082
Orthosulfamuron	1.63	0.0006	0.75	0.0002	1.89	0.0010	2.45	0.0027
DB amine	0.82	0.0003	0.93	0.0003	1.77	0.0009	1.27	0.0014
DBS amide	2.99	0.0011	0.75	0.0002	7.48	0.0038	3.81	0.0042
-Aqueous soluble	16.03	0.0059	17.88	0.0059	18.12	0.0091	41.51	0.0457
OBS acid	6.79	0.0025	5.91	0.0020	9.65	0.0048	34.79	0.0383
Unknowns [2]	*9.24*	0.0034	11.97	0.0040	8.47	0.0043	*6.81*	0.0075
Aqueous reflux extract	6.25	0.0023	6.36	0.0021	5.58	0.0028	5.18	0.0057
Nonextractable	70.92	0.0261	73.03	0.0241	67.73	0.0340	40.78	0.0449
-pH 4.8 Buffer extract	*4.62*	0.0017						
-Cellulase hydrolysate (cellulose)	22.28	0.0082						
-Amylase hydrolysate (starch)	5.71	0.0021						
-Pectinase hydrolysate (pectin)	3.26	0.0012						
-Protease hydrolysate (protein)	3.26	0.0012						
-DMSO hydrolysate (lignin)	0.82	0.0003						
-Solids	NR	NR						
-Cellulose solid							9.45	0.0104
-Lignin precipitate							15.35	0.0169

[1] Shading indicates that the extraction step and/or characterization analysis was not conducted for the matrix in question. Percent TRR values in *italics* were calculated by the study reviewer from the reported ppm value in raw data. NR = Not reported. Metabolites for husked rice and straw were only reported as % of the TLC chromatogram; the study reviewer has estimated these values as %TRR and ppm for informational purposes.
[2] One unknown peak in grain and husked rice, and two unknown peaks in straw and hulls.

TABLE C.2.3. Summary of Characterization and Identification of Radioactive Residues in Rice Matrices Following Application of [^{14}C-5-Pyrimidinyl] or [^{14}C-U-Phenyl] Orthosulfamuron at ~0.067 lb ai/A.

Compound	PY Label				PH Label			
	Rice, grain		Rice, straw		Rice, grain		Rice, straw	
	TRR = 0.0632 ppm		TRR = 0.0898 ppm		TRR = 0.0368 ppm		TRR = 0.1101 ppm	
	%TRR	ppm	%TRR	ppm	% TRR	ppm	% TRR	ppm
Orthosulfamuron	--	--	--	--	1.63	0.0006	2.45	0.0027
DOP urea	12.97	0.0082	21.16	0.0190	--	--	--	--
DB amine	--	--	--	--	0.82	0.0003	1.27	0.0014
DBS amide	--	--	--	--	2.99	0.0011	3.81	0.0042
DBS acid	--	--	--	--	6.79	0.0025	34.79	0.0383

TABLE C.2.3. Summary of Characterization and Identification of Radioactive Residues in Rice Matrices Following Application of [^{14}C-5-Pyrimidinyl] or [^{14}C-U-Phenyl] Orthosulfamuron at ~0.067 lb ai/A.

Compound	PY Label				PH Label			
	Rice, grain		Rice, straw		Rice, grain		Rice, straw	
	TRR = 0.0632 ppm		TRR = 0.0898 ppm		TRR = 0.0368 ppm		TRR = 0.1101 ppm	
	%TRR	ppm	%TRR	ppm	% TRR	ppm	% TRR	ppm
Unknowns (aqueous)	15.66	0.0099	27.28	0.0245	9.24	0.0034	6.81	0.0075
Hexane soluble	2.22	0.0014	--	--	0.82	0.0003	--	--
Aqueous reflux	6.49	0.0041	5.12	0.0046	6.25	0.0023	5.18	0.0057
pH 4.8 Buffer	4.59	0.0029	--	--	4.62	0.0017	--	--
Cellulose	25.32	0.0160	11.02	0.0099	22.28	0.0082	9.45	0.0104
Starch	8.70	0.0055	--	--	5.71	0.0021	--	--
Pectin	3.80	0.0024	--	--	3.26	0.0012	--	--
Protein	3.48	0.0022	--	--	3.26	0.0012	--	--
Lignin	1.74	0.0011	13.81	0.0124	0.82	0.0003	15.35	0.0169
Total identified	12.97	0.0082	21.16	0.0190	12.23	0.0045	42.32	0.0466
Total characterized	72.00	0.0455	57.23	0.0514	56.26	0.0207	36.79	0.0405
Total extractable	84.98	0.0537	53.45	0.0480	68.49	0.0252	54.14	0.0596
Unextractable (PES)[1]	NR	NR	41.65	0.0374	NR	NR	40.78	0.0449
Accountability[2]	>85		95.1		>68		94.9	

[1] Residues remaining after exhaustive extractions. NR = Not reported.
Nonextractable residues after chemical and enzyme treatments were not reported for grain; however, based on the extracts/hydrolysates released, nonextractable residues would be ≤0.01 ppm in grain.
Nonextractable residues in straw were further characterized as cellulose and lignin bound residues precipitated under basic and acidic conditions (as reported in the table); uncharacterized nonextractable residues were <0.02 ppm in straw.
[2] Accountability = (Total extractable + Total unextractable)/(TRR from combustion analysis; see TABLE C.2.1) * 100.

C.3. Proposed Metabolic Profile

Based on the rice metabolism study, the petitioner states that TRR levels were extremely low in both rice and straw and that no significant degradation compounds were present in either rice or straw. The majority of the residue was bound, and the radioactivity was incorporated into natural components.

FIGURE C.3.1. Proposed Metabolic Profile of Orthosulfamuron in Rice

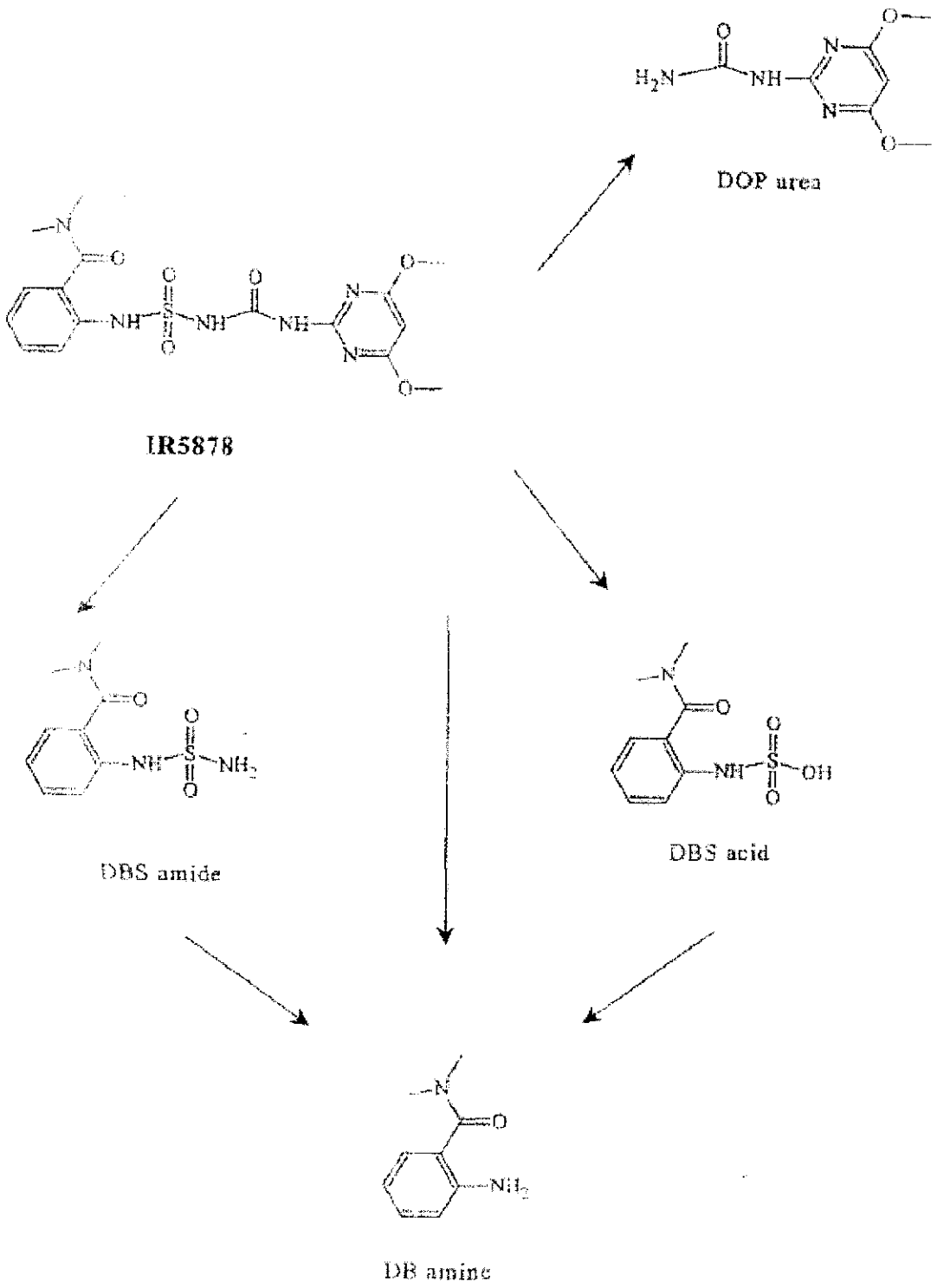
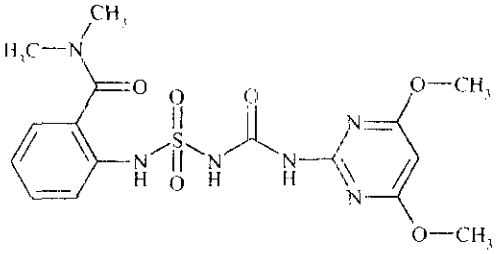
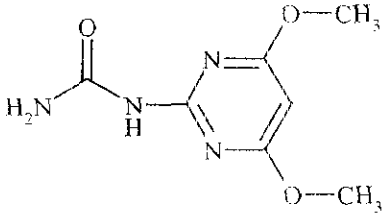
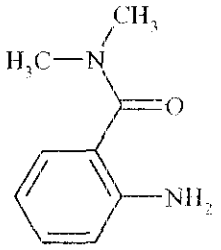
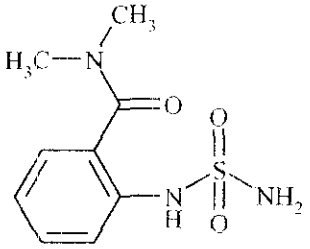
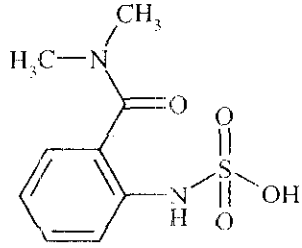


TABLE C.3.1. Identification of Compounds from Metabolism Study

Common name/code Figure C.3.1 ID No.	Chemical name	Chemical structure
Orthosulfamuron	2-[[[[[(4,6-dimethoxy-2-pyrimidinyl)-amino]carbonyl]amino]sulfonyl]amino]-N,N-dimethylbenzamide	
DOP urea	N-(4,6-dimethoxypyrimidin-2-yl)urea	
DB amine	2-amino-N,N-dimethylbenzamide	
DBS amide	2-sulfamoylamino-N, N-dimethylbenzamide	
DBS acid	2-sulfoamino-N,N-dimethylbenzamide	

D. CONCLUSION

Total radioactive residues (TRR) in rice matrices were 0.0632 ppm and 0.0898 ppm in samples of rice whole grain and straw, respectively, harvested 112 days after a single foliar application of [^{14}C-5-pyrimidinyl]orthosulfamuron (PY label) at 0.070 lb ai/A. TRR were 0.0368 ppm and 0.1101 ppm in samples of rice whole grain and straw, respectively, harvested 112 days after a single foliar application of [^{14}C-U-phenyl]orthosulfamuron (PH label) at 0.066 lb ai/A. Subsamples of whole grain were separated into husked rice and hulls. In both PY- and PH-label grain, TRR appear to be concentrated in hulls of rice grain, and husked rice contains lower radioactivity than the whole grain. TRR were 0.0584 ppm and 0.1128 ppm in PY-label husked rice and hulls, and 0.0330 ppm and 0.0502 ppm in PH-label husked rice and hulls, respectively.

The distribution of radioactivity was similar between the two labels. Samples of rice grain were first extracted with hexane, which only released ~1-2% TRR. A large amount of radioactivity was released from both rice grain (~21-29% TRR) and straw (~48-49% TRR) with solvent extraction. Partitioning of the solvent extract separated the organic and aqueous soluble residues, respectively as 13.0% TRR and 15.7% TRR in PY-label grain; 5.4% TRR and 16.0% TRR in PH-label grain; 21.2% TRR and 27.3% TRR in PY-label straw; and 7.5% TRR and 41.5% TRR in PH-label straw. Additional extraction with aqueous sodium bicarbonate at reflux released only ≤6% TRR from rice grain and straw. The majority of the radioactivity (62-71% TRR, <0.04 ppm) in rice grain remained bound or nonextractable, and was further subjected to various enzyme hydrolyses, which released an additional 40-48% TRR. Nonextractable radioactivity (41-42% TRR, <0.05 ppm) in rice straw following solvent extraction was subjected to base and acid hydrolyses to precipitate cellulose and lignin (25% TRR). Uncharacterized nonextractable residues were ≤0.02 ppm in rice grain and straw.

Residues were identified and quantitated by normal and reverse phase TLC. The metabolite profile differed between the PY and PH labels. Parent, orthosulfamuron, was not identified in PY-label grain and straw, and was only identified in PH-label grain and straw at trace levels (<0.003 ppm).

DOP urea was the only residue identified in PY-label grain and straw at 13.0% TRR and 21.2% TRR, respectively; DOP urea was not identified in PH-label matrices. The metabolite DBS acid was the major metabolite identified in PH-label straw at 34.8% TRR; DBS acid was identified in PH-label grain, but at lower levels (6.8% TRR). Metabolites DB amine and DBS amide were also identified in both grain and straw at minor levels (<4% TRR). The remaining unknowns, present at 15.7% TRR in PY-label grain, 27.3% TRR in PY-label straw, 9.2% TRR in PH-label grain and 6.8% TRR in PH-label straw, were characterized as organic or aqueous soluble and individually accounted for <0.01 ppm. Nonextractable residues were characterized as natural components accounting approximately for: 22-25% TRR as cellulose, 6-9% TRR as starch, 3-4% TRR as pectin, 3-4% TRR as protein, and 1-2% TRR as lignin in grain (both labels); and 9-11% TRR as cellulose and 14-15% TRR as lignin in straw (both labels).

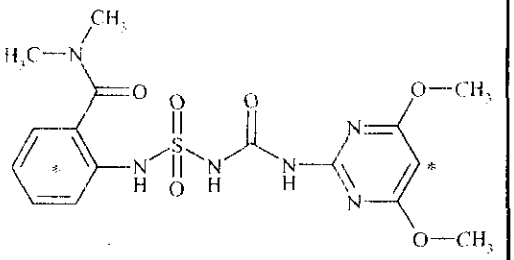
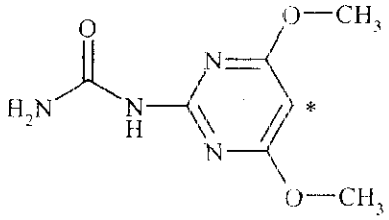
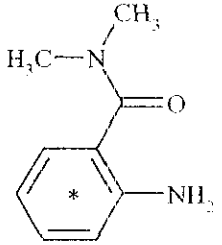
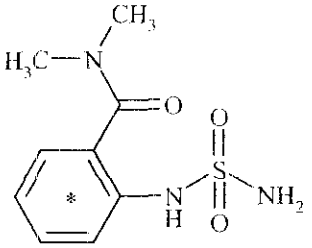
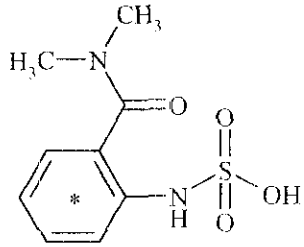
In the rice metabolism study, TRR levels were extremely low in rice matrices and no significant degradation compounds were present in either grain or straw. The majority of the residue was bound, and the radioactivity was incorporated into natural components.

E. REFERENCES

None.

F. DOCUMENT TRACKING

Petition Number: 5F6967
DP Barcode: D319614
PC Code: 108209

APPENDIX I. Chemical Names and Structures of Reference Standards Used in Rice Metabolism Study.		
Common name, Company code	Chemical name	Chemical structure
[^{14}C]Orthosulfamuron; IR5878 (PY and PH label)	2-[[[[[(4,6-dimethoxy-2-pyrimidinyl)-amino]carbonyl]amino]sulfonyl]amino]-N,N-dimethylbenzamide	
[^{14}C]DOP urea	N-(4,6-dimethoxypyrimidin-2-yl)urea	
[^{14}C]DB amine	2-amino-N,N-dimethylbenzamide	
[^{14}C]DBS amide	2-sulfamoylamino-N, N-dimethylbenzamide	
[^{14}C]DBS acid	2-sulfoamino-N,N-dimethylbenzamide	

Orthosulfamuron/IR5878/PC Code 108209/Isagro S.p.A.
DACO 7.4.3/OPPTS 860.1850/OECD IIA 6.6.3, 6.8.7 and IIIA 8.6
Confined Accumulation in Rotational Crops - Carrot, Lettuce & Wheat

Primary Evaluator	Douglas Dotson, Chemist, RAB2 D. Dotson	Date: 2/14/2007
Peer Reviewer	Dennis McNeilly, Chemist, RAB2	Date: 2/14/2007

This DER was originally prepared under contract by Dynamac Corporation (2275 Research Boulevard, Suite 300; Rockville, MD 20850; submitted 06/12/2006). The DER has been reviewed by the Health Effects Division (HED) and revised to reflect current Office of Pesticide Programs (OPP) policies.

STUDY REPORT:

46578988 Rizzo, F.; Garau, S.; Pizzingrilli, G. (2004) Uptake, Translocation and Metabolism of [(^{14}C)-Phenyl]IR5878 in Rotated Crops of Wheat, Carrots and Lettuce. Project Number: MEF.02.05_A2_06.06/02. Unpublished study prepared by Isagro S.R.L. (Formerly Agrimont). 246 p.

EXECUTIVE SUMMARY:

Isagro S.p.A. has submitted a confined rotational crop study with [^{14}C-U-phenyl]ortho-sulfamuron (PH label; specific activity 106.998-110.660 µCi/mg). The radiolabeled test substance was mixed with acetonitrile/water and applied micro-dropwise to sandy loam soil in plastic pots, maintained outdoors, at a nominal rate of 0.067 lb ai/A. Carrot (root vegetable), lettuce (leafy vegetable), and wheat (small grain) were planted in the treated soil as representative rotational crops at plantback intervals (PBIs) of ca. 30, 120, and 365 days. The in-life and analytical phases of the study were conducted at Isagro Ricerca (Novara, Italy).

After a single application of [^{14}C-U-phenyl]orthosulfamuron (PH label) to bare soil at 0.073 lb ai/A, total radioactive residues (TRR) accumulated at ≥0.01 ppm in the following: carrot tops planted 33 and 128 days after application, wheat forage planted 29 and 121 days after application, and wheat straw planted 29, 121, and 365 days after application. TRR were below 0.01 ppm in carrot root, lettuce, and wheat grain rotated crop matrices at all plantback intervals (PBIs), and in carrot tops and wheat forage planted at the 373/365-day PBI. In general, TRR were highest at the ~30 or 120-day PBI, and greatly reduced at the ~365-day PBI. Residues in carrot tops were 0.0442 ppm at the 33-day PBI, 0.0368 ppm at the 128-day PBI, and 0.0063 ppm at the 373-day PBI; residues in wheat forage were 0.0286 ppm at the 29-day PBI, 0.0310 ppm at the 121-day PBI, and 0.0019 ppm at the 365-day PBI; and residues in wheat straw were 0.5763 ppm at the 29-day PBI, 0.8730 ppm at the 121-day PBI, and 0.1458 ppm at the 365-day PBI.

Rotated crop matrices with TRR >0.01 ppm were extracted (carrot tops and wheat forage, 33/29- and 128/121-day PBIs, and wheat straw, all PBIs). The majority of the radioactivity (72-94% TRR) was extracted from the rotated crop matrices using ACN/ammonium bicarbonate; small amounts of radioactivity (1-5% TRR) were subsequently released with acetone. The ACN

extracts were partitioned into DCM, ethyl acetate and aqueous soluble phases for metabolite analysis. The majority of the extractable residue was aqueous soluble. Nonextractable residues in the extracted crop matrices were <0.01 ppm in carrot tops and wheat forage, and 0.031-0.074 ppm (7-21% TRR) in wheat straw. Nonextractable residues were partially characterized as cellulose and lignin (see below) and accountabilities ranged from 89 to 101%. The extraction procedures extracted sufficient residues from all PBIs. Metabolites were quantitated and identified by co-chromatography with the respective radiolabeled standard using normal and reverse phase TLC.

Total identified residues ranged from 51 to 76% TRR in rotated carrot tops, wheat forage, and wheat straw. The parent, orthosulfamuron, was only identified in rotated wheat straw (all PBIs) at trace levels (<1% TRR; 0.001-0.004 ppm).

The major metabolite identified in rotated crop matrices was DBS-acid. DBS-acid accounted for 63.2% and 75.7% TRR (0.028 ppm) in carrot tops from the 33- and 128-day PBIs, respectively; 50.8% and 51.1% TRR (0.015-0.016 ppm) in wheat forage from the 29- and 121-day PBIs, respectively; and 63.7%, 65.8%, and 44.8% TRR (0.066-0.575 ppm) in wheat straw from the 29-, 121-, and 365-day PBIs, respectively. DBS-amide was identified as a minor metabolite in wheat forage and straw accounting for 9.2-9.8% TRR (0.003 ppm) in wheat forage from the 29- and 121-day PBIs, and 3.2-4.1% TRR (0.005-0.028 ppm) in wheat straw from all PBIs; DBS-amide was not identified in carrot tops. DB-amine was identified as a minor residue present only in wheat straw (<1-2.6% TRR; 0.004-0.005 ppm).

An unknown metabolite, characterized as a conjugate of DBS-acid, was determined to be a significant residue in wheat forage and accounted for 28.2% TRR (0.008 ppm) at the 29-day PBI and 19.2% TRR (0.006 ppm) at the 121-day PBI. Unknown 2 was found in carrot tops (1.9-6.5% TRR, ≤0.003 ppm) and wheat straw (5.4-8.6% TRR, 0.008-0.068 ppm) at minor levels. Another unknown metabolite, characterized as two N-glucosides of hydroxylated DB-amine, was determined to be a minor residue in carrot tops (3.8-4.7% TRR, 0.002 ppm), wheat forage (5.9-6.5% TRR, 0.002 ppm), and wheat straw (4.2-5.7% TRR, 0.008-0.050 ppm). The remaining extractable residues were characterized as one or two unknowns in the aqueous phase present at <0.01 ppm in carrot tops and wheat forage and ≤0.02 ppm in wheat straw, and a single unknown in the organic phases present at <0.01 ppm in wheat straw.

Nonextractable residues were partially characterized as cellulose and lignin bound residues accounting for ~7-8% TRR in carrot tops (33- and 128-day PBIs), and 5-11% TRR in wheat straw (all PBIs); the uncharacterized nonextractable residues represented <0.05 ppm in these rotated crop matrices.

STUDY/WAIVER ACCEPTABILITY/DEFICIENCIES/CLARIFICATIONS:

Under the conditions and parameters used in the study, the confined rotational crop residue data are classified as scientifically acceptable. A confined rotational crop study with [^{14}C-5-pyrimidinyl]orthosulfamuron was separately submitted, refer to the DER for MRID 46578966. The petitioner cited in the subject confined rotational crop study that the results of the two studies were consistent and any quantitative differences in the TRR were likely due to the lower water solubility of DOP-urea (the major metabolite found in the PY-labeled study) compared to that of DBS-acid (the major metabolite found in the PH-labeled study) and, therefore, lower availability of residues for uptake into the plant from the soil

The acceptability of this study for regulatory purposes is addressed in the U.S. EPA Residue Chemistry Summary Document, D332290, D. Dotson, 2/14/2007.

COMPLIANCE:

Signed and dated Good Laboratory Practice (GLP), Quality Assurance and Data Confidentiality statements were provided. The GLP Compliance Statements cite that the study was not conducted in accordance with GLP Standards as defined by the USA EPA, but in compliance with OECD principles of GLP as defined by the Number 1 [ENV/MC/CHEM(98)17] on "The OECD principles of GLP;" Council Directives 88/320/EEC and 90/18/EEC as actuated by the Italian Decrees N° 120 of 1/27/92 and subsequent Ministerial Decree of 8/5/99; and EC directive 2004/9/EC of the European Parliament and of the Council.

A. BACKGROUND INFORMATION

Orthosulfamuron is a postemergence herbicide that Isagro S.p.A. is proposing for use on rice grown in the United States for the control of annual and perennial broadleaf weeds, sedges, and barnyard grass. Orthosulfamuron belongs to the sulfamoylurea class of herbicides. It reportedly acts by inhibiting the plant enzyme acetolactate synthase which is active in the biosynthesis of valine, leucine, and isoleucine.

TABLE A.1. Test Compound Nomenclature.

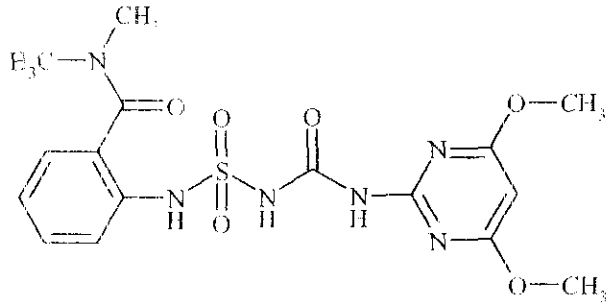
Compound	
Common name	Orthosulfamuron
Company experimental name	IR5878
IUPAC name	1-(4,6-dimethoxypyrimidin-2-yl)-3-[2-(dimethylcarbamoyl)phenylsulfamoyl]urea

TABLE A.1. Test Compound Nomenclature.	
CAS name	2-[[[[[(4,6-dimethoxy-2-pyrimidinyl)-amino]carbonyl]amino]sulfonyl]amino]-N,N-dimethylbenzamide
CAS registry number	213464-77-8
End-use product (EP)	0.51% G formulation (IR5878 0.5 GR; EPA Co. No. 80289) 51.5% WG formulation (IR5878 50 WG; EPA Co. No. 80289)

Table 2. Physicochemical Properties of the Technical Grade of Orthosulfamuron.		
Parameter	Value	Reference (MRID)
Color	White	46219004
Physical State	Fine Powder at 20°C	46219005
Odor	Odorless	46219006
pH	4.35 at 25°C (1% aqueous dispersion)	46219013
Density	1.45 g/mL at 20°C	46219008
Water solubility at 20°C	pH 4 buffer: 0.062 g/L pH 7 buffer: 0.63 g/L pH 8.5 buffer: 39 g/L	46219009
Solvent solubility at 20°C	n-heptane: 0.23 mg/L xylene: 130 mg/L acetone: 20 g/L ethyl acetate: 3.3 g/L dichloromethane: 56 g/L methanol: 8.3 g/L	Electronic communication, J. Messina to E. Kraft, 9/6/2006
Vapor pressure	1.1×10^{-4} at 20°C	46219010
Dissociation constant, pK_a	The test material becomes increasingly less soluble in water as the pH is lowered and undergoes degradation (hydrolysis) at neutral to acidic pHs. The test material is predicted to have 5 overlapping dissociation constants.	46219011
Octanol/water partition coefficient, Log(K_{OW})	pH 4: 2.0 pH 7: 1.3	46219012
UV/visible absorption spectrum	at pH 6.9, A=0.49 and $\varepsilon = 2.1 \times 10^4$ at 238 nm	46219001

B. EXPERIMENTAL DESIGN

Orthosulfamuron uniformly radiolabeled in the phenyl ring was radiodiluted with unlabeled orthosulfamuron and prepared in acetonitrile (ACN)/water. The radiolabeled test solution was applied micro-dropwise to the surface of sandy loam soil in 18 plastic pots (36 cm wide x 78 cm long x 30 cm high) at a nominal rate of 0.067 lb ai/A. After aging for ca. 30, 120, and 365 days, representative rotated crops of carrot (root vegetable), lettuce (leafy vegetable), and wheat (small grain) were planted in the treated soil. A single pot was used for planting of carrot and lettuce at each plantback interval (PBI), and four pots were used for planting of wheat at each PBI; additional pots were treated for soil analyses. Four pots of soil were treated with ACN/water for controls: one pot each for planting of carrot and lettuce, and two pots for planting of wheat.

Plants were grown to maturity outdoors in accordance with usual and customary agricultural practices for each crop in that location. Plants were treated with chemicals and fertilizers, and irrigated as needed. Temperature, relative humidity, and sunlight exposure were recorded daily.

Immature wheat forage was collected at the tillering growth stage. Carrot root and tops, lettuce leaves, as well as wheat grain and straw were collected at maturity.

B.1. Test Site and Crop Information

TABLE B.1.1. Test Site Information.

Testing Environment and location	Soil characteristics						
	Type	% Sand	% Silt	% Clay	%OM	pH	CEC (meq/100 g)
Pots maintained outdoors at Isagro Ricerca Srl (Novara, Italy)	loamy sand	75.5	21.5	3	2.20	5.34	12.69

TABLE B.1.2. Crop Information.

Crop; crop group	Variety	Plantback intervals (days)	Growth stage at harvest	Harvested Matrix	Harvesting procedure
Carrot; Vegetable, root and tuber, group 1, and Vegetable, leaves of root and tuber, group 2	Mezzalunga Nantese 3	33, 128, 373	Maturity: 126 DAP	root and tops	Whole plant pulled from the pot; excess soil was brushed off; root and top were separated and the root washed with water.
Lettuce; Vegetable, leafy, except brassica, group 4	Kagraner Sommer 2	33, 128, 373	Maturity: 43 DAP	leaves	Plants cut close to the soil surface and rinsed with water.
Wheat; Grain, cereal, group 15, and Grain, cereal, forage, fodder, and straw, group 16	Gemini	29, 121, 365	Immature (tillering stage): 66 DAP Maturity: 118 DAP	forage grain and straw	Plants cut close to the soil surface; mature plants were separated into grain and straw.

B.2. Test Materials

TABLE B.2.1. Test Material Characteristics.

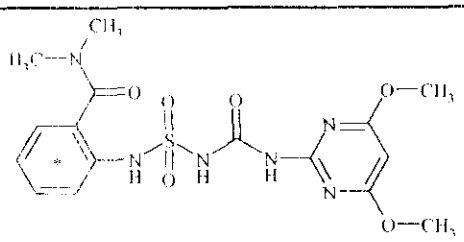
Chemical structure	
Radiolabel position	[^{14}C-U-phenyl]orthosulfamuron (PH-label)
Lot No.	182 and 209
Purity	>98% (TLC and HPLC)

TABLE B.2.1. Test Material Characteristics.	
Specific activity	Lot 182 test substance: 5.700 MBq/mg (154.058 µCi/mg) Lot 209 test substance: 7.357 MBq/mg (198.850 µCi/mg) Radiodiluted test solutions: 3.959-4.131 MBq/mg (106.998-110.660 µCi/mg)

B.3. Study Use Pattern

TABLE B.3.1. Use Pattern Information.	
Chemical name	[^{14}C-U-phenyl]orthosulfamuron
Application method	The radiolabeled test material was radiodiluted with unlabeled orthosulfamuron and prepared in ACN:water (3:7, v:v) on the day of application, and applied micro-dropwise to the bare soil.
Application rate	77.30-90.70 g ai/ha Mean of applications made to treated pots = 0.073 lb ai/A (81.84 g ai/ha)
Number of application (s)	One
Timing of application (s)	Preplant to bare soil
DAT[1] (days)	Target plantback intervals of 30, 120, and 365 days; refer to Table B.1.2. for actual plantback intervals

[1] DAT = Days after treatment

B.4. Identification/Characterization of Residues

B.4.1. Sample Handling and Preparation

All rotated crop samples were immediately stored frozen (-20°C) until they were processed. For processing, whole crop samples were cut into pieces and finely ground with dry ice. The homogenized samples were frozen until extraction and analysis.

Rotational crop commodities with a TRR >0.01 ppm were subjected to extraction procedures. Carrot root from all PBIs, 373-day PBI carrot tops, lettuce from all PBIs, 365-day PBI wheat forage, and wheat grain from all PBIs were not extracted. A brief discussion of the extraction processes follows.

An aliquot of the rotated crop matrix was extracted three times with ACN:50 mM ammonium bicarbonate (NH_4HCO_3; 5:5, v:v), then once with acetone, and centrifuged. Each supernatant was separately collected and brought to volume with additional extraction solvent for radioassay. The ACN extracts of carrot leaves (33- and 128-day PBIs), wheat forage (29- and 121-day PBIs) and wheat straw (from all PBIs), with >0.01 ppm extracted residues, were sequentially partitioned with dichloromethane (DCM) and ethyl acetate, after concentration to aqueous. The resulting DCM, ethyl acetate, and aqueous phases were brought to volume with the respective solvent and reserved for TLC analysis.

Nonextractable residues (>10% TRR) of carrot leaves (33- and 128-day PBIs) and wheat straw (all PBIs) were fractionated to determine incorporation into natural components. The bound residue was refluxed with 5% sodium hydroxide (110°C for 3 hours) and centrifuged to isolate

the cellulose fraction (solids) from the soluble fraction. The soluble fraction was then acidified with 6 N HCl (pH 2) to precipitate the lignin fraction.

The extraction flowcharts are presented in Figures B.4.1.1 (crops), B.4.1.2 (ACN extract), and B.4.1.3 (nonextractable residues). These flowcharts were copied from MRID 46578988 without alteration.

Figure B.4.1.1. Extraction Scheme for Rotated Crop Matrices.

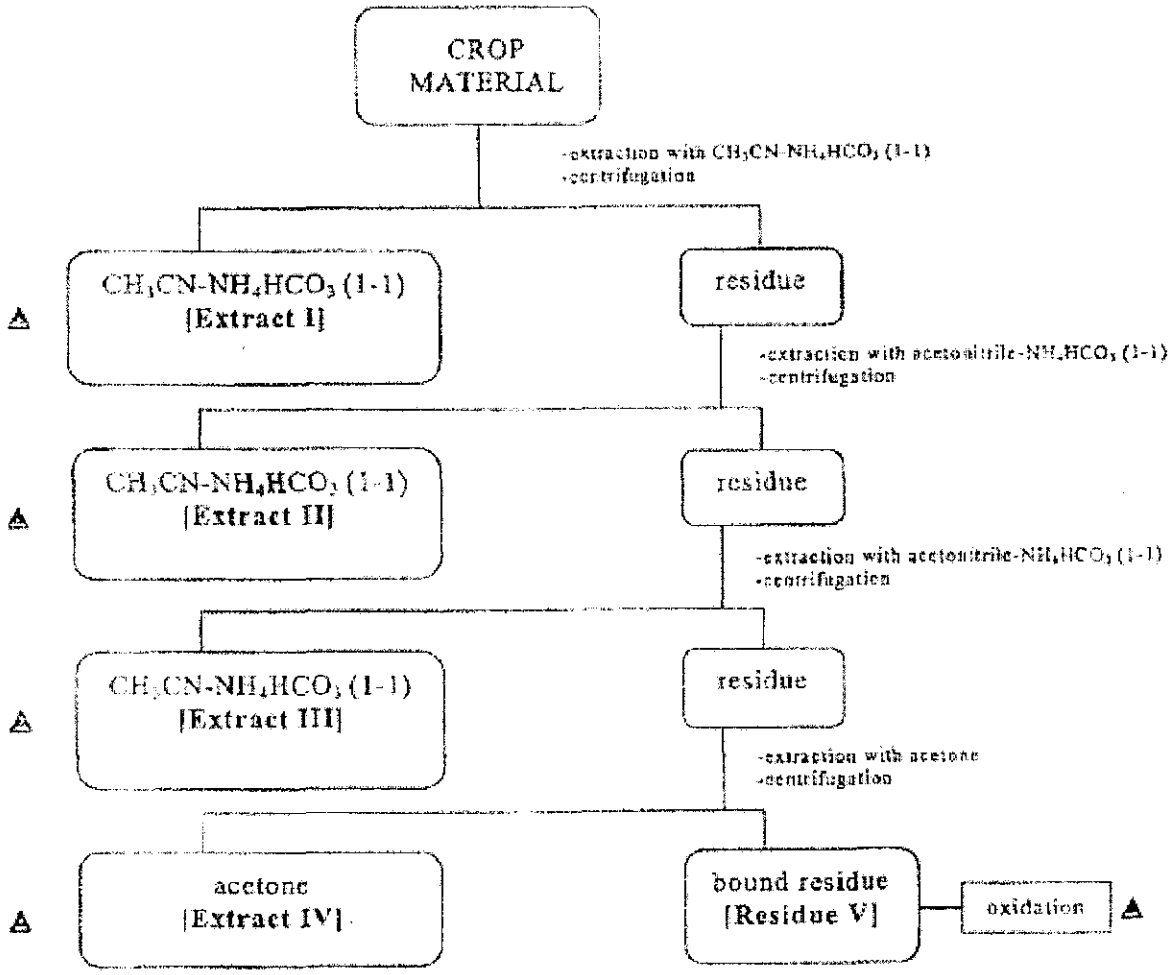


Figure B.4.1.2. Partitioning Scheme for the ACN Extract.

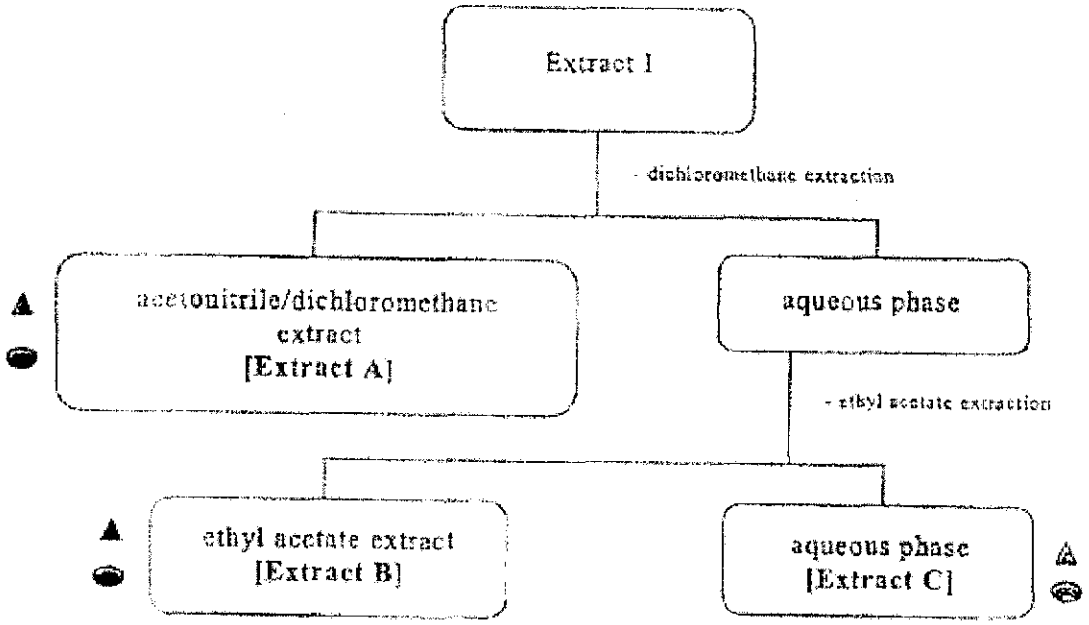
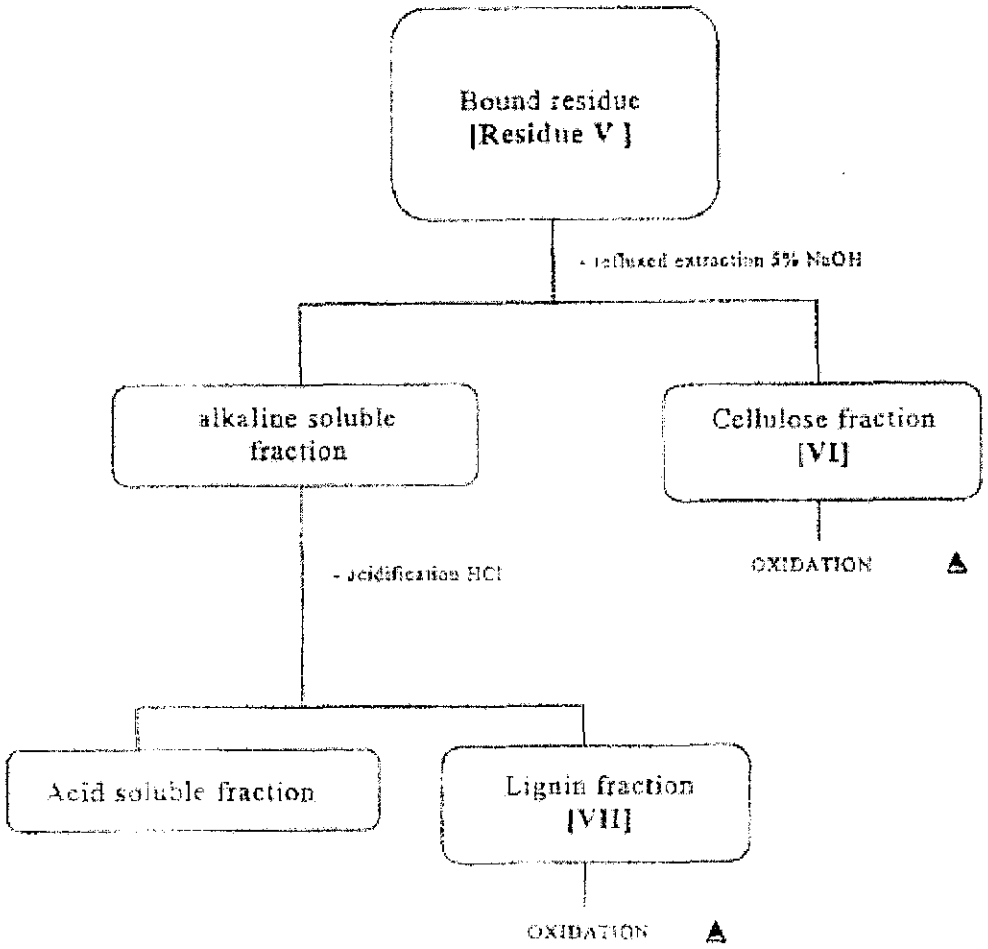


Figure B.4.1.3. Fractionation Scheme for Nonextractable Residues.



B.4.2. Analytical Methodology

Total radioactive residues (TRR) in rotational crop matrices were determined by combustion/LSC of the finely ground sample. Aliquots of ground carrot root and leaves, lettuce, and wheat forage were freeze-dried to remove water prior to combustion. Extracts and hydrolysates were radioassayed by LSC and nonextractable residues were radioassayed by combustion/LSC. The limit of determination for LSC determinations was twice the background.

The DCM, ethyl acetate, and aqueous phases of the ACN:NH_4HCO_3 extract of carrot leaves, wheat forage, and wheat straw were each subjected to TLC analysis for identification and quantitation of metabolites. Normal phase TLC analyses were conducted using silica gel 60 F_{254} plates and a solvent system of chloroform:methanol:ammonium hydroxide (70:27:3, v:v:v). Reverse phase TLC analyses were conducted using RP-18 F_{254S} plates and a solvent system of ACN:water (92:8, v:v). Samples were analyzed by both normal and reverse phase TLC systems, except for the aqueous phase which could only be analyzed by normal phase TLC; normal phase TLC was used for quantitation of residues because of better separation of the ^{14}C-compounds.

TLC plates were evaluated using radioimaging; luminescence was detected by a photo multiplier. Extracts from different PBIs, extracts from different crops, and different extracts from one crop were co-chromatographed to compare the metabolic profiles; 29-day PBI wheat straw was chosen as representative of all rotated crops and was used for metabolite identification. Metabolites were identified by co-chromatography with radiolabeled reference standards using both TLC systems (except for the aqueous phase which could only be analyzed using normal phase TLC); chemical names and structures for the reference standards are presented in Appendix I.

To facilitate identification of polar metabolites, 3-week old wheat plants were cut and dipped in aqueous solutions of radiolabeled orthosulfamuron, DBS-acid, or DB-amine. After 24 hours the cut plants were placed in fresh water and maintained in a growth chamber for ca. 36 hours. Treatment with DBS-acid was unsuccessful because the cut plant could not survive the low PH of the treatment solution. The treated cut plants were extracted with ACN:NH_4HCO_3 and centrifuged; the extracts were reserved for TLC analysis, and co-chromatographed with metabolites isolated from 29-day PBI wheat straw by preparative TLC.

To aid in characterizing polar metabolites, the aqueous extract of 29-day PBI wheat straw was subjected to enzyme and acid hydrolyses. Separate aliquots of the extract were incubated at 25°C for 48 hours with β-glucosidase in 0.1 M acetate buffer (pH 4.8) or esterase in 0.1 M ammonium sulfate buffer (pH 8). Polar metabolites isolated from the aqueous extract were subjected to acid hydrolysis under different conditions: (i) 0.1 N HCl at 40 °C for 4 hours; (ii) 2.0 N HCl at 40 °C for 4 hours; (iii) 4.0 N HCl at 40 °C for 5 hours; (iv) 4.0 N HCl at 70 °C for 2.5 hours; (v) 4.0 N HCl at 80 °C for 4 hours; or (vi) 6.0 N HCl at 80 °C for 4 hours. Metabolite 2, hydrolyzed with 6 N HCl at 80 °C for 4 hours, was co-chromatographed with *o*-aminobenzoic acid, ^{14}C-DBS-acid, and ^{14}C-DB-amine acid reference standards using normal and reverse phase TLC.

C. RESULTS AND DISCUSSION

The storage conditions and intervals for rotational crop samples are presented in Table C.1. All samples were analyzed for TRR within 2 months of harvest. Samples were extracted and analyzed by TLC within 5 months of harvest, therefore, no storage stability data are required to support the sample storage intervals and conditions of the confined rotational crop study.

After a single application of [^{14}C-U-phenyl]orthosulfamuron (PH label) to bare soil at 0.073 lb ai/A, total radioactive residues (TRR) accumulated at ≥0.01 ppm in the following: carrot tops planted 33 and 128 days after application, wheat forage planted 29 and 121 days after application, and wheat straw planted 29, 121, and 365 days after application. TRR were below 0.01 ppm in carrot root, lettuce, and wheat grain rotated crop matrices at all plantback intervals (PBIs), and in carrot tops and wheat forage planted at the 373/365-day PBI. In general, TRR were highest at the ~30 or 120-day PBI, and greatly reduced at the ~365-day PBI. Residues in carrot tops were 0.0442 ppm at the 33-day PBI, 0.0368 ppm at the 128-day PBI, and 0.0063 ppm at the 373-day PBI; residues in wheat forage were 0.0286 ppm at the 29-day PBI, 0.0310 ppm at

the 121-day PBI, and 0.0019 ppm at the 365-day PBI; and residues in wheat straw were 0.5763 ppm at the 29-day PBI, 0.8730 ppm at the 121-day PBI, and 0.1458 ppm at the 365-day PBI.

Rotated crop matrices with TRR >0.01 ppm were extracted; the extraction profiles and distribution of the radioactivity in these rotational crops are presented in Tables C.2.2.1 (carrot tops and wheat forage, 33/29- and 128/121-day PBIs) and C.2.2.2. (wheat straw, all PBIs). The majority of the radioactivity (72-94% TRR) was extracted from the rotated crop matrices using ACN/ammonium bicarbonate. Small amounts of radioactivity (1-5% TRR) were subsequently released with acetone. The ACN extracts were partitioned into DCM, ethyl acetate, and aqueous soluble phases for metabolite analysis. The majority of the extractable residue was aqueous soluble. Nonextractable residues in the extracted crop matrices were <0.01 ppm in carrot tops and wheat forage and 0.031-0.074 ppm (7-21% TRR) in wheat straw. Nonextractable residues were partially characterized as cellulose and lignin (see below) and accountabilities ranged from 89 to 101%. The extraction procedures extracted sufficient residues from all PBIs.

The characterization and identification of residues in rotational crop matrices which were analyzed are summarized in Tables C.2.3.1. (carrot tops and wheat forage, 33/29- and 128/121-day PBIs) and C.2.3.2. (wheat straw, all PBIs). Total identified residues ranged from 51 to 76% TRR in rotated carrot tops, wheat forage, and wheat straw. The parent, orthosulfamuron, was only identified in rotated wheat straw (all PBIs) at trace levels (<1% TRR; 0.001-0.004 ppm).

The major metabolite identified in rotated crop matrices was DBS-acid. DBS-acid accounted for 63.2% and 75.7% TRR (0.028 ppm) in carrot tops from the 33- and 128-day PBIs, respectively; 50.8% and 51.1% TRR (0.015-0.016 ppm) in wheat forage from the 29- and 121-day PBIs, respectively; and 63.7%, 65.8%, and 44.8% TRR (0.066-0.575 ppm) in wheat straw from the 29-, 121-, and 365-day PBIs, respectively. DBS-amide was identified as a minor metabolite in wheat forage and straw accounting for 9.2-9.8% TRR (0.003 ppm) in wheat forage from the 29- and 121-day PBIs, and 3.2-4.1% TRR (0.005-0.028 ppm) in wheat straw from all PBIs. DBS-amide was not identified in carrot tops. DB-amine was identified as a minor residue present only in wheat straw (<1-2.6% TRR; 0.004-0.005 ppm). Identifications of orthosulfamuron, DB-amine and DB-amide in the organic extracts of 30-day PBI straw, and DBS-acid in the aqueous extract of 30-day PBI straw were confirmed using normal phase and reverse phase TLC co-chromatography with the respective radiolabeled reference standard.

An unknown metabolite, characterized as a conjugate of DBS-acid, was determined to be a significant residue in wheat forage and accounted for 28.2% TRR (0.008 ppm) at the 29-day PBI and 19.2% TRR (0.006 ppm) at the 121-day PBI; Unknown 2 was found in carrot tops (1.9-6.5% TRR, ≤0.003 ppm) and wheat straw (5.4-8.6% TRR, 0.008-0.068 ppm) at minor levels. Another unknown metabolite, characterized as two N-glucosides of hydroxylated DB-amine, was determined to be a minor residue in carrot tops (3.8-4.7% TRR, 0.002 ppm), wheat forage (5.9-6.5% TRR, 0.002 ppm), and wheat straw (4.2-5.7% TRR, 0.008-0.050 ppm). The remaining extractable residues were characterized as one or two unknowns in the aqueous phase present at <0.01 ppm in carrot tops and wheat forage and ≤0.02 ppm in wheat straw, and a single unknown in the organic phases present at <0.01 ppm in wheat straw.

Unknown metabolite 2 from the aqueous extract of 30-day PBI straw was not hydrolyzed by β-glucosidase or esterase, but was fractionated into three metabolites (a, b, and c) with acid hydrolysis (6 N HCl at 80°C). Compound a was identified as DB-amine and Compound c was identified as *o*-aminobenzoic acid using normal and reverse phase TLC co-chromatography with reference standards. However, metabolite 2 was determined not to be a conjugate of DB-amine, because it was found only in the cut plant treated with orthosulfamuron and not the cut plant treated with DB-amine. Because DBS-acid is known to hydrolyze readily to DB-amine under acidic conditions and based on the enzyme and acid hydrolysis results, Unknown metabolite 2 was characterized as a conjugate of DBS-acid. In addition, as enzyme hydrolysis was unsuccessful, metabolite 2 is likely to be a conjugate with more glucose molecules, and because acid hydrolysis under weaker conditions was unsuccessful, metabolite 2 was not an N-glucoside.

Unknown metabolite 3 from the aqueous extract of 30-day PBI straw was fractionated into two metabolites. Neither compound was hydrolyzed by β-glucosidase or esterase, but both were easily hydrolyzed with acid hydrolysis (0.1 N HCl at 40°C) and yielded a new compound slightly more polar than DB-amine (likely by hydroxylation of the aromatic ring). Compounds 3a and 3b were characterized as N-glucosides of hydroxylated DB-amine. Compounds with similar TLC retention times were also observed in the extracts of cut plants treated with DB-amine.

All extracted crop matrices with nonextractable residues >10% TRR (or >0.05 ppm) were treated with base and acid to precipitate cellulose- and lignin-bound residues. Cellulose and lignin bound residues accounted for ~7-8% TRR in carrot tops (33- and 128-day PBIs), and 5-11% TRR in wheat straw (all PBIs); the uncharacterized nonextractable residues represented <0.05 ppm in these rotated crop matrices.

C.1. Storage Stability

Samples of rotated crop matrices were stored frozen (-20°C) after harvest. All samples were ground within 2-20 days of harvest, and TRR were determined within 1-27 days of grinding. Only the carrot top and wheat forage and straw samples were chromatographically analyzed, and analysis occurred within ~6 months of harvest. Extracts were stored at 4°C until analysis.

Matrix	Plantback interval (days)	Storage Temp. (°C)	Actual Storage Duration [1]
Carrot root	33, 128, 373	-20	19 days (TRR analysis)
Carrot tops	33, 128 373	-20	49 days (<2 months to TLC analysis) 42 days (TRR analysis)
Lettuce	33, 128, 373	-20	28 days (TRR analysis)
Wheat, forage	29, 121 365	-20	140 days (<5 months to TLC analysis) 22 days (TRR analysis)
Wheat, straw	29 121, 365	-20	26-27 days (<1 month to TLC analysis) 74 days (<3 months to TLC analysis)
Wheat, grain	29, 121 365	-20	125-127 days (~4 months to extraction) 14 days (TRR analysis)

[1] Only the carrot tops, wheat forage, and wheat straw extracts were analyzed; these samples were analyzed within 1-7 days of extraction.

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

TABLE C.2.1. Total Radioactive Residues (TRR) in Rotated Crop Matrices and the Treated Soil.

Matrix	Plantback interval (days)	PH Label (ppm)
Carrot, root	33	0.0057
	128	0.0043
	373	0.0007
Carrot, tops	33	0.0442
	128	0.0368
	373	0.0063
Lettuce, leaves	33	0.0042
	128	0.0020
	373	0.0003
Wheat, forage	29	0.0286
	121	0.0310
	365	0.0019
Wheat, straw	29	0.5763
	121	0.8730
	365	0.1458
Wheat, grain	29	0.0094
	121	0.0087
	365	0.0024
Soil [1]	0	0.069
	29 (wheat planting)	0.027
	33 (carrot and lettuce planting)	0.033
	121 (wheat planting)	0.046
	128 (carrot and lettuce planting)	0.035
	365 (wheat planting)	0.031
	373 (carrot and lettuce planting)	0.017

[1] The petitioner reported the mean values obtained from all analyses on soils with the same aging period; TRR were calculated by the summation of extractable (acetone) and nonextractable residues in soil.

TABLE C.2.2.1. Distribution of the Parent and the Metabolites in Rotational Carrot Tops and Wheat Forage Following Application of [Phenyl-^{14}C]Orthosulfamuron at 0.073 lb ai/A. [1]

Metabolite Fraction	Carrot, tops 33-day PBI		Carrot, tops 128-day PBI		Wheat, forage 29-day PBI		Wheat, forage 121-day PBI	
	TRR = 0.0442 ppm		TRR = 0.0368 ppm		TRR = 0.0286 ppm		TRR = 0.0310 ppm	
	%TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm
ACN/NH_4HCO_3 Extract	73	0.0323	79	0.0294	94	0.0272	89	0.0275
-DCM phase	--	--	--	--	7.69	0.0022	8.39	0.0026
DBS-amide					7.69	0.0022	8.39	0.0026
-Ethyl acetate phase	2.50	0.0011	1.90	0.0007	6.99	0.0020	3.87	0.0012
DBS-amide	--	--	--	--	*1.40*	0.0004	*1.29*	0.0004
Unknown 2	2.50	0.0011	1.90	0.0007	*4.90*	0.0014	*1.94*	0.0006
Unknown 3 [2]	--	--	--	--	*0.70*	0.0002	*0.32*	0.0001
-Aqueous phase	74.43	0.0329	85.05	0.0320	83.21	0.0238	78.06	0.0242

TABLE C.2.2.1. Distribution of the Parent and the Metabolites in Rotational Carrot Tops and Wheat Forage Following Application of [Phenyl-^{14}C]Orthosulfamuron at 0.073 lb ai/A. [1]

Metabolite Fraction	Carrot, tops 33-day PBI		Carrot, tops 128-day PBI		Wheat, forage 29-day PBI		Wheat, forage 121-day PBI	
	TRR = 0.0442 ppm		TRR = 0.0368 ppm		TRR = 0.0286 ppm		TRR = 0.0310 ppm	
	%TRR	ppm	%TRR	ppm	%TRR	ppm	%TRR	ppm
DBS acid	63.16	0.0279	75.66	0.0278	50.76	0.0145	51.14	0.0159
Unknown 1	3.71	0.0016	4.72	0.0017	3.34	0.0010	4.24	0.0013
Unknown 2	*4.07*	0.0018	--	--	*24.48*	0.0070	*17.10*	0.0053
Unknown 3 [2]	*3.85*	0.0017	*4.62*	0.0017	*5.94*	0.0017	*5.48*	0.0017
Acetone	3	0.0012	5	0.0020	--	--	--	--
Nonextractable	12.67	0.0056	11.76	0.0044	6.29	0.0018	5.16	0.0016
-Cellulose solid	5.66	0.0025	4.35	0.0016				
-Lignin precipitate	2.04	0.0009	2.17	0.0008				

[1] Shading indicates that the extraction step and/or characterization analysis was not conducted for the matrix in question. Percent TRR values in *italics* were calculated by the study reviewer from the reported ppm value.
[2] Unknown 3 was further fractionated into two peaks present at 0.0011 ppm and 0.0006 ppm in carrot tops, and ≤0.0012 ppm and 0.0007 ppm in wheat forage.

TABLE C.2.2.2. Distribution of the Parent and the Metabolites in Rotational Wheat Straw Following Application of [Phenyl-^{14}C]Orthosulfamuron at 0.073 lb ai/A. [1]

Metabolite Fraction	Wheat, straw 29-day PBI		Wheat, straw 121-day PBI		Wheat, straw 365-day PBI	
	TRR = 0.5763 ppm		TRR = 0.8730 ppm		TRR = 0.1458 ppm	
	%TRR	ppm	%TRR	ppm	%TRR	ppm
ACN/NH_4HCO_3 Extracts	85	0.4918	85	0.7401	72	0.1043
-DCM phase	5.04	0.0291	3.91	0.0341	7.13	0.0104
Orthosulfamuron	*0.38*	0.0022	0.22	0.0019	*0.48*	0.0007
DBS-amide	*3.40*	0.0196	*2.67*	0.0233	*2.54*	0.0037
DB-amine	*0.76*	0.0044	0.59	0.0038	2.62	0.0040
Unknown 9	*0.49*	0.0028	0.59	0.0052	1.28	0.0019
-Ethyl acetate phase	3.40	0.0196	2.60	0.0227	3.77	0.0055
Orthosulfamuron	*0.38*	0.0022	--	--	--	--
DBS-acid	--	--	*0.29*	0.0025	--	--
DBS-amide	*0.68*	0.0039	*0.57*	0.0050	*0.75*	0.0011
DB-amine	*0.16*	0.0009	--	--	--	--
Unknown 2	*1.80*	0.0104	*1.51*	0.0132	*2.61*	0.0038
Unknown 3	*0.29*	0.0017	*0.24*	0.0021	*0.41*	0.0006
Unknown 9	*0.09*	0.0005	--	--	--	--
-Aqueous phase	77.62	0.4473	79.62	0.6951	61.80	0.0901
DBS-acid	63.66	0.3669	*65.52*	0.5720	44.81	0.0655
Unknown 1	3.24	0.0187	2.36	0.0206	4.97	0.0073
Unknown 2 [2]	*6.82*	0.0393	*6.25*	0.0546	*2.61*	0.0038
Unknown 3	*3.90*	0.0225	*5.50*	0.0480	*5.21*	0.0076
Unknown 5	--	--	--	--	4.12	0.0060
Acetone	1	0.0044	1	0.0119	--	--
Nonextractable	12.85	0.0741	6.99	0.0610	20.99	0.0306
-Cellulose solid	2.76	0.0159	2.94	0.0257	6.88	0.0100

TABLE C.2.2.2. Distribution of the Parent and the Metabolites in Rotational Wheat Straw Following Application of [Phenyl-^{14}C]Orthosulfamuron at 0.073 lb ai/A. [1]

Metabolite Fraction	Wheat, straw 29-day PBI		Wheat, straw 121-day PBI		Wheat, straw 365-day PBI	
	TRR = 0.5763 ppm		TRR = 0.8730 ppm		TRR = 0.1458 ppm	
	%TRR	ppm	%TRR	ppm	%TRR	ppm
-Lignin precipitate	2.48	0.0143	2.05	0.0179	4.25	0.0062

[1] Percent TRR values in *italics* were calculated by the study reviewer from the reported ppm value.
[2] Unknown 2 was characterized in the aqueous phase of 30-day PBI straw as a conjugate of DBS-acid by enzyme and acid hydrolyses and co-TLC with treated cut plants.
[3] Unknown 3 was further fractionated into two peaks (3a and 3b) present at ≤0.0316 ppm and ≤0.0184 ppm (totals) in wheat straw, and characterized in the aqueous phase of 30-day PBI straw as N-glucosides of hydroxylated DB-amine by enzyme and acid hydrolyses and co-TLC with treated cut plants.

TABLE C.2.3.1. Summary of Characterization and Identification of Radioactive Residues in Rotational Carrot Tops and Wheat Forage Following Application of [Phenyl-^{14}C]Orthosulfamuron at 0.073 lb ai/A.

Compound	Carrot, tops 33-day PBI		Carrot, tops 128-day PBI		Wheat, forage 29-day PBI		Wheat, forage 121-day PBI	
	TRR = 0.0442 ppm		TRR = 0.0368 ppm		TRR = 0.0286 ppm		TRR = 0.0310 ppm	
	% TRR	ppm	% TRR	ppm	% TRR	ppm	% TRR	ppm
DBS-acid	63.16	0.0279	75.66	0.0278	50.76	0.0145	51.14	0.0159
DBS-amide	--	--	--	--	9.15	0.0026	9.77	0.0030
Unknown 2 [1]	6.49	0.0029	1.90	0.0007	28.16	0.0081	19.24	0.0060
Unknown 3a [2]	2.40	0.0011	2.95	0.0011	4.10	0.0012	3.75	0.0011
Unknown 3b [2]	1.40	0.0006	1.72	0.0006	2.39	0.0007	2.19	0.0007
Other Unknowns	3.71	0.0016	4.72	0.0017	3.34	0.0010	4.24	0.0013
Acetone extractable	3	0.0012	5	0.0020	--	--	--	--
Cellulose	5.66	0.0025	4.35	0.0016	--	--	--	--
Lignin	2.04	0.0009	2.17	0.0008	--	--	--	--
Total identified	63.16	0.0279	75.66	0.0278	59.91	0.0171	60.91	0.0189
Total characterized	24.70	0.0108	22.81	0.0085	37.99	0.0110	29.42	0.0091
Total extractable	76	0.0335	84	0.0314	94	0.0272	89	0.0275
Unextractable (PES) [3]	12.67	0.0056	11.76	0.0044	6.29	0.0018	5.16	0.0016
Accountability [4]	88.5		97.3		101		93.9	

[1] Unknown 2 was characterized in the aqueous phase of 30-day PBI straw as a conjugate of DBS-acid.
[2] Unknowns 3a and 3b were characterized in the aqueous phase of 30-day PBI straw as N-glucosides of hydroxylated DB-amine.
[3] Residues remaining after exhaustive extractions. Nonextractable residues were further characterized in carrot tops as cellulose- and lignin-bound residues precipitated under basic and acidic conditions (as reported in the table); uncharacterized nonextractable residues were ≤0.01 ppm.
[4] Accountability = (Total extractable + Total unextractable)/(TRR from combustion analysis; see TABLE C.2.1) * 100.

TABLE C.2.3.2. Summary of Characterization and Identification of Radioactive Residues in Rotational Wheat Straw Following Application of [Phenyl-^{14}C]Orthosulfamuron at 0.073 lb ai/A.

Compound	Wheat, straw 29-day PBI		Wheat, straw 121-day PBI		Wheat, straw 365-day PBI	
	TRR = 0.5763 ppm		TRR = 0.8730 ppm		TRR = 0.1458 ppm	
	% TRR	ppm	% TRR	ppm	% TRR	ppm
Orthosulfamuron	0.76	0.0044	0.22	0.0019	0.48	0.0007
DBS-acid	63.66	0.3669	65.81	0.5745	44.81	0.0655

TABLE C.2.3.2. Summary of Characterization and Identification of Radioactive Residues in Rotational Wheat Straw Following Application of [Phenyl-^{14}C]Orthosulfamuron at 0.073 lb ai/A.

Compound	Wheat, straw 29-day PBI		Wheat, straw 121-day PBI		Wheat, straw 365-day PBI	
	TRR = 0.5763 ppm		TRR = 0.8730 ppm		TRR = 0.1458 ppm	
	% TRR	ppm	% TRR	ppm	% TRR	ppm
DBS-amide	4.08	0.0235	3.22	0.0281	3.28	0.0048
DB-amine	0.92	0.0053	0.59	0.0038	2.62	0.0040
Unknown 2 [1]	8.62	0.0497	7.78	0.0678	5.44	0.0076
Unknown 3a [2]	2.65	0.0153	3.62	0.0316	3.56	0.0052
Unknown 3b [2]	1.55	0.0089	2.11	0.0184	2.07	0.0030
Other Unknowns	3.82	0.0221	2.95	0.0258	10.37	0.0152
Acetone extractable	1	0.0044	1	0.0119	--	--
Cellulose	2.76	0.0159	2.94	0.0257	6.88	0.0100
Lignin	2.48	0.0143	2.05	0.0179	4.25	0.0062
Total identified	69.42	0.4001	69.84	0.6083	51.19	0.0750
Total characterized	21.88	0.1262	21.45	0.1872	32.57	0.0472
Total extractable	86	0.4962	86	0.7520	72	0.1043
Unextractable (PES) [3]	12.85	0.0741	6.99	0.0610	20.99	0.0306
Accountability [4]	99.0		93.1		92.5	

[1] Unknown 2 was characterized in the aqueous phase of 30-day PBI straw as a conjugate of DBS acid.
[2] Unknowns 3a and 3b were characterized in the aqueous phase of 30-day PBI straw as N-glucosides of hydroxylated DB-amine.
[3] Residues remaining after exhaustive extractions. Nonextractable residues were further characterized in wheat straw as cellulose- and lignin-bound residues precipitated under basic and acidic conditions (as reported in the table); uncharacterized nonextractable residues were <0.05 ppm.
[4] Accountability = (Total extractable + Total unextractable)/(TRR from combustion analysis; see TABLE C.2.1) * 100.

C.3. Proposed Metabolic Profile

FIGURE C.3.1. Proposed Metabolic Profile of Orthosulfamuron in Rotated Crops. (Based on metabolites identified in both [^{14}C-U-phenyl]- and [^{14}C-5-pyrimidinyl]IR5878 studies)

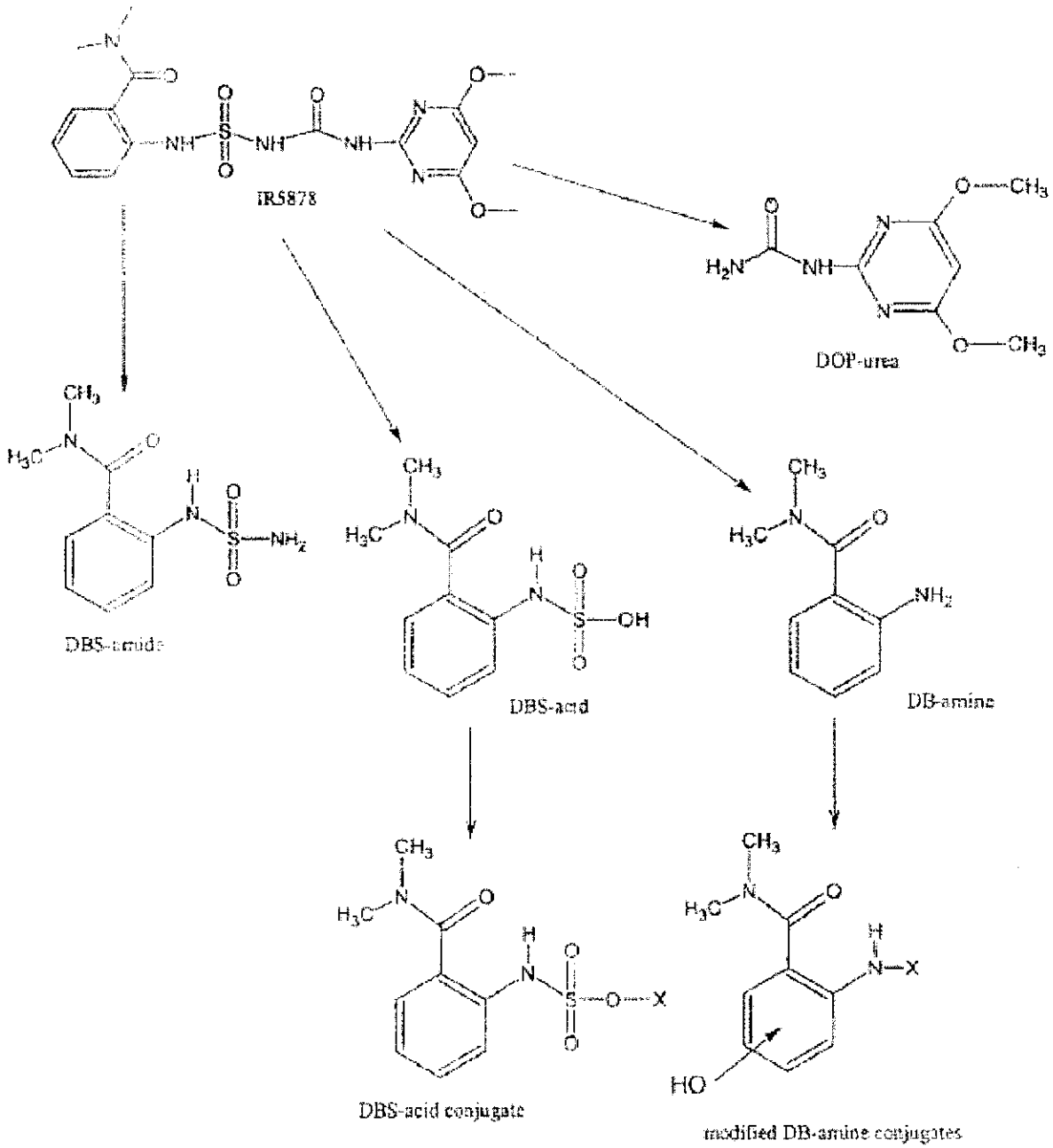


TABLE C.3.1. Identification of Compounds from the Confined Rotational Crop Study.

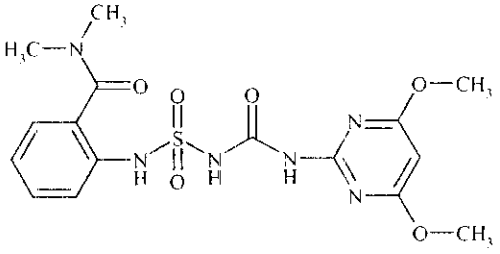
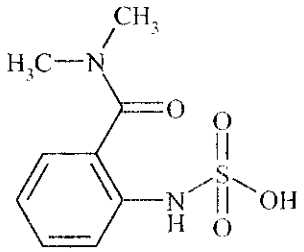
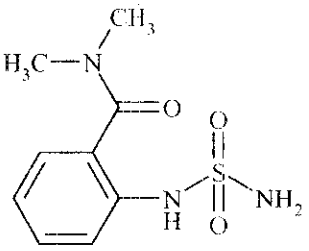
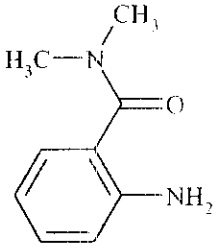
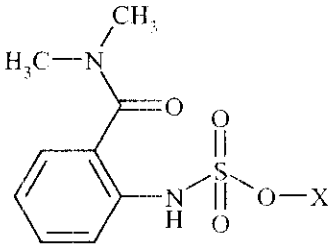
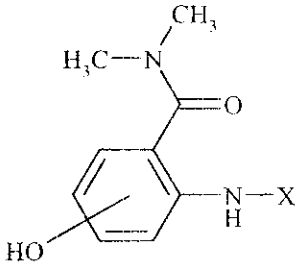
Common name/code Figure C.3.1 ID No.	Chemical name	Chemical structure
Orthosulfamuron	2-[[[[[(4,6-dimethoxy-2-pyrimidinyl)-amino]carbonyl]amino]sulfonyl]amino]-N,N-dimethylbenzamide	
DBS-acid	2-sulfoamino-N,N-dimethylbenzamide	
DBS-amide	2-sulfamoylamino-N, N-dimethylbenzamide	
DB-amine	2-amino-N,N-dimethylbenzamide	

TABLE C.3.1. Identification of Compounds from the Confined Rotational Crop Study.		
Common name/code Figure C.3.1 ID No.	Chemical name	Chemical structure
DBS-acid conjugate		
Hydroxy-DB-amine conjugates		

D. CONCLUSION

After a single application of [^{14}C-U-phenyl]orthosulfamuron (PH label) to bare soil at 0.073 lb ai/A, total radioactive residues (TRR) accumulated at ≥0.01 ppm in the following: carrot tops planted 33 and 128 days after application, wheat forage planted 29 and 121 days after application, and wheat straw planted 29, 121, and 365 days after application. TRR were below 0.01 ppm in carrot root, lettuce, and wheat grain rotated crop matrices at all plantback intervals (PBIs), and in carrot tops and wheat forage planted at the 373/365-day PBI. In general, TRR were highest at the ~30 or 120-day PBI, and greatly reduced at the ~365-day PBI. Residues in carrot tops were 0.0442 ppm at the 33-day PBI, 0.0368 ppm at the 128-day PBI, and 0.0063 ppm at the 373-day PBI; residues in wheat forage were 0.0286 ppm at the 29-day PBI, 0.0310 ppm at the 121-day PBI, and 0.0019 ppm at the 365-day PBI; and residues in wheat straw were 0.5763 ppm at the 29-day PBI, 0.8730 ppm at the 121-day PBI, and 0.1458 ppm at the 365-day PBI.

The majority of the radioactivity (72-94% TRR) was extracted from the rotated crop matrices (33/29- and 128/121-day PBI carrot tops and wheat forage, and wheat straw from all PBIs), using ACN/ammonium bicarbonate. Small amounts of radioactivity (1-5% TRR) were subsequently released with acetone. The ACN extracts were partitioned into DCM, ethyl acetate and aqueous soluble phases for metabolite analysis. The majority of the extractable residue was aqueous soluble. Nonextractable residues in the extracted crop matrices were <0.01 ppm in carrot tops and wheat forage, and 0.031-0.074 ppm (7-21% TRR) in wheat straw.

Total identified residues ranged from 51 to 76% TRR in rotated carrot tops, wheat forage, and wheat straw. The parent, orthosulfamuron, was only identified in rotated wheat straw (all PBIs) at trace levels (<1% TRR; 0.001-0.004 ppm).

The major metabolite identified in rotated crop matrices was DBS-acid. DBS-acid accounted for 63.2-75.7% TRR in carrot tops from the 33- and 128-day PBIs; 50.8-51.1% TRR in wheat forage from the 29- and 121-day PBIs; and 44.8-65.8% TRR in wheat straw from all PBIs. DBS-amide was identified as a minor metabolite (<10% TRR) in wheat forage and straw; DBS-amide was not identified in carrot tops. DB-amine was identified as a minor residue (<3% TRR) present only in wheat straw.

An unknown metabolite, characterized as a conjugate of DBS-acid, was determined to be a significant residue in wheat forage and accounted for 28.2% TRR at the 29-day PBI and 19.2% TRR at the 121-day PBI. Unknown 2 was found in carrot tops and wheat straw at minor (<9% TRR) levels. Another unknown metabolite, characterized as two N-glucosides of hydroxylated DB-amine, was determined to be a minor (<7% TRR) residue in carrot tops, wheat forage, and wheat straw. Nonextractable residues were characterized as cellulose- and lignin-bound residues (~5-11% TRR) in carrot tops (33- and 128-day PBIs) and wheat straw (all PBIs). Uncharacterized nonextractable residues represented <0.05 ppm in these rotated crop matrices.

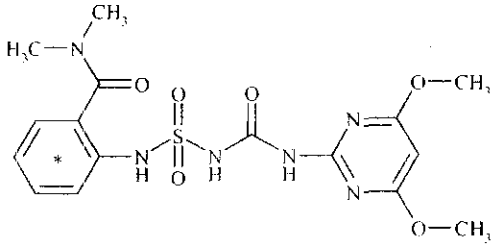
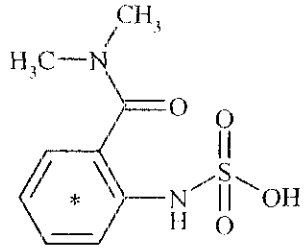
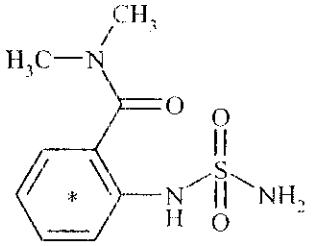
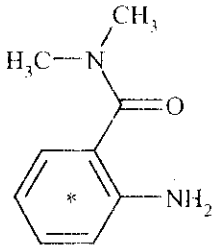
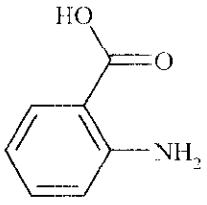
E. REFERENCES

None.

F. DOCUMENT TRACKING

Petition Number: 5F6967
DP Barcode: D319614
PC Code: 108209

APPENDIX I. Chemical Names and Structures of Reference Standards Used in Confined Rotational Crop Study.

Common name; Company code	Chemical name	Chemical structure
[^{14}C-phenyl]orthosulfamuron; IR5878	2-[[[[[(4,6-dimethoxy-2-pyrimidinyl)-amino]carbonyl]amino]sulfonyl]amino]-N,N-dimethylbenzamide	
[^{14}C-phenyl]DBS-acid	2-sulfoamino-N,N-dimethylbenzamide	
[^{14}C-phenyl] DBS-amide	2-sulfamoylamino-N, N-dimethylbenzamide	
[^{14}C-phenyl] DB-amine	2-amino-N,N-dimethylbenzamide	
o-aminobenzoic acid		

Orthosulfamuron/IR5878/PC Code 108209/Isagro S.p.A.
DACO 7.4.1/7.4.2/OPPTS 860.1500/OECD IIA 6.3.1, 6.3.2, 6.3.3 and IIIA 8.3.1, 8.3.2, 8.3.3
Crop Field Trial - Rice

Primary Evaluator Douglas Dotson, Chemist, RAB2 D. Dotson Date: 2/14/2007

Peer Reviewer Dennis McNeilly, Chemist, RAB2 Date: 2/14/2007

This DER was originally prepared under contract by Dynamac Corporation (2275 Research Boulevard, Suite 300; Rockville, MD 20850; submitted 06/12/2006). The DER has been reviewed by the Health Effects Division (HED) and revised to reflect current Office of Pesticide Programs (OPP) policies.

STUDY REPORT:

46578964 Willard, T. (2004) Magnitude of the Residue of IR5878 in Rice Raw Agricultural Commodities: Final Study Report. Project Number: AA030701, A2_06.03/04, 1205W. Unpublished study prepared by American Agricultural Services and PTRL West, Inc. and Shoffner Farm Research. 210 p.

EXECUTIVE SUMMARY:

Isagro S.p.A. submitted field trial data for orthosulfamuron on rice. Fourteen field trials were conducted in the United States in Regions 4 (AR, LA, MS; 9 trials), 5 (MO; 1 trial), 6 (TX; 2 trials), and 10 (CA; 2 trials) during the 2003 growing season. At each test location, a single broadcast spray application of the 50% water-dispersible granular (WG) formulation was made at 0.066-0.070 lb ai/A (73.6-78.0 g ai/ha) to moist/wet soil (not flooded) when rice was in the 2-3 leaf stage. Application was made using ground equipment in ~15-21 gal/A with an adjuvant added to the spray mixture. Samples of mature rice grain and straw were harvested from all test sites 91-119 days after application.

Samples of rice grain and straw were analyzed for residues of orthosulfamuron using an LC/MS/MS method based on the "Enforcement Method (including Validation) for the Determination of Residues of IR 5878 in Rice Grain, Rice Green Plant and Rice Straw" as presented in Report ISA-0102V, Dr. Specht & Partner, 2002. This method is adequate for data collection based on acceptable concurrent method recoveries. The validated LOQ was 0.05 ppm in/on rice grain and straw, and the limit of detection was 0.02 ppm.

The maximum storage interval of crop samples from harvest to analysis was 97 days (3.2 months) for rice grain and 113 days (3.7 months) for rice straw. The results of a storage stability study were submitted (refer to the 860.1380 DER for MRID 46578983) and indicate that residues of orthosulfamuron are stable under frozen storage conditions in/on fortified samples of rice grain and straw for up to 12 months. These data are adequate to support the storage intervals of samples from the rice field trials.

Residues of orthosulfamuron were each below the LOQ (<0.05 ppm) in/on all samples (n = 28) of rice grain and straw harvested at maturity following a single broadcast application of the 50% WG formulation at 0.066-0.070 lb ai/A to rice grown in un-flooded fields.

No residue decline data were included in the submission. These data are not required because application was made prior to the formation of the edible portion of the crop.

STUDY/WAIVER ACCEPTABILITY/DEFICIENCIES/CLARIFICATIONS:

Under the conditions and parameters used in the study, the field trial residue data are classified as scientifically acceptable. The acceptability of this study for regulatory purposes is addressed in the U.S. EPA Residue Chemistry Summary Document, D332290, D. Dotson, 2/14/2007.

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

Orthosulfamuron is a postemergence herbicide that Isagro S.p.A. is proposing for use on rice grown in the United States for the control of annual and perennial broadleaf weeds, sedges, and barnyard grass. Orthosulfamuron belongs to the sulfamoylurea class of herbicides. It reportedly acts by inhibiting the plant enzyme acetolactate synthase which is active in the biosynthesis of valine, leucine, and isoleucine.

TABLE A.1. Test Compound Nomenclature.

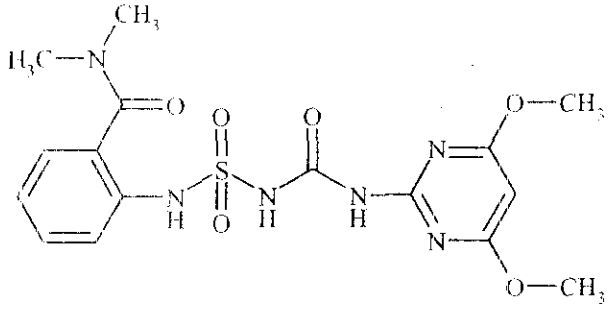
Compound	
Common name	Orthosulfamuron
Company experimental name	IR5878
IUPAC name	1-(4,6-dimethoxypyrimidin-2-yl)-3-[2-(dimethylcarbamoyl)phenylsulfamoyl]urea
CAS name	2-[[[[[(4,6-dimethoxy-2-pyrimidinyl)-amino]carbonyl]amino]sulfonyl]amino]-N,N-dimethylbenzamide
CAS registry number	213464-77-8
End-use product (EP)	0.51% G formulation (IR5878 0.5 GR; EPA Co. No. 80289) 51.5% WG formulation (IR5878 50 WG; EPA Co. No. 80289)

Table 2. Physicochemical Properties of the Technical Grade of Orthosulfamuron.

Parameter	Value	Reference (MRID)
Color	White	46219004
Physical State	Fine Powder at 20°C	46219005
Odor	Odorless	46219006
pH	4.35 at 25°C (1% aqueous dispersion)	46219013
Density	1.45 g/mL at 20°C	46219008
Water solubility at 20°C	pH 4 buffer: 0.062 g/L pH 7 buffer: 0.63 g/L pH 8.5 buffer: 39 g/L	46219009
Solvent solubility at 20°C	n-heptane: 0.23 mg/L xylene: 130 mg/L acetone: 20 g/L ethyl acetate: 3.3 g/L dichloromethane: 56 g/L methanol: 8.3 g/L	Electronic communication, J. Messina to E. Kraft, 9/6/2006
Vapor pressure	1.1×10^{-4} at 20°C	46219010
Dissociation constant, pK_a	The test material becomes increasingly less soluble in water as the pH is lowered and undergoes degradation (hydrolysis) at neutral to acidic pHs. The test material is predicted to have 5 overlapping dissociation constants.	46219011
Octanol/water partition coefficient, $Log(K_{OW})$	pH 4: 2.0 pH 7: 1.3	46219012
UV/visible absorption spectrum	at pH 6.9, A=0.49 and $\varepsilon = 2.1 \times 10^4$ at 238 nm	46219001

B. EXPERIMENTAL DESIGN

Fourteen field trials were conducted in the U.S. during the 2003 growing season in Regions 4 (AR, LA, MS; 9 trials), 5 (MO; 1 trial), 6 (TX; 2 trials), and 10 (CA; 2 trials).

Each field test consisted of one untreated plot and one treated plot (400-4000 sq. ft.), separated by a levee. A single broadcast application of a 50% water-dispersible granular (WG) formulation of orthosulfamuron was made to moist/wet soil (not flooded) when rice was at the 2-3 leaf stage. Application was made at 0.066-0.070 lb ai/A (73.6-78.0 g ai/ha) using ground equipment (pressurized CO_2 backpack boom sprayer) in ~15-21 gal/A with 0.2% (v:v) of surfactant added to the spray mixture.

Permanent flooding was established 3-10 days after treatment and irrigation was applied in order to maintain the flood level typical for the test area. Irrigation was stopped during the last 2-4 weeks of the study to allow the crop to dry down. The test crops were grown and maintained according to typical practices for each region. Fertilizer and maintenance pesticides were applied as needed. Trial site conditions are presented in Table B.1.1. The crop varieties grown are identified in Table C.3. Average minimum and maximum temperatures and total precipitation recordings for each month of the study period were reported, along with the average

10-year historical data. Weather conditions were similar to those reported in historical weather data.

Samples of rice grain and straw were harvested at maturity.

B.1. Study Site Information

TABLE B.1.1. Trial Site Conditions.

Trial Identification: City, State; Year (Trial ID #)	Soil characteristics			
	Type	%OM [1]	pH [1]	CEC [1] (meq/g)
Shoffner, AR; 2003 (AR1)	Sandy clay loam	1.2	7.5	NR
Tuckerman, AR; 2003 (AR2)	Sandy loam	0.6	7.3	NR
Cord, AR; 2003 (AR3)	Silty clay	1.7	7.7	NR
Porterville, CA; 2003 (CA1)	Clay loam	0.8	6.8	NR
Live Oak, CA; 2003 (CA2)	Clay loam	1	7.9	NR
Washington, LA; 2003 (LA1)	Silty loam	1.0	7.6	NR
Bunkie, LA; 2003 (LA2)	Clay	1.3	7.4	NR
Ville Platte, LA; 2003 (LA3)	Silty clay loam	1.4	7.3	NR
Dudley, MO; 2003 (MO1)	Silty loam	0.9	8.1	NR
Greenville, MS; 2003 (MS1)	Silty clay	2.4	7.5	NR
Shaw, MS; 2003 (MS2)	Silty clay	2.5	7.7	NR
Cleveland, MS; 2003 (MS3)	Silt loam	2.0	7.4	NR
Brookshire, TX; 2003 (TX1)	Sandy loam	0.8	7.1	NR
Fulshear, TX; 2003 (TX2)	Sandy loam	2.0	6.8	NR

[1] NR = Not recorded

TABLE B.1.2. Study Use Pattern.

Location: City, State; Year (Trial ID)	EP [1]	Application					Tank Mix/ Adjuvants [4]
		Method; Timing	Vol. (GPA [2]) [L/ha]	Rate (lb ai/A) [g ai/ha]	RTI [3] (days)	Total Rate (lb ai/A) [g ai/ha]	
Shoffner, AR; 2003 (AR1)	50% WG	1. Broadcast to unflooded fields; 2-3 leaf stage, crop height 4 inches	20.1 [187.9]	0.067 [75.0]	NA	0.067 [75.0]	ADJ 1012
Tuckerman, AR; 2003 (AR2)	50% WG	1. Broadcast to unflooded fields; 2-3 leaf stage, crop height 6-7 inches	20.2 [188.9]	0.067 [75.5]	NA	0.067 [75.5]	ADJ 009
Cord, AR; 2003 (AR3)	50% WG	1. Broadcast to unflooded fields; 2-3 leaf stage, crop height 6-7 inches	20.4 [190.7]	0.068 [76.2]	NA	0.068 [76.2]	ADJ 010
Porterville, CA; 2003 (CA1)	50% WG	1. Broadcast to unflooded fields; 2-3 leaf stage, crop height 4 inches	14.8 [138.4]	0.066 [73.9]	NA	0.066 [73.9]	ADJ 1012
Live Oak, CA; 2003 (CA2)	50% WG	1. Broadcast to unflooded fields; 3 leaf stage, crop height 6 inches	15 [140.3]	0.067 [74.6]	NA	0.067 [74.6]	ADJ 010

TABLE B.1.2. Study Use Pattern.

Location: City, State; Year (Trial ID)	EP[1]	Application					Tank Mix/ Adjuvants[4]
		Method; Timing	Vol. (GPA[2]) [L/ha]	Rate (lb ai/A) [g ai/ha]	RTI[3] (days)	Total Rate (lb ai/A) [g ai/ha]	
Washington, LA; 2003 (LA1)	50% WG	1. Broadcast to unflooded fields; 2-3 leaf stage, crop height 4-6 inches	20.9 [195.4]	0.067 [74.6]	NA	0.067 [74.6]	ADJ 1012
Bunkie, LA; 2003 (LA2)	50% WG	1. Broadcast to unflooded fields; 2 leaf stage, crop height 3 inches	20.6 [192.6]	0.067 [75.4]	NA	0.067 [75.4]	ADJ 009
Ville Platte, LA; 2003 (LA3)	50% WG	1. Broadcast to unflooded fields; 3 leaf stage, crop height 4-5 inches	20.3 [189.8]	0.067 [75.3]	NA	0.067 [75.3]	ADJ 010
Dudley, MO; 2003 (MO1)	50% WG	1. Broadcast to unflooded fields; 2-3 leaf stage, crop height 6-7 inches	15.7 [146.8]	0.066 [73.6]	NA	0.066 [73.6]	ADJ 010
Greenville, MS; 2003 (MS1)	50% WG	1. Broadcast to unflooded fields; 3-4 leaf stage, crop height 8 inches	18.3 [171.1]	0.069 [77.8]	NA	0.069 [77.8]	ADJ 1012
Shaw, MS; 2003 (MS2)	50% WG	1. Broadcast to unflooded fields; 2-3 leaf stage, crop height 5-6 inches	15.4 [144.0]	0.067 [75.3]	NA	0.067 [75.3]	ADJ 009
Cleveland, MS 2003 (MS3)	50% WG	1. Broadcast to unflooded fields; 2-3 leaf stage, crop height 5-6 inches	17.5 [163.6]	0.070 [78.0]	NA	0.070 [78.0]	ADJ 010
Brookshire, TX; 2003 (TX1)	50% WG	1. Broadcast to unflooded fields; 2-3 leaf stage, crop height 4-5 inches	19.8 [185.1]	0.068 [76.2]	NA	0.068 [76.2]	ADJ 1012
Fulshear, TX; 2003 (TX2)	50% WG	1. Broadcast to unflooded fields; 2-3 leaf stage, crop height 4 inches	20.1 [187.9]	0.067 [75.3]	NA	0.067 [75.3]	ADJ 010

[1] EP = End-use Product; IR5878 50 WG
[2] GPA = Gallons per acre [L/ha = Liters per hectare], application volume calculated by the petitioner (L/ha = GPA x 9.35).
[3] RTI = Retreatment Interval; NA = Not applicable, a single application was made.
[4] An adjuvant was added to the spray mixture at 0.2% (v/v).

TABLE B.1.3. Trial Numbers and Geographical Locations.

NAFTA Growing Zones	Rice		
	Submitted	Requested	
		Canada	U.S.
1			
1A			
2			
3			
4	9		7
5	1		1
5A			
5B			
6	2		2
7			
7A			
8			
9			
10	2		2
11			
12			
13			
14			
15			
16			
17			
18			
19			
20			
21			
Total	14 [1]		12 [2]

[1] Data from 2 additional field trials conducted with the G formulation in Zone 10 have been submitted separately (refer to the DER for MRID 46578986).

[2] The number of requested field trials represents a 25 percent reduction in the number of trials due to the pesticidal use resulting in no quantifiable residues.

B.2. Sample Handling and Preparation

A single untreated and duplicate treated samples of rice grain and straw were collected from each trial site 91-119 days after application. All samples were frozen at the field facility within 3 hours of sampling. Samples were shipped by freezer truck to PTRL West, Inc. (Hercules, CA), where samples were stored frozen (<-18°C) prior to analysis. Samples were homogenized in the presence of dry ice prior to residue analysis.

B.3. Analytical Methodology

Samples of rice grain and straw were analyzed for residues of orthosulfamuron at PTRL West, Inc. (Hercules, CA) using an LC/MS/MS method based on the "Enforcement Method (including Validation) for the Determination of Residues of IR 5878 in Rice Grain, Rice Green Plant and Rice Straw" as presented in Report ISA-0102V, Dr. Specht & Partner, 2002. A method description was included with the subject submission. The validated LOQ was 0.05 ppm in/on rice grain and straw. The limit of detection was 0.02 ppm (3 times the standard deviation of recoveries in matrix).

Briefly, homogenized samples of rice grain and straw were extracted twice with acetonitrile (ACN):0.02 M triethylamine (4:1, v:v) and filtered. Sodium chloride was added to the filtrate to induce separation of the aqueous and ACN phases. An aliquot of the ACN layer was then partitioned with hexane. The resulting ACN phase was collected and evaporated to dryness by rotary evaporation. Residues were redissolved in methanol, and water was added. The final sample solution was microfilterfuged to remove any particulate matter and analyzed by HPLC/MS/MS. Residues were quantitated using external standards.

C. RESULTS AND DISCUSSION

Sample storage conditions and intervals are summarized in Table C.2. The maximum storage interval of crop samples from harvest to analysis was 97 days (3.2 months) for rice grain and 113 days (3.7 months) for rice straw. The results of a storage stability study were submitted (refer to the 860.1380 DER for MRID 46578983) which indicate that residues of orthosulfamuron are stable under frozen storage conditions in/on fortified samples of rice grain and straw for up to 12 months. These data are adequate to support the storage intervals of samples from the rice field trials.

Concurrent recovery data are presented in Table C.1. The LC/MS/MS method is adequate for data collection based on acceptable concurrent recovery data. Concurrent recoveries ranged from 88% to 110% (mean = 97% with a standard deviation (s.d.) of 6%) for rice grain and 92-104% (mean = 99 with a s.d. of 4%) for rice straw each fortified with orthosulfamuron at 0.05 ppm. Sample chromatograms were supplied which indicated that the matrix was relatively free of interferences. Apparent residues of orthosulfamuron were nondetectable (<0.02 ppm) in/on 14 samples each of untreated rice grain and straw.

The results of the rice field trials are reported in Table C.3. A summary of the residue data is presented in Table C.4. Residues of orthosulfamuron were below the LOQ (<0.05 ppm) in/on all samples of rice grain and straw harvested at maturity following a single broadcast application of the 50% WG formulation at 0.066-0.070 lb ai/A to rice in unflooded fields.

The petitioner noted that rice in the treated plot at the TX1 field site appeared to be growing normally, but a significant amount of "blank" heads were present at harvest. The entire plot had to be harvested for sufficient sample.

TABLE C.1. Summary of Concurrent Recoveries of Orthosulfamuron from Rice Grain and Straw.

Matrix	Spike level (ppm)	Sample size (n)	Recoveries (%)	Mean ± std dev (%)
Rice grain	0.05	10	88, 92, 92, 94, 96, 98, 100, 100, 102, 110	97 ± 6
Rice straw	0.05	10	92, 94, 96, 96, 100, 100, 102, 102, 104, 104	99 ± 4

TABLE C.2. Summary of Storage Conditions.

Matrix (RAC)	Storage Temperature (°C)	Actual Storage Duration [1]	Interval of Demonstrated Storage Stability [2]
Rice grain	<-18	51-97 days (1.7-3.2 months)	Stable under frozen storage conditions in/on fortified samples of rice grain and straw for up to 12 months.
Rice straw	<-18	53-113 days (1.7-3.7 months)	

[1] All samples were analyzed within 0-9 days of extraction.
[2] Refer to 860.1380 DER for MRID 46578983.

TABLE C.3 Residue Data from Crop Field Trials with Orthosulfamuron.

Trial ID (City, State; Year)	Zone	Crop; Variety	Commodity or Matrix	Total Rate (lb ai/A) [g ai/ha]	PHI (days)	Orthosulfamuron Residues (ppm) [1]
Shoffner, AR; 2003 (AR1)	4	Rice; Francis	Grain	0.067 [75.0]	118	ND, ND
			Straw		118	ND, ND
Tuckerman, AR; 2003 (AR2)	4	Rice; Wells	Grain	0.067 [75.5]	103	ND, ND
			Straw		103	ND, ND
Cord, AR; 2003 (AR3)	4	Rice; Wells	Grain	0.068 [76.2]	115	ND, ND
			Straw		115	ND, ND
Porterville, CA; 2003 (CA1)	10	Rice; NFD-181	Grain	0.066 [73.9]	105	ND, ND
			Straw		105	ND, ND
Live Oak, CA; 2003 (CA2)	10	Rice; M 205	Grain	0.067 [74.6]	117	ND, ND
			Straw		117	ND, ND
Washington, LA; 2003 (LA1)	4	Rice; Cocodrie	Grain	0.067 [74.6]	91	ND, ND
			Straw		91	ND, ND
Bunkie, LA; 2003 (LA2)	4	Rice; Cocodrie	Grain	0.067 [75.4]	106	ND, ND
			Straw		106	ND, ND
Ville Platte, LA; 2003 (LA3)	4	Rice; Cocodrie	Grain	0.067 [75.3]	92	ND, ND
			Straw		92	ND, ND
Dudley, MO; 2003 (MO1)	5	Rice; Wells	Grain	0.066 [73.6]	105	ND, ND
			Straw		105	ND, ND
Greenville, MS; 2003 (MS1)	4	Rice; Cocodrie	Grain	0.069 [77.8]	107	ND, ND
			Straw		107	ND, ND
Shaw, MS; 2003 (MS2)	4	Rice; Cocodrie	Grain	0.067 [75.3]	119	ND, ND
			Straw		119	ND, ND
Cleveland, MS; 2003 (MS3)	4	Rice; Cocodrie	Grain	0.070 [78.0]	103	ND, ND
			Straw		103	ND, ND

TABLE C.3. Residue Data from Crop Field Trials with Orthosulfamuron.

Trial ID (City, State; Year)	Zone	Crop: Variety	Commodity or Matrix	Total Rate (lb ai/A) [g ai/ha]	PHI (days)	Orthosulfamuron Residues (ppm) [1]
Brookshire, TX; 2003 (TX1)	6	Rice: Cocodrie	Grain	0.068 [76.2]	112	ND, ND
			Straw		112	ND, ND
Fulshear, TX; 2003 (TX2)	6	Rice: Cocodrie	Grain	0.067 [75.3]	105	ND, ND
			Straw		105	ND, ND

[1] Residues below the method LOD (<0.02 ppm) are reported as ND (nondetectable).

TABLE C.4. Summary of Residue Data from Crop Field Trials with Orthosulfamuron.

Commodity	Total Applic. Rate (lb ai/A) [g ai/ha]	PHI (days)	Residue Levels (ppm) [1]						
			n	Min.	Max.	HAFT [2]	Median (STMdR)	Mean (STMR)	Std. Dev.
Rice grain	0.066-0.070 [73.6-78.0]	91-119	28	<0.05	<0.05	<0.05	<0.025	<0.025	N/A
Rice straw		91-119	28	<0.05	<0.05	<0.05	<0.025	<0.025	N/A

[1] The method LOQ was <0.05 ppm. The median, mean, and standard deviation were calculated using half the LOQ (<0.025 ppm) for all residues reported as ND in Table C.3.
[2] HAFT = Highest Average Field Trial

D. CONCLUSION

The submitted field trial data reflect the use of a broadcast application of a 50% WG formulation of orthosulfamuron at a rate of 0.066-0.070 lb ai/A (73.6-78.0 g ai/ha) to rice. Application was made to moist/wet soil (not flooded) when rice was in the 2-3 leaf stage. An acceptable method was used for quantitation of residues in/on rice grain and straw.

E. REFERENCES

None.

F. DOCUMENT TRACKING

Petition Number: 5F6957
DP Barcode: D319614
PC Code: 108209

Orthosulfamuron/IR5878/PC Code 108209/Isagro S.p.A.
DACO 7.4.1/7.4.2/OPPTS 860.1500/OECD IIA 6.3.1, 6.3.2, 6.3.3 and IIIA 8.3.1, 8.3.2, 8.3.3
Crop Field Trial - Rice

Primary Evaluator Douglas Dotson, Chemist, RAB2 D. Dotson Date: 2/14/2007

Peer Reviewer Dennis McNeilly, Chemist, RAB2 Date: 2/14/2007

This DER was originally prepared under contract by Dynamac Corporation (2275 Research Boulevard, Suite 300; Rockville, MD 20850; submitted 06/12/2006). The DER has been reviewed by the Health Effects Division (HED) and revised to reflect current Office of Pesticide Programs (OPP) policies.

STUDY REPORT:

46578986 Willard, T. (2005) Magnitude of the Residue of IR5878 0.5 GR in Rice Raw Agricultural Commodities: Final Study Report. Project Number: AA040702, A2_06.03/05, 1292W. Unpublished study prepared by American Agricultural Services, PTRL West, Inc. and Research for Hire. 100 p.

EXECUTIVE SUMMARY:

Isagro S.p.A. submitted field trial data for orthosulfamuron on rice. Two field trials were conducted in the United States in Region 10 (CA) during the 2004 growing season. At each test location, a single broadcast application of the 0.5% granular (G) formulation was made to emerged rice (3 leaf stage) growing in flooded rice paddies at 0.067 lb ai/A (75.0-75.1 g ai/ha). Application was made using ground equipment without an adjuvant. Samples of mature rice grain and straw were harvested 116-136 days after application.

Samples of rice grain and straw were analyzed for residues of orthosulfamuron using an LC/MS/MS method based on the "Enforcement Method (including Validation) for the Determination of Residues of IR 5878 in Rice Grain, Rice Green Plant and Rice Straw" as presented in Report ISA-0102V, Dr. Specht & Partner, 2002. This method is adequate for data collection based on acceptable concurrent method recoveries. The validated LOQ was 0.05 ppm in/on rice grain and straw, and the limit of detection was 0.02 ppm.

The maximum storage interval of crop samples from harvest to analysis was 78 days (2.6 months) for rice grain and 79 days (2.6 months) for rice straw. The results of a storage stability study were submitted (refer to the 860.1380 DER for MRID 46578983) and indicate that residues of orthosulfamuron are stable under frozen storage conditions in/on fortified samples of rice grain and straw for up to 12 months. These data are adequate to support the storage intervals of samples from the rice field trials.

Residues of orthosulfamuron were below the LOQ (<0.05 ppm) in/on all four samples of rice grain and straw harvested at maturity following a single broadcast application of the 0.5% G formulation at 0.067 lb ai/A to rice in flooded rice paddies.

No residue decline data were included in the submission. These data are not required because application was made prior to the formation of the edible portion of the crop.

STUDY/WAIVER ACCEPTABILITY/DEFICIENCIES/CLARIFICATIONS:

Under the conditions and parameters used in the study, the field trial residue data are classified as scientifically acceptable. The acceptability of this study for regulatory purposes is addressed in the U.S. EPA Residue Chemistry Summary Document, D332290, D. Dotson, 2/14/2007.

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

Orthosulfamuron is a postemergence herbicide that Isagro S.p.A. is proposing for use on rice grown in the United States for the control of annual and perennial broadleaf weeds, sedges, and barnyard grass. Orthosulfamuron belongs to the sulfamoylurea class of herbicides. It reportedly acts by inhibiting the plant enzyme acetolactate synthase which is active in the biosynthesis of valine, leucine, and isoleucine.

TABLE A.1. Test Compound Nomenclature.

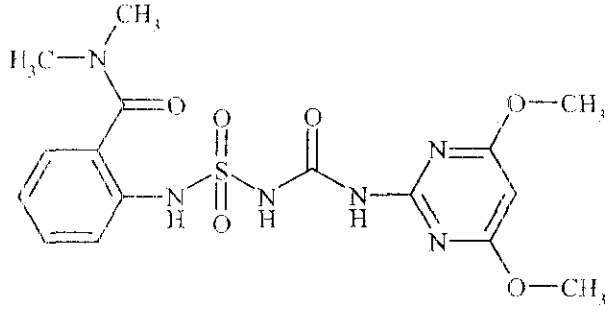
Compound	
Common name	Orthosulfamuron
Company experimental name	IR5878
IUPAC name	1-(4,6-dimethoxypyrimidin-2-yl)-3-[2-(dimethylcarbamoyl)phenylsulfamoyl]urea
CAS name	2-[[[[[(4,6-dimethoxy-2-pyrimidinyl)-amino]carbonyl]amino]sulfonyl]amino]-N,N-dimethylbenzamide
CAS registry number	213464-77-8
End-use product (EP)	0.51% G formulation (IR5878 0.5 GR; EPA Co. No. 80289) 51.5% WG formulation (IR5878 50 WG; EPA Co. No. 80289)

Table 2. Physicochemical Properties of the Technical Grade of Orthosulfamuron.		
Parameter	Value	Reference (MRID)
Color	White	46219004
Physical State	Fine Powder at 20°C	46219005
Odor	Odorless	46219006
pH	4.35 at 25°C (1% aqueous dispersion)	46219013
Density	1.45 g/mL at 20°C	46219008
Water solubility at 20°C	pH 4 buffer: 0.062 g/L pH 7 buffer: 0.63 g/L pH 8.5 buffer: 39 g/L	46219009
Solvent solubility at 20°C	n-heptane: 0.23 mg/L xylene: 130 mg/L acetone: 20 g/L ethyl acetate: 3.3 g/L dichloromethane: 56 g/L methanol: 8.3 g/L	Electronic communication, J. Messina to E. Kraft, 9/6/2006
Vapor pressure	1.1×10^{-4} at 20°C	46219010
Dissociation constant, pK_a	The test material becomes increasingly less soluble in water as the pH is lowered and undergoes degradation (hydrolysis) at neutral to acidic pHs. The test material is predicted to have 5 overlapping dissociation constants.	46219011
Octanol/water partition coefficient, $Log(K_{OW})$	pH 4: 2.0 pH 7: 1.3	46219012
UV/visible absorption spectrum	at pH 6.9, A=0.49 and $\varepsilon = 2.1 \times 10^4$ at 238 nm	46219001

B. EXPERIMENTAL DESIGN

Two field trials were conducted in the U.S. during the 2004 growing season in Region 10 (California; 2 trials).

Each field test consisted of one untreated plot and one treated plot. A single broadcast application of a 0.5% granular (G) formulation of orthosulfamuron was made to emerged rice (3 leaf stage) growing in flooded rice paddies. Application was made at 0.067 lb ai/A (75.0-75.1 g ai/ha) using ground equipment (shaker or spreader). Samples of rice grain and straw were harvested at maturity.

The test crops were grown and maintained according to typical practices for each region. Irrigation was applied in order to maintain the flood level typical for the test area. Irrigation was stopped during the last 3-9 weeks of the study to allow the crop to dry down. Fertilizer and maintenance pesticides were applied as needed. Trial site conditions are presented in Table B.1.1. The crop varieties grown are identified in Table C.3. Average minimum and maximum temperatures and total precipitation recordings for each month of the residue study period were reported, along with the average 10-year historical data. Weather conditions were similar to those recorded in historical weather data.

B.1. Study Site Information

TABLE B.1.1. Trial Site Conditions.

Trial Identification: City, State; Year (Trial ID #)	Soil characteristics			
	Type	%OM [1]	pH [1]	CEC [1] (meq/g)
Porterville, CA; 2004 (CA1)	Clay loam	NR	NR	NR
Live Oak, CA; 2004 (CA2)	Clay loam	NR	NR	NR

[1] NR = Not reported.

TABLE B.1.2. Study Use Pattern.

Location: City, State; Year (Trial ID)	EP[1]	Application					Tank Mix/ Adjuvants
		Method; Timing	Vol. (GPA[2]) [L/ha]	Rate (lb ai/A) [g ai/ha]	RTI[3] (days)	Total Rate (lb ai/A) [g ai/ha]	
Porterville, CA; 2004 (CA1)	0.5% G	1. Broadcast to flooded field; 3 leaf stage, crop height 8 inches	NA	0.067 [75.0]	NA	0.067 [75.0]	None
Live Oak, CA 2004 (CA2)	0.5% G	1. Broadcast to flooded field; 3 leaf stage, crop height 6 inches	NA	0.067 [75.1]	NA	0.067 [75.1]	None

[1] EP = End-use Product; IR5878 0.5GR
[2] GPA = Gallons per acre [L/ha = Liters per hectare]; Not applicable (NA), granular formulation.
[3] RTI = Retreatment Interval; NA, a single application was made.

TABLE B.1.3. Trial Numbers and Geographical Locations.

NAFTA Growing Zones	Rice		
	Submitted	Requested	
		Canada	U.S.
1			
1A			
2			
3			
4			7
5			1
5A			
5B			
6			2
7			
7A			
8			
9			
10	2		2
11			
12			
13			
14			
15			
16			
17			
18			
19			
20			
21			
Total	2 [1]		12 [2]

[1] Data from 11 additional field trials conducted with the WG formulation in Zones 4, 5, 6, and 10 have been submitted separately (refer to the DER for MRID 46578964).

[2] The number of requested field trials represents a 25 percent reduction in the number of trials due to the pesticidal use resulting in no quantifiable residues.

B.2. Sample Handling and Preparation

A single untreated and duplicate treated samples of rice grain and straw were collected from each trial site 116-136 days after application. All samples were frozen within 2 hours of sampling. Samples were shipped by freezer truck to PTRL West, Inc. (Hercules, CA), where samples were stored frozen (ca. -18°C) prior to analysis. Samples were homogenized in the presence of dry ice prior to residue analysis.

B.3. Analytical Methodology

Samples of rice grain and straw were analyzed for residues of orthosulfamuron at PTRL West, Inc. (Hercules, CA) using an LC/MS/MS method based on the "Enforcement Method (including Validation) for the Determination of Residues of IR 5878 in Rice Grain, Rice Green Plant and Rice Straw" as presented in Report ISA-0102V, Dr. Specht & Partner, 2002. A method description was included with the subject submission. The validated LOQ was 0.05 ppm in/on rice grain and straw. The limit of detection was 0.02 ppm (3 times the standard deviation of recoveries in matrix).

Briefly, homogenized samples of rice grain and straw were extracted twice with acetonitrile (ACN):0.02 M triethylamine (4:1, v:v) and filtered. Sodium chloride was added to the filtrate to induce separation of the aqueous and ACN phases. An aliquot of the ACN layer was partitioned with hexane. The ACN phase was evaporated to dryness and residues were redissolved in methanol and water for HPLC/MS/MS analysis. Residues were quantitated using external standards.

C. RESULTS AND DISCUSSION

Sample storage conditions and intervals are summarized in Table C.2. The maximum storage interval of crop samples from harvest to analysis was 78 days (2.6 months) for rice grain and 79 days (2.6 months) for rice straw. The results of a storage stability study were submitted (refer to the 860.1380 DER for MRID 46578983) which indicate that residues of orthosulfamuron are stable under frozen storage conditions in/on fortified samples of rice grain and straw for up to 12 months. These data are adequate to support the storage intervals of samples from the rice field trials.

Concurrent recovery data are presented in Table C.1. The LC/MS/MS method used is adequate for data collection based on acceptable concurrent recovery data. Concurrent recoveries ranged from 86% to 88% (mean = 87% with a standard deviation (s.d.) of 1%) for rice grain and 88-96% (mean = 93 with a s.d. of 4%) for rice straw each fortified with orthosulfamuron at 0.05 ppm. Sample chromatograms were supplied which indicated that the matrix was relatively free of interferences. Apparent residues of orthosulfamuron were nondetectable (<0.02 ppm) in/on two samples each of untreated rice grain and straw.

The results of the rice field trials are reported in Table C.3. A summary of the residue data is presented in Table C.4. Residues of orthosulfamuron were below the LOQ (<0.05 ppm) in/on all samples of rice grain and straw harvested at maturity following a single broadcast application of the 0.5% G formulation at 0.067 lb ai/A to rice in flooded rice paddies.

TABLE C.1. Summary of Concurrent Recoveries of Orthosulfamuron from Rice Grain and Straw.

Matrix	Spike level (ppm)	Sample size (n)	Recoveries (%)	Mean ± std dev (%)
Rice grain	0.05	3	86, 86, 88	87 ± 1.2
Rice straw	0.05	3	88, 94, 96	93 ± 4.2

TABLE C.2. Summary of Storage Conditions.

Matrix (RAC)	Storage Temperature (°C)	Actual Storage Duration [1]	Interval of Demonstrated Storage Stability [2]
Rice grain	<-18 [3]	44-78 days (1.4-2.6 months)	Stable under frozen storage conditions in/on fortified samples of rice grain and straw for up to 12 months.
Rice straw	<-18 [3]	45-79 days (1.5-2.6 months)	

[1] All samples were analyzed within 0-5 days of extraction.
[2] Refer to 860.1380 DER for MRID 46578983.
[3] The petitioner noted that the freezer temperature did not maintain -18°C throughout the period of sample storage. The freezer air temperature rose to a maximum of +8°C when freezer door was opened for a short period to remove samples. The samples remained frozen during the entire storage period.

TABLE C.3. Residue Data from Crop Field Trials with Orthosulfamuron.

Trial ID (City, State; Year)	Zone	Crop; Variety	Commodity or Matrix	Total Rate (lb ai/A) [g ai/ha]	PHI (days)	Orthosulfamuron Residues (ppm)
Porterville, CA; 2004 (CA1)	10	Rice; Koshihikari	Grain	0.067 [75.0]	136	ND, ND
			Straw		136	ND, ND
Live Oak, CA; 2004 (CA2)	10	Rice; M 204	Grain	0.067 [75.1]	116	ND, ND
			Straw		116	ND, ND

[1] Residues below the method LOD (<0.02 ppm) are reported as ND (nondetectable).

TABLE C.4. Summary of Residue Data from Crop Field Trials with Orthosulfamuron.

Commodity	Total Applic. Rate (lb ai/A) [g ai/ha]	PHI (days)	Residue Levels (ppm) [1]						
			n	Min.	Max.	HAFT [2]	Median (STMdR)	Mean (STMR)	Std. Dev.
Rice grain	0.067 [75.0-75.1]	116-136	4	<0.05	<0.05	<0.05	<0.025	<0.025	N/A
Rice straw		116-136	4	<0.05	<0.05	<0.05	<0.025	<0.025	N/A

[1] The method LOQ was <0.05 ppm. The median, mean, and standard deviation were calculated using half the LOQ (<0.025 ppm) for all residues reported as ND in Table C.3.
[2] HAFT = Highest Average Field Trial

D. CONCLUSION

The submitted field trial data reflect the use of a broadcast application of a 0.5% G formulation of orthosulfamuron at a rate of 0.067 lb ai/A (75.0-75.1 g ai/ha) to rice. Application was made to emerged rice (3 leaf stage) growing in flooded rice paddies. An acceptable method was used for quantitation of residues in/on rice grain and straw.

E. REFERENCES

None.

F. DOCUMENT TRACKING

Petition Number: 5F6957
DP Barcode: D319614
PC Code: 108209

Primary Evaluator	Douglas Dotson, Chemist, RAB2	Date: 2/14/2007
Peer Reviewer	Dennis McNeilly, Chemist, RAB2	Date: 2/14/2007

This DER was originally prepared under contract by Dynamac Corporation (2275 Research Boulevard, Suite 300; Rockville, MD 20850; submitted 06/12/2006). The DER has been reviewed by the Health Effects Division (HED) and revised to reflect current Office of Pesticide Programs (OPP) policies.

STUDY REPORT:

46578965 Willard, T. (2004) Magnitude of the Residue of IR5878 in Rice Processed Commodities: Final Study Report. Project Number: AA030702, A2_06.05/01, AR. Unpublished study prepared by American Agricultural Services and South Texas Ag Research, Inc. and PTRL West, Inc. 159 p.

EXECUTIVE SUMMARY:

Isagro S.p.A. submitted a processing study on rice. In two trials conducted in Arkansas and California, mature rice grain was harvested 103 or 105 days after a single broadcast application of the 50% water-dispersible granular (WG) formulation made at either 0.066-0.067 lb ai/A (73.9-74.9 g ai/ha; 1x the field trial application rate) or 0.200 lb ai/A (223.7-224.5 g ai/ha; ~3x the proposed field trial application rate) to moist/wet soil (not flooded) when rice was in the 2-3 leaf stage. The harvested rice grain samples were processed into polished rice, bran, and hulls using simulated commercial processing procedures.

Samples of rice grain and its processed commodities (polished rice, bran, and hulls) were analyzed for residues of orthosulfamuron using an LC/MS/MS method based on the "Enforcement Method (including Validation) for the Determination of Residues of IR 5878 in Rice Grain, Rice Green Plant and Rice Straw" as presented in Report ISA-0102V, Dr. Specht & Partner, 2002. This method is adequate for data collection based on acceptable method recoveries. The validated LOQ was 0.05 ppm and the limit of detection was 0.02 ppm.

The maximum storage interval of the study samples from collection/processing to analysis was 86 days (2.8 months) for rice grain and 49-56 days (1.6-1.8 months) for the processed rice commodities. To support sample storage conditions and intervals, the petitioner submitted the results of a storage stability study (refer to the 860.1380 DER for MRID 46578983) which indicate that residues of orthosulfamuron are stable under frozen storage conditions in/on fortified samples of rice grain and straw for up to 12 months. The available storage stability data support the storage intervals and conditions of the RAC (rice grain) but no storage stability data are available for the processed commodities of rice.

Residues of orthosulfamuron were less than the method LOD (<0.02 ppm) in/on rice grain from both treatment rates. Residues of orthosulfamuron were also less than the method LOD in all samples of polished rice, bran, and hulls processed from rice grain bearing nonquantifiable orthosulfamuron residues. Processing factors could not be calculated because residues were below the LOD in/on the RAC and the processed commodities.

The maximum theoretical concentration factor for rice is 8x (OPPTS GLN 860.1520, Table 1). According to Table 3 of OPPTS 860.1520, the theoretical concentration factors based on separation into components for rough rice grain commodities are 5.0x for hulls and 7.7x for bran.

STUDY/WAIVER ACCEPTABILITY/DEFICIENCIES/CLARIFICATIONS:

Under the conditions and parameters used in the study, the processed commodity residue data are classified as scientifically acceptable to support nonquantifiable residues in the rice grain RAC treated at 1x and 3x the nominal field rate. However, the data are not acceptable to support the nonquantifiable residues in the processed commodities of rice because supporting storage stability data are not available for rice processed commodities. Storage stability data, investigating the stability of orthosulfamuron residues in polished rice, hulls, and bran stored frozen for up to 2 months, are required.

The acceptability of this study for regulatory purposes is addressed in the U.S. EPA Residue Chemistry Summary Document, D332290, D. Dotson, 2/14/2007.

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

Orthosulfamuron is a postemergence herbicide that Isagro S.p.A. is proposing for use on rice grown in the United States for the control of annual and perennial broadleaf weeds, sedges, and barnyard grass. Orthosulfamuron belongs to the sulfamoylurea class of herbicides. It reportedly acts by inhibiting the plant enzyme acetolactate synthase which is active in the biosynthesis of valine, leucine, and isoleucine.

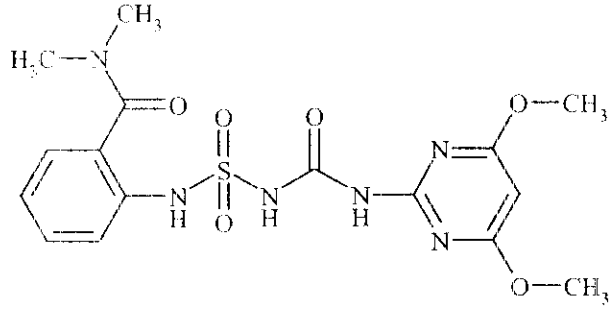
TABLE A.1. Test Compound Nomenclature.	
Compound	
Common name	Orthosulfamuron
Company experimental name	IR5878
IUPAC name	1-(4,6-dimethoxypyrimidin-2-yl)-3-[2-(dimethylcarbamoyl)phenylsulfamoyl]urea
CAS name	2-[[[[[(4,6-dimethoxy-2-pyrimidinyl)-amino]carbonyl]amino]sulfonyl]amino]-N,N-dimethylbenzamide
CAS registry number	213464-77-8
End-use product (EP)	0.51% G formulation (IR5878 0.5 GR; EPA Co. No. 80289) 51.5% WG formulation (IR5878 50 WG; EPA Co. No. 80289)

Table 2. Physicochemical Properties of the Technical Grade of Orthosulfamuron.		
Parameter	Value	Reference (MRID)
Color	White	46219004
Physical State	Fine Powder at 20°C	46219005
Odor	Odorless	46219006
pH	4.35 at 25°C (1% aqueous dispersion)	46219013
Density	1.45 g/mL at 20°C	46219008
Water solubility at 20°C	pH 4 buffer: 0.062 g/L pH 7 buffer: 0.63 g/L pH 8.5 buffer: 39 g/L	46219009
Solvent solubility at 20°C	n-heptane: 0.23 mg/L xylene: 130 mg/L acetone: 20 g/L ethyl acetate: 3.3 g/L dichloromethane: 56 g/L methanol: 8.3 g/L	Electronic communication, J. Messina to E. Kraft, 9/6/2006
Vapor pressure	1.1×10^{-4} at 20°C	46219010
Dissociation constant, pK_a	The test material becomes increasingly less soluble in water as the pH is lowered and undergoes degradation (hydrolysis) at neutral to acidic pHs. The test material is predicted to have 5 overlapping dissociation constants.	46219011
Octanol/water partition coefficient. Log(K_{OW})	pH 4: 2.0 pH 7: 1.3	46219012
UV/visible absorption spectrum	at pH 6.9, A=0.49 and $\varepsilon = 2.1 \times 10^4$ at 238 nm	46219001

B. EXPERIMENTAL DESIGN

Two rice processing studies were conducted in 2003 in AR and CA. Each site included an untreated plot, a plot treated at the nominal field rate, and a plot treated at an exaggerated rate. The plots were 2,400-4,000 sq. ft. and separated by a levee. Rice plants in nonflooded fields were treated early post-emergence (2-3 leaf growth stage) with the 50% WG formulation of orthosulfamuron at ~0.067 lb ai/A (1x the field trial application rate) or ~0.200 lb ai/A (3x the field trial application rate). The broadcast spray application was made using ground equipment in ~15 gal/A, with 0.2% (v:v) of surfactant added to the spray mixture.

Permanent flooding was established 3-7 days after treatment and irrigation was applied in order to maintain the flood level typical for the test area. Irrigation was stopped during the last 2-4 weeks of the study to allow the crop to dry down. The test crops were grown and maintained according to typical practices for each region. Rice grain was harvested at maturity and processed using simulated commercial processing procedures into polished rice, hulls, and bran.

B.1. Application and Crop Information

TABLE B.1.1. Study Use Pattern.

Location (City, State; Year)	EP [1]	Application					Tank Mix/ Adjuvants
		Method; Timing	Vol. (GPA[2]) [L/ha]	Rate (lb ai/A)	RTI [3] (days)	Total Rate (lb ai/A)	
Proctor, AR; 2003	50% WG	1. Broadcast to unflooded fields; 2-3 leaf stage, crop height 3 inches	15.3 [143.1]	0.067 [74.9]	NA	0.067 [74.9]	ADJ 1012 (0.2% v/v)
	50% WG	1. Broadcast to unflooded fields; 2-3 leaf stage, crop height 3 inches	15.4 [144.0]	0.200 [224.5]	NA	0.200 [224.5]	ADJ 1012 (0.2% v/v)
Porterville, CA; 2003	50% WG	1. Broadcast to unflooded fields; 2-3 leaf stage, crop height 4 inches	14.8 [138.4]	0.066 [73.9]	NA	0.066 [73.9]	ADJ 1012 (0.2% v/v)
	50% WG	1. Broadcast to unflooded fields; 2-3 leaf stage, crop height 5 inches	14.9 [139.3]	0.200 [223.7]	NA	0.200 [223.7]	ADJ 1012 (0.2% v/v)

[1] EP = End-use Product; IR5878 50 WG
[2] GPA = Gallons per acre [L/ha = Liters per hectare]; application volume calculated by the petitioner (L/ha = GPA x 9.35)
[3] RTI = Retreatment Interval; Not applicable (NA) because a single application was made.

B.2. Sample Handling and Processing Procedures

A single bulk sample of untreated and treated rice grain was collected from each plot (untreated, 1x treatment rate regime, and 3x treatment rate regime) for processing into hulls, bran, and polished rice using simulated commercial practices. Mature rough rice grain samples were shipped at ambient temperatures to South Texas Ag Research, Inc. (Brookshire, TX) via overnight delivery either on the day of harvest (CA trial) or the day after harvest (AR trial). Upon receipt at the processing facility samples were stored frozen (<-10°C). Processing was conducted within 51-52 days of sample collection for the AR trial and 34-36 days of sample collection for the CA trial.

Prior to processing, a whole grain RAC sample was collected from each bulk sample and frozen. The remaining bulk sample was placed in a dryer for 3.2-26.8 hours to thaw and reduce the moisture content to <10.5%. The dried rough rice was cleaned and hulled. The hulled rice was milled to remove the bran and leave the polished rice. The petitioner submitted adequate descriptions of the processing procedures including material balance summaries. Immediately after processing, all samples were frozen until shipment by ACDS freezer truck to PTRL West, Inc. (Hercules, CA) for residue analysis. Samples of rice grain and polished rice were homogenized in the presence of dry ice prior to residue analysis. The hull and bran samples required no processing prior to extraction. Samples were stored frozen (ca. -18°C) until analysis.

The rice processing procedures are summarized in the flow chart below (Figure 1), which was copied without alteration from MRID 46578965.

FILE R143130

DER for MRID #s 46578669-69, -60, -62, -63, -64, -61, 88-65, 65

Page 121 is not included in this copy.

Pages _____ through _____ are not included in this copy.

The material not included contains the following type of information:

_____ Identity of product inert ingredients.

_____ Identity of product impurities.

_____ Description of the product manufacturing process.

_____ Description of quality control procedures.

_____ Identity of the source of product ingredients.

_____ Sales or other commercial/financial information.

_____ A draft product label.

_____ The product confidential statement of formula.

_____ Information about a pending registration action.

✓ FIFRA registration data.

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

_____ The document is not responsive to the request.

_____ Internal deliberative information.

_____ Attorney-Client work product.

_____ Claimed Confidential by submitter upon submission to the Agency.

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

B.3. Analytical Methodology

Samples of rice grain and rice processed commodities (polished rice, bran, and hulls) were analyzed for residues of orthosulfamuron at PTRL West, Inc. (Hercules, CA) using an LC/MS/MS method based on the "Enforcement Method (including Validation) for the Determination of Residues of IR 5878 in Rice Grain, Rice Green Plant and Rice Straw" as presented in Report ISA-0102V, Dr. Specht & Partner, 2002. A method description was included with the subject submission. The validated LOQ was 0.05 ppm and the limit of detection was 0.02 ppm (3 times the standard deviation of recoveries in matrix).

Briefly, homogenized samples (rice grain and polished rice) and nonhomogenized samples (rice bran and hulls) were extracted twice with acetonitrile (ACN):0.02 M triethylamine (4:1, v:v) and filtered. Sodium chloride was added to the filtrate to induce separation of the aqueous and ACN phases. An aliquot of the ACN layer was partitioned with hexane. The ACN phase was evaporated to dryness, and residues were redissolved in methanol and water for HPLC/MS/MS analysis. Residues were quantitated using external standards.

C. RESULTS AND DISCUSSION

Mature rice grain was harvested 103 or 105 days after a single broadcast foliar application of the 50% WG formulation made at either 0.066-0.067 lb ai/A (1x the field trial application rate) or 0.200 lb ai/A (~3x the field trial application rate) to moist/wet soil (not flooded) when rice was in the 2-3 leaf stage. The rice grain samples were processed into polished rice, bran, and hulls using simulated commercial processing procedures.

Sample storage intervals and conditions are summarized in Table C.2. Rice grain and processed rice commodities (polished rice, bran, and hulls) were stored frozen following harvest/processing until analysis. The maximum storage interval of the study samples from collection/processing to analysis was 86 days (2.8 months) for rice grain and 49-56 days (1.6-1.8 months) for the processed rice commodities. To support sample storage conditions and intervals, the petitioner submitted the results of a storage stability study (refer to the DER for MRID 46578983) which indicate that residues of orthosulfamuron are stable under frozen storage conditions in/on fortified samples of rice grain and straw for up to 12 months. The available storage stability data support the storage intervals and conditions of the RAC (rice grain) but no storage stability data are available for the processed commodities of rice.

Concurrent recovery data from the rice processing study are presented in Table C.1. The LC/MS/MS method used is adequate for data collection based on acceptable concurrent recovery data. Concurrent recoveries ranged from 94% to 102% (mean = 98% with a standard deviation of 3%) for rice grain and its processed commodities each fortified with orthosulfamuron at 0.05 ppm. Sample chromatograms were supplied which indicated that the matrix was relatively free of interferences. Apparent residues of orthosulfamuron were nondetectable (<0.02 ppm) in/on two samples each of untreated rice grain and its processed commodities polished rice, bran, and hulls.

Residues of orthosulfamuron were less than the method LOD (<0.02 ppm) in/on rice grain harvested 103 or 105 days following a single broadcast foliar application of the 50% WG formulation made at either 0.066-0.067 lb ai/A or 0.200 lb ai/A. Residues of orthosulfamuron were also less than the method LOD in polished rice, bran, and hulls processed from rice grain bearing nonquantifiable orthosulfamuron residues. Processing factors could not be calculated because residues were below the LOD in/on the RAC and the processed commodities.

TABLE C.1. Summary of Concurrent Recoveries of Orthosulfamuron from Rice Matrices.

Matrix	Spike level (ppm)	Sample size (n)	Recoveries (%)	Mean ± std dev (%)
Rice grain	0.05	2	100, 100	98 ± 3
Polished rice	0.05	2	94, 94	
Bran	0.05	2	98, 100	
Hulls	0.05	2	98, 102	

TABLE C.2. Summary of Storage Conditions.

Matrix	Storage Temperature (°C)	Actual Storage Duration [1]	Interval of Demonstrated Storage Stability
Rice grain (RAC)	<-18	84-86 days (2.8 months)	Stable under frozen storage conditions in/on fortified samples of rice grain and straw for up to 12 months. [2]
Polished rice	<-18	35-49 days (1.2-1.6 months)	None available for the processed commodities of rice.
Bran	<-18	42-56 days (1.4-1.8 months)	
Hulls	<-18	36-50 days (1.2-1.6 months)	

[1] Actual storage duration from harvest to analysis for RAC and processing to analysis for processed commodities; samples were processed within 34-52 days of harvest. All samples were analyzed on the day of extraction.
[2] Refer to 860.1380 DER for MRID 46578983.

TABLE C.3. Residue Data from Rice Processing Study with Orthosulfamuron.

RAC (Study ID)	Processed Commodity	Total Rate (lb ai/A)	PHI (days)	Orthosulfamuron Residues (ppm)	Processing Factor
Rice (AR)	Grain (RAC)	0.067 [74.9]	103	ND [1]	--
	Polished rice		103	ND	NC [2]
	Hulls		103	ND	NC
	Bran		103	ND	NC
	Grain (RAC)	0.200 [224.5]	103	ND	--
	Polished rice		103	ND	NC
	Hulls		103	ND	NC
	Bran		103	ND	NC

TABLE C.3. Residue Data from Rice Processing Study with Orthosulfamuron.

RAC (Study ID)	Processed Commodity	Total Rate (lb ai/A)	PHI (days)	Orthosulfamuron Residues (ppm)	Processing Factor
Rice (CA)	Grain (RAC)	0.066 [73.9]	105	ND	--
	Polished rice		105	ND	NC
	Hulls		105	ND	NC
	Bran		105	ND	NC
	Grain (RAC)	0.200 [223.7]	105	ND	--
	Polished rice		105	ND	NC
	Hulls		105	ND	NC
	Bran		105	ND	NC

[1] Residues below the method LOD (<0.02 ppm) are reported as ND (nondetectable).
[2] NC = Not calculated. The processing factor could not be calculated because residues were below the LOD in both the RAC and the processed sample.

D. CONCLUSION

The submitted field trial data reflect the use of a broadcast foliar application of a 50% WG formulation of orthosulfamuron at a rate of 0.066-0.067 lb ai/A (73.9-74.9 g ai/ha) or 0.200 lb ai/A (223.7-224.5 g ai/ha) to rice. Application was made to moist/wet soil (not flooded) when rice was in the 2-3 leaf stage. Processing factors for orthosulfamuron in polished rice, bran, and hulls could not be calculated because residues were nondetectable in both the RAC (rice grain) and all rice processed commodities. An acceptable method was used for quantitation of residues in/on rice grain and its processed commodities.

E. REFERENCES

None.

F. DOCUMENT TRACKING

Petition Number: 5F6957
DP Barcode: D319614
PC Code: 108209

Primary Evaluator Douglas Dotson, Chemist, RAB2 D. Dotson Date: 2/14/2007

Peer Reviewer Dennis McNeilly, Chemist, RAB2 Date: 2/14/2007

This DER was originally prepared under contract by Dynamac Corporation (2275 Research Boulevard, Suite 300; Rockville, MD 20850; submitted 06/12/2006). The DER has been reviewed by the Health Effects Division (HED) and revised to reflect current Office of Pesticide Programs (OPP) policies.

STUDY REPORTS:

46578982 Zini, G.; Crisippi, T. (2004) Storage Stability of IR5878 in Rice Green Plants Stored in the Dark Below -20°C: (Final Report). Project Number: 2376, A2_06.00/02. Unpublished study prepared by Isagro S.R.L. (Formerly Agrimont). 98 p.

46578983 Rose, J. (2004) Storage Stability of IR5878 in Rice Grain and Straw Stored in the Dark Below -20°C: (Final Report). Project Number: 1176W, A2_06.00/01. Unpublished study prepared by PTRL West, Inc. 68 p.

EXECUTIVE SUMMARY:

Isagro S.p.A. has submitted storage stability studies with orthosulfamuron in rice. Untreated samples of rice green plant, grain, and straw from the orthosulfamuron rice crop field trials were fortified with orthosulfamuron standard at 0.5 ppm. Samples were stored at -20°C in the dark and analyzed at storage intervals of approximately 0, 1, 3, 6, and 12 months. The study on rice green plants (MRID 46578982) was conducted by Isagro Ricerca S.r.l. (Novara, Italy), and the study on rice grain and straw (MRID 46578983) was conducted by PTRL West, Inc. (Hercules, CA). The results indicate that residues of orthosulfamuron are stable at -20°C for up to 368 days (12 months) in/on rice grain and straw, and for up to 408 days (13 months) in/on rice green plants.

Rice samples were analyzed for residues of orthosulfamuron using an LC/MS/MS method which is the same method that was used for data collection for the magnitude of the residue trials. Based on the concurrent method recovery data, the LC/MS/MS analytical method is adequate for the determination of residues of orthosulfamuron in rice matrices. The validated LOQ was 0.05 ppm in/on rice grain, straw, and green plant.

STUDY/WAIVER ACCEPTABILITY/DEFICIENCIES/CLARIFICATIONS:

Under the conditions and parameters used in the study, the storage stability data are classified as scientifically acceptable. The acceptability of this study for regulatory purposes is addressed in the U.S. EPA Residue Chemistry Summary Document, D332290, D. Dotson, 2/14/2007.

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; however we note that the rice green plant study (conducted in Italy) was not performed in accordance with US EPA GLPs, but was conducted in compliance with OECD principles of GLP.

A. BACKGROUND INFORMATION

Orthosulfamuron is a postemergence herbicide that Isagro S.p.A. is proposing for use on rice grown in the United States for the control of annual and perennial broadleaf weeds, sedges, and barnyard grass. Orthosulfamuron belongs to the sulfamoylurea class of herbicides. It reportedly acts by inhibiting the plant enzyme acetolactate synthase which is active in the biosynthesis of valine, leucine, and isoleucine.

TABLE A.1. Test Compound Nomenclature.

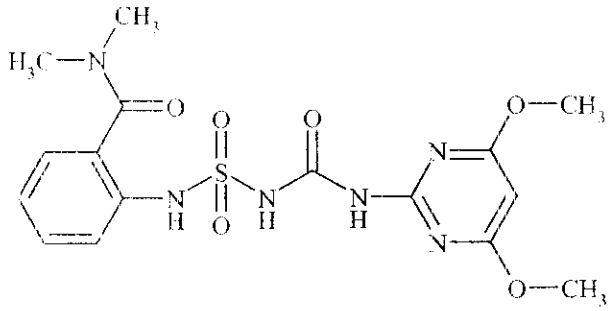
Compound	
Common name	Orthosulfamuron
Company experimental name	IR5878
IUPAC name	1-(4,6-dimethoxypyrimidin-2-yl)-3-[2-(dimethylcarbamoyl)phenylsulfamoyl]urea
CAS name	2-[[[[[(4,6-dimethoxy-2-pyrimidinyl)-amino]carbonyl]amino]sulfonyl]amino]-N,N-dimethylbenzamide
CAS registry number	213464-77-8
End-use product (EP)	0.51% G formulation (IR5878 0.5 GR; EPA Co. No. 80289) 51.5% WG formulation (IR5878 50 WG; EPA Co. No. 80289)

Table 2. Physicochemical Properties of the Technical Grade of Orthosulfamuron.		
Parameter	Value	Reference (MRID)
Color	White	46219004
Physical State	Fine Powder at 20°C	46219005
Odor	Odorless	46219006
pH	4.35 at 25°C (1% aqueous dispersion)	46219013
Density	1.45 g/mL at 20°C	46219008
Water solubility at 20°C	pH 4 buffer: 0.062 g/L pH 7 buffer: 0.63 g/L pH 8.5 buffer: 39 g/L	46219009
Solvent solubility at 20°C	n-heptane: 0.23 mg/L xylene: 130 mg/L acetone: 20 g/L ethyl acetate: 3.3 g/L dichloromethane: 56 g/L methanol: 8.3 g/L	Electronic communication, J. Messina to E. Kraft, 9/6/2006
Vapor pressure	1.1×10^{-4} at 20°C	46219010
Dissociation constant, pK_a	The test material becomes increasingly less soluble in water as the pH is lowered and undergoes degradation (hydrolysis) at neutral to acidic pHs. The test material is predicted to have 5 overlapping dissociation constants.	46219011
Octanol/water partition coefficient, $Log(K_{OW})$	pH 4: 2.0 pH 7: 1.3	46219012
UV/visible absorption spectrum	at pH 6.9, A= 0.49 and $\varepsilon = 2.1 \times 10^4$ at 238 nm	46219001

B. EXPERIMENTAL DESIGN

B.1. Sample Handling and Preparation

Untreated samples of rice green plant, grain, and straw, from the orthosulfamuron crop field trials, were homogenized with dry ice and fortified with orthosulfamuron standard at 0.5 ppm in methanol. Samples were stored at -20°C in the dark and analyzed at storage intervals of approximately 0, 1, 3, 6, and 12 months. Freshly fortified samples were also analyzed at each storage interval.

B.2. Analytical Methodology

Samples of rice green plants, grain and straw were analyzed for residues of orthosulfamuron using an LC/MS/MS method entitled "Enforcement Method (including Validation) for the Determination of Residues of IR5878 in Rice Grain, Rice Green Plant and Rice Straw" as presented in Report ISA-0102V, Dr. Specht & Partner, 2002. A complete method description was included with the rice grain and straw submission; minor method differences were used for the rice green plant study (refer to the DER for MRID 46578960 for method details). The

validated LOQ was 0.05 ppm in/on rice green plants, grain, and straw. The limit of detection was 0.02 ppm (3 times the standard deviation of recoveries in matrix) for rice grain and straw and 0.03 ppm for green plant.

Briefly, homogenized samples of rice grain and straw were extracted twice with acetonitrile (ACN):0.02 M triethylamine (4:1, v:v) and filtered. Sodium chloride was added to the filtrate to induce separation of the aqueous and ACN phases. An aliquot of the ACN layer was then partitioned with hexane. The resulting ACN phase was collected and evaporated to dryness by rotary evaporation. Residues were redissolved in methanol, and water was added. The final sample solution was microfilterfuged to remove any particulate matter and analyzed by HPLC/MS/MS. Because of poor recoveries from straw at the 0- and 1-month intervals, the method was modified for samples from later storage intervals to include soaking the straw samples in extraction solvent overnight.

For homogenized green plants, the method was the same as described for grain and straw except hexane pre-saturated with ACN was used for partitioning, and the dried ACN residues were dissolved in dichloromethane, filtered, dried again, and finally dissolved in ACN for analysis.

C. RESULTS AND DISCUSSION

Based on the concurrent method recovery data (see Table C.1), the LC/MS/MS analytical method is adequate for the determination of residues of orthosulfamuron in rice matrices. Because of poor recoveries from straw at the 0- and 1-month intervals, the method was modified to include soaking the straw samples in extraction solvent overnight. This modification resulted in improved recoveries of fortified samples analyzed at later intervals. Concurrent recoveries ranged from 70% to 122% from rice green plant, 73-99% from rice grain, and 59-101% from rice straw samples fortified with orthosulfamuron standard at 0.5 ppm. Apparent residues were below the LOQ in/on all unfortified samples.

The results of the storage stability study are presented in Table C.2. Based on the reported data, residues of orthosulfamuron are stable at -20°C for up to 368 days (12 months) in/on rice grain and straw, and 408 days (13 months) in/on rice green plants. A graph of the storage stability of residues of orthosulfamuron in/on rice matrices over time is presented in Figure C.1.

TABLE C.1. Summary of Concurrent Recoveries of Orthosulfamuron from Rice Matrices.

Matrix	Spike Level (ppm)	Storage Interval (days)	Sample Size (n)	Recoveries (%)	Mean ± SD [1] (%)
Rice green plants	0.5	30	2	70.36, 73.66	72.0
		100	2	95.07, 120.02	108
		170	2	118.07, 122.11	120
		408	2	86.57, 98.24	92.4
Rice grain	0.5	32	2	78, 98	88
		~3 months	2	97, 99	98
		203	2	73, 79	76
		368	2	86, 89	88
Rice straw	0.5	37	2	59, 70	65

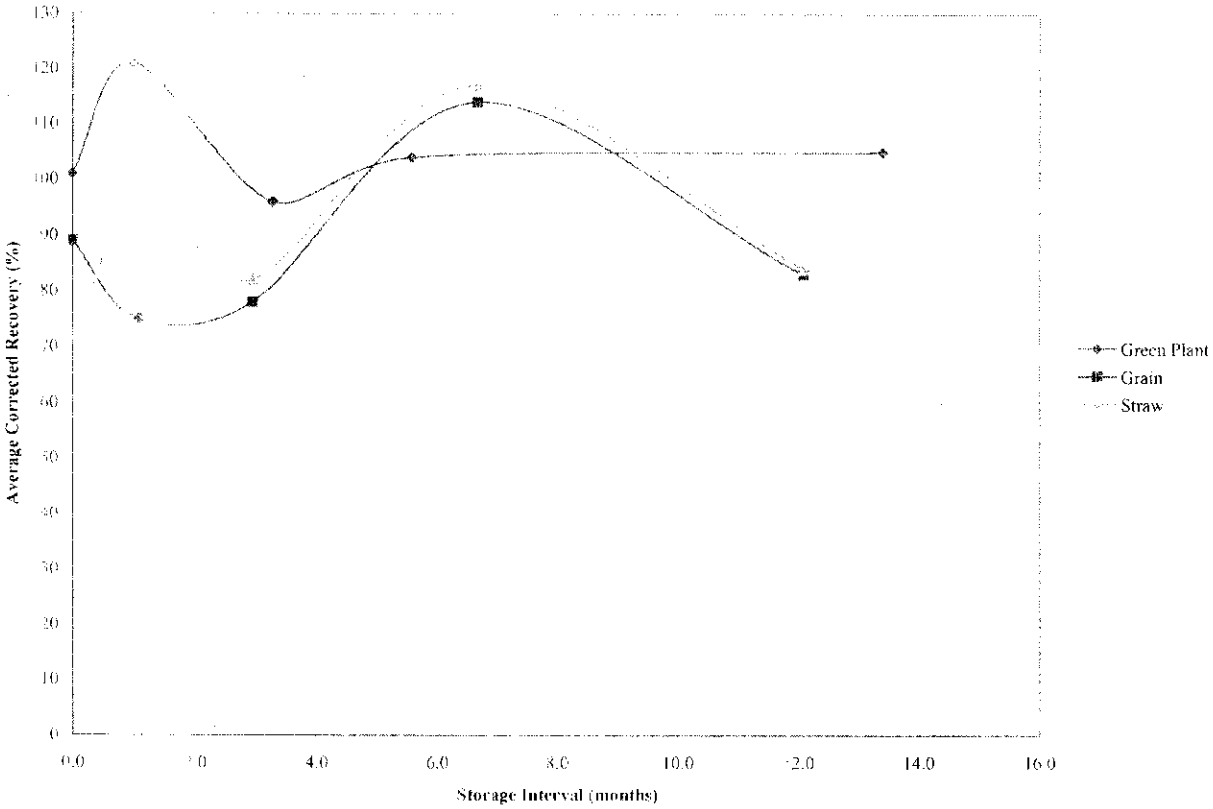
TABLE C.1. Summary of Concurrent Recoveries of Orthosulfamuron from Rice Matrices.					
Matrix	Spike Level (ppm)	Storage Interval (days)	Sample Size (n)	Recoveries (%)	Mean ± SD [1] (%)
		~3 months	2	95, 101	98
		203	2	93, 93	93
		368	2	79, 91	85

[1] Because only two values were available for each determination, standard deviations were not calculated.

TABLE C.2. Stability of Orthosulfamuron Residues in Rice Matrices Following Storage at -20°C.						
Commodity	Spike Level (ppm)	Storage Interval (days)	Recovered Residues (ppm)	Mean Recovered Residues (ppm)	Mean Recovery (%)	Corrected % Recovery [1]
Rice green plants	0.515	0	0.5192, 0.5248	0.5220	101.36	--
		30	0.4452, 0.4507	0.4479	86.98	121
		100	0.5296, 0.5361	0.5329	103.47	96
		170	0.6157, 0.6670	0.6413	124.53	104
		408	0.4728, 0.5232	0.4980	96.70	105
Rice grain	0.5	0	0.426, 0.459	0.443	89	--
		32	0.301, 0.362	0.332	66	75
		~3 months	0.378, 0.382	0.380	76	78
		203	0.428, 0.443	0.436	87	114
		368	0.362, 0.368	0.365	73	83
Rice straw	0.5	0	0.337, 0.378	0.358	72	--
		37	0.310, 0.331	0.321	64	99
		~3 months	0.385, 0.414	0.400	80	82
		203	0.538, 0.553	0.546	109	117
		368	0.349, 0.357	0.353	71	84

[1] Corrected for mean concurrent recovery (see TABLE C.1.); 0-time recoveries were not corrected as they represent fresh fortifications.

FIGURE C.1. Graph of Orthosulfamuron Stability in Rice Green Plants, Grain, and Straw.



D. CONCLUSION

The submitted storage stability results adequately demonstrate the stability of orthosulfamuron in/on rice green plants, grain, and straw stored frozen for up to 12-13 months. An acceptable method was used for the quantitation of residues in rice matrices.

E. REFERENCES

None.

F. DOCUMENT TRACKING

Petition Number: 5F6957
DP Barcode: D319614
PC Code: 108209

13544

R143130

Chemical: Orthosulfamuron

PC Code:
108209

HED File Code: 11000 Chemistry Reviews
Memo Date: 2/14/2007
File ID: 00000000
Accession #: 000-00-0119

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