

US EPA ARCHIVE DOCUMENT

5



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

OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

November 8, 2004

MEMORANDUM

SUBJECT: Thidiazuron Residue Chemistry Studies (4)

DP Barcode No.: D294543
Reregistration Case No.: 4092
PC Code: 120301
MRID No.: 46298401, 46298402, 46298403, 46298404

FROM: Toiya Jimerson, Chemist
Reregistration Branch 1
Health Effects Division (7509C)

THRU: Whang Phang, Ph.D., Branch Senior Scientist
Reregistration Branch 1
Health Effects Division (7509C)

TO: Stephanie Plummer, Chemical Review Manager
Special Review and Reregistration Division (7508C)

Attached are four residue chemistry reviews of the eleven studies submitted in support of the reregistration of thidiazuron. The MRID number and the citation of each study are listed below:

- 46298401 Magnitude of Residue in Rotational Crops Following Cotton Treated with DROPP® SC
- 46298402 Independent Laboratory Validation of Aventis Method AW/01/01 Thidiazuron: Analytical Method for the Determination of Thidiazuron and its Metabolite Photo-Thidiazuron in Crop Matrices Using LC/MS/MS

46298403 Independent Laboratory Validation of Aventis Method AW/02/01 Thidiazuron:
Analytical Method for the Determination of 1,2,3-thidiazol-5-ylurea
(AE F132345) Residues in Crop Matrices Using LC/MS/MS

46298404 Stability of Thidiazuron and its Metabolites 4-Hydroxy Thidiazuron and Phenyl
Urea in Milk and Beef Tissues During Frozen Storage

7509C:RRB1:TJimerson:CM#2:Rm 722H:308-0070:11/8/04
WPhang: 11/8/04



Primary Evaluator Toiya Jimerson, Chemist, Date: 11/8/04
Reregistration Branch 1,
Health Effects Division

Reviewer Felecia Fort, Chemist,
Reregistration Branch 1,
Health Effects Division

Whang Phang, Ph.D.,
Branch Senior Scientist
Reregistration Branch 1
Health Effects Division

STUDY REPORTS:

MRID No. 46298401 Frank A. Norris (March 28, 2002) Magnitude of Residue in Rotational Crops Following Cotton Treated with DROPP® SC. Lab Project Number: B003793. Unpublished study prepared by Frank A. Norris. 248 pages.

EXECUTIVE SUMMARY:

Thidiazuron (41.9%a.i., soluble concentrate) was applied to Norfolk Sandy Loam, Amarillo Sandy Loam, and Hanford Fine Sandy Loam soils at the maximum application rate (1x) of 0.3 lb a.i./A/season (0.337 kg a.i./ha/season) with a pre-harvest interval (PHI) of 5 days. Application sites were located in EPA Region IV, Pikeville, North Carolina and Leland, Mississippi (discontinued); Region VI, Levelland, TX; and Region IX, Fresno, California. Each test site consisted of one untreated control plot and one treated plot. Three trials were performed for each crop group at each application site. Leafy vegetable, root, and small grain crops were planted at a number of plant back intervals (PBI). Wheat had the shortest PBI at 2 weeks after the last thidiazuron application. Leafy vegetable, root, and other small grain crops were planted back at intervals of 1 month, 4 months, and 8 months.

Residue of thidiazuron and its metabolites were extracted using two methods. Aventis CropScience method AW/01/01 was used to quantify residues of thidiazuron and the crop metabolite photo-thidiazuron; method AW/02/01 was used to extract the soil metabolite 1,2,3-thidiazol-5-ylurea. Independent Laboratory Validations (ILV's) for both methods have been reviewed and accepted by RRB1/HED (MRIDs 46298402, 46298403).

Method verification and procedural recoveries were included in this study, both yielding acceptable results. Average method verification recoveries were 93%, 85%, and 78% for thidiazuron, photo-thidiazuron, and thidiazol urea, respectively. Average procedural recoveries for controls fortified at 0.05ppm (LOQ) were 89%, 88%, and 81% for thidiazuron, photo-



thidiazuron, and thiadiazol urea, respectively. No thidiazuron residues greater than LOQ (>0.05 ppm) were found in any of the treated rotational crops. Therefore, samples were not analyzed after the shortest PBI, which is 2-weeks for wheat and 1-month for root, leafy vegetable, and other small grain crops. Samples were not analyzed because it was expected that the recovered residues would be even lower in later plant back intervals. No new tolerances are needed.

Although the submitted field trial residue data was adequate, this study is unacceptable due to the inadequacy of the confined accumulation in rotational crops study (DP Barcode D241202, 10/26/04, T. Jimerson). An unknown metabolite was not characterized during the confined study, violating OPPTS Guideline No. 860.1300.

STUDY/WAIVER ACCEPTABILITY/DEFICIENCIES/CLARIFICATIONS:

Under the conditions and parameters used in the study, the field trial residue data are classified as scientifically unacceptable due to the inadequacy of the submitted confined accumulation in rotational crops study.

Additionally, there were several protocol amendments made to this study. Specifically, amendments number one and two had an impact on the root and leafy vegetable crop data in California and Texas. However, these deviations from protocol were not significant enough to impact the validity of the residue data submitted.

1. Radishes were planted 14 days late in one plot for the four month plant-back in Fresno, California.
2. Spinach planted at the 8 month plant back did not survive in the heat in Levelland, Texas.
3. Analysis discontinued for all but the crop matrices from the earliest plant-back interval.
4. Leland, Mississippi trial was terminated due to the pending sale of the Mississippi research station.
5. North Carolina research station added to study as a replacement for the Mississippi station.
6. Lot numbers were identified for test substances used in the study.
7. Errors in protocol sample numbers were corrected.
8. Replacement principal field investigator was named for Fresno, California trial.
9. Principal investigator named for this study, and method of analysis determined.

The acceptability of this study for regulatory purposes will be addressed in the forthcoming U.S. EPA Residue Chemistry Summary Document.

COMPLIANCE:

Signed and dated GLP, Quality Assurance and Data Confidentiality statements were provided. Reported deviations from regulatory requirements are relatively minor and include the



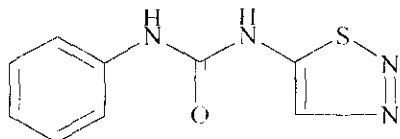
following:

1. The weather data reported by the National Oceanic and Atmospheric Administration (NOAA) or state weather services were collected by groups not under a GLP program. [40 CFR 160.185(a)(2)]
2. Some field and laboratory notebook entries and raw data were not properly entered according to GLP's, but were corrected as discovered during the review of the data according to GLP's. [40 CFR 160.130(e)]
3. The soil characterization laboratories did not have a copy of the study protocol. The laboratory was informed that characterization was part of a GLP study and the analyses were performed under their standard operating procedures. [40 CFR 160.130(a)]
4. The following exceptions were noted at one or more of the field or analytical facilities:
 - a. Inadequate training files. [40 CFR 160.29(b)]
 - b. Some equipment maintenance/calibration logs and SOP's did not contain all the required elements. [40 CFR 160.63(b)(c)]

A. BACKGROUND INFORMATION

Thidiazuron has one registered use as a cotton defoliant. Currently, the combined residues of thidiazuron and its aniline containing metabolites are regulated (40 CFR 180.403). The Metabolism Assessment Review Committee (9/29/95, F. Fort) has determined that the residue of concern in plant commodities consists of the parent thidiazuron only, and that the residues of concern in animal commodities consist of the parent thidiazuron, 4-hydroxythidiazuron, and phenyl urea. Tolerances are established in/on cottonseed (0.4 ppm), cottonseed hulls (0.8 ppm), eggs (0.1 ppm), and milk (0.05 ppm). Additional tolerances are set at 0.2 ppm for the fat, meat and meat by-products of cattle, goats, hogs, horses, poultry and sheep.

The confined accumulation in rotational crops study submitted was unacceptable because an unknown metabolite, accounting for 0.046 ppm (26.4 % TRR) in 14-DAT wheat straw and 0.010 ppm (8.8% TRR) in 120-DAT wheat straw, was not characterized during the confined study.

TABLE A.1. Thidiazuron- Test Compound Nomenclature	
Compound	Chemical Structure 
Common name	Thidiazuron



IUPAC name	1-phenyl-3-(1,2,3-thiadiazol-5-yl)urea
CAS name	N-phenyl-N'-1,2,3-thiadiazol-5-ylurea
CAS #	51707-55-2
End-use product/EP	DROPP® SC

TABLE A.2. Photo-thidiazuron- Test Compound Nomenclature

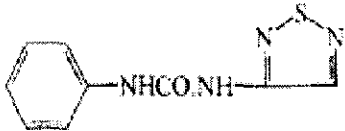
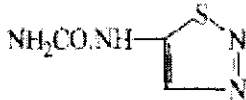
Compound	Chemical Structure 
Common name	Photo-thidiazuron
IUPAC name	1-phenyl-3-(1,2,5-thiadiazol-3-yl)urea
CAS name	N-phenyl-N'-1,2,5-thiadiazol-3-ylurea
CAS #	71769-74-9

TABLE A.3. Thiadiazol urea- Test Compound Nomenclature for Thiadiazol urea

Compound	Chemical Structure 
Common name	Thiadiazol urea
IUPAC name	1,2,3-thiadiazol-5-ylurea
CAS name	1,2,3-thiadiazol-5-ylurea
CAS #	89270-19-9

B. EXPERIMENTAL DESIGN

B.1. Study Site Information



TABLE B.1.1 Soil Characterization.

Study Location (City, State/Year)	Soil characteristics			
	Type	%OM	pH (in 1:1 soil:water ratio)	CEC (meq/100g)
Pikeville, NC/2000	Norfolk Sandy Loam	1.6	5.4	4.7
Levelland, TX/2000	Amarillo Sandy Loam	0.9	8.0	18.5
Fresno, CA/2000	Hanford Fine Sandy Loam	0.5	6.4	5.6

The actual temperature and rainfall average recordings were within average historical values for the residue study period. Irrigation was used to supplement as needed for each site. No meteorological abnormalities were reported during the conduct of the study. Recorded weather data and irrigation application data are presented in Appendix I.

TABLE B.1.2. Study Use Pattern.

Trial ID (City, State/Year)	EP ¹	Application					Tank Mix Adjuvants ³
		Crop Stage at Application	Rate (lb a.i./A)	RTI ² (days)	Method	Total Rate (lb a.i./A)	
Pikeville, NC/2000	DROPP* SC	Late Season	0.1001; 0.2022	7	Broadcast Foliar Spray	0.3023	N/S
Levelland, TX/2000	DROPP* SC	Late Season	0.0997; 0.2002	7	Foliar Broadcast	0.2999	N/S
Fresno, CA/2000	DROPP* SC	Late Season	0.0982; 0.1985	7	Ground Broadcast	0.2967	N/S

¹EP = End-use Product

²RTI = Retreatment Interval

³N/S = Not Specified

B.2. Analytical Methodology

Residue of thidiazuron and its metabolites were extracted using two methods. Aventis CropScience method AW/01/01 was used to quantify residues of thidiazuron and the crop metabolite photo-thidiazuron; method AW/02/01 was used to extract the soil metabolite 1,2,3-thidiazol-5-ylurea. Independent Laboratory Validations (ILV's) for both methods have been reviewed and accepted.

AW/01/01

Extractable residues of thidiazuron and photo-thidiazuron are removed from crops by blending with 80/20: acetonitrile/water. After blending the resulting extract is filtered, and



diluted to a volume of 200 mL. A 250 μ L aliquot of the forementioned extract is diluted to a final volume of 10 mL with 20/80: acetonitrile/water. This solution is injected into an LC/MS/MS instrument for quantitation.

AW/02/01

Residues of 1,2,3-thidiazol-5-ylurea are extracted from crops by blending with 90/10: methanol/water. The resulting extract is then filtered, and diluted to a volume of 200 mL with 90/10: methanol/water. A 1 μ L aliquot of the forementioned extract is diluted to a final volume of 25 mL with 80/20: water/methanol. This solution is injected into an LC/MS/MS instrument for quantitation.

The limit of quantitation for this study was defined as the lowest level of fortification, which was 0.05 ppm for all analytes. Limits of quantitation and detection (LOQ and LOD) were also calculated for each individual analyte. The LOQ was calculated by adding the average residue value for the untreated control samples to ten times the standard deviation of recoveries from the lowest fortification level (0.05 ppm). Calculated LOQ's were 0.038, 0.063, and 0.040 ppm for thidiazuron, photo-thidiazuron, and thidiazol urea, respectively. LOD's were calculated by adding the average residue value for the untreated control samples to three times the standard deviation of recoveries from the lowest fortification level (0.05 ppm). The LOD's were 0.012, 0.020, and 0.012 ppm for thidiazuron, photo-thidiazuron, and thidiazol urea, respectively.

C. RESULTS AND DISCUSSION

Method verification and procedural recoveries were included in this study, both yielding acceptable results. Method verification control samples were fortified at 0.05, 0.25, and 2.50 ppm. Average method verification recoveries were 93%, 85%, and 78% for thidiazuron, photo-thidiazuron, and thidiazol urea, respectively (Table R-1).



Table R-1. METHOD VERIFICATION RECOVERIES
Recovery of Thidiazuron and Metabolites from Fortified Rotational Crop Control Samples ^a

Analytical Set No.	Sample No.	Crop Matrix	Spike (ppm)	Thidiazuron (TDZ)			Photo-TDZ (isomer)			Thidiazoyl urea		
				ppm	UTC	Recovery	ppm	UTC	Recovery	ppm	UTC	Recovery
Method Verification Recoveries:												
MV1	AV00-011272	Wheat Straw	0.05	0.0421	0.0000	84	0.0351	0.0000	70			
MV1	AV00-011272	Wheat Straw	0.05	0.0459	0.0000	92	0.0366	0.0000	73			
MV1	AV00-011272	Wheat Straw	0.25	0.2375	0.0000	95	0.1971	0.0000	79			
MV1	AV00-011272	Wheat Straw	0.25	0.2339	0.0000	94	0.1665	0.0000	67			
MV1	AV00-011272	Wheat Straw	2.50	1.8588	0.0000	74	2.0434	0.0000	82			
MV1	AV00-011272	Wheat Straw	2.50	2.3150	0.0000	93	2.4828	0.0000	99			
MV2	AV00-011291	Radish Leaves	0.05	0.0472	0.0009	93	0.0474	0.0023	90			
MV2	AV00-011291	Radish Leaves	0.05	0.0390	0.0009	76	0.0453	0.0023	86			
MV2	AV00-011291	Radish Leaves	0.25	0.2483	0.0009	99	0.2370	0.0023	94			
MV2	AV00-011291	Radish Leaves	0.25	0.2633	0.0009	105	0.2648	0.0023	105			
MV2	AV00-011291	Radish Leaves	2.50	2.6472	0.0009	106	2.2199	0.0023	89			
MV2	AV00-011291	Radish Leaves	2.50	2.6472	0.0009	106	2.3055	0.0023	92			
MV3	AV00-011291	Radish Leaves	0.05							0.0420	0.0000	84
MV3	AV00-011291	Radish Leaves	0.05							0.0407	0.0000	81
MV3	AV00-011291	Radish Leaves	0.25							0.1990	0.0000	80
MV3	AV00-011291	Radish Leaves	0.25							0.1931	0.0000	77
MV3	AV00-011291	Radish Leaves	2.50							2.3293	0.0000	93
MV3	AV00-011291	Radish Leaves	2.50							1.9925	0.0000	80
MV4	AV00-011347	Wheat Straw	0.05							0.0333	0.0000	67
MV4	AV00-011347	Wheat Straw	0.05							0.0359	0.0000	72
MV4	AV00-011347	Wheat Straw	0.25							0.1900	0.0000	76
MV4	AV00-011347	Wheat Straw	0.25							0.1866	0.0000	75
MV4	AV00-011347	Wheat Straw	2.50							2.0863	0.0000	83
MV4	AV00-011347	Wheat Straw	2.50							1.8313	0.0000	73
Average Recoveries:				93%±11% (n=12)			85%±12% (n=12)			78%±7% (n=12)		

^a Recoveries were corrected for apparent residues in the associated controls.

Procedural recovery control samples were analyzed concurrently with the treated samples. Average procedural recoveries for controls fortified at 0.05ppm (LOQ) were 89%, 88%, and 81% for thidiazuron, photo-thidiazuron, and thidiazoyl urea, respectively (Table R-3).

Table R-3. PROCEDURAL RECOVERIES
Recovery of Thidiazuron and Metabolites from Fortified Rotational Crop Control Samples ^a

Analytical Set No.	Sample No.	Crop Matrix	Spike (ppm)	Thidiazuron (TDZ)			Photo-TDZ (isomer)			Thidiazoyl urea		
				ppm	UTC	Recovery	ppm	UTC	Recovery	ppm	UTC	Recovery
1	AV00-012013	Mustard	0.05	0.0439	0.0000	88	0.0518	0.0000	94			
2	AV00-011344	Wheat Hay	0.05	0.0403	0.0012	78	0.0464	0.0067	79			
3	AV00-011266	Wheat Forage	0.05	0.0495	0.0051	89	0.0408	0.0052	71			
4	AV00-011990	Wheat Grain	0.05	0.0474	0.0000	95	0.0480	0.0000	96			
5	AV00-011272	Wheat Straw	0.05	0.0422	0.0027	79	0.400	0.0000	80			
6	AV00-011290	Radish Root	0.05	0.0501	0.0000	100	0.0511	0.0006	101			
7	AV00-011366	Carrot Leaves	0.05	0.0511	0.0030	96	0.0470	0.0000	94			
8	AV00-011290	Radish Roots	0.05							0.0487	0.0000	97
9	AV00-011990	Wheat Grain	0.05							0.0416	0.0000	83
10	AV00-011366	Carrot Leaves	0.05							0.0470	0.0000	94
11	AV00-012013	Mustard	0.05							0.0403	0.0000	81
12	AV00-011344	Wheat Hay	0.05							0.0367	0.0000	73
13	AV00-011272	Wheat Straw	0.05							0.0354	0.0000	71
14	AV00-011266	Wheat Forage	0.05							0.0334	0.0000	67
Average Recoveries:				89%±8% (n=7)			88%±11% (n=7)			81%±12% (n=7)		

^a Recoveries were corrected for apparent residues in the associated controls.



No thidiazuron residues greater than LOQ (>0.05 ppm) were found in any of the treated rotational crops. Therefore, samples were not analyzed after the shortest PBI, which is 2-weeks for wheat and 1-month for root, leafy vegetable, and other small grain crops. Samples were not analyzed because it was expected that the recovered residues would be even lower in later plant back intervals.

The limit of quantitation for this study was defined as 0.05 ppm for all analytes. Limits of quantitation and detection (LOQ and LOD) were also calculated for each individual analyte. Calculated LOQ's were 0.038, 0.063, and 0.040 ppm for thidiazuron, photo-thidiazuron, and thiadiazol urea, respectively. The LOD's were 0.012, 0.020, and 0.012 ppm for thidiazuron, photo-thidiazuron, and thiadiazol urea, respectively. Additionally, chromatograms of control samples for various crop matrices were submitted, and are free from interferences.

Data depicting the frozen storage stability of thidiazuron in cotton commodities stored for intervals reflecting the maximum storage intervals incurred by samples is required in order for any magnitude of residue study to be accepted. The maximum storage interval incurred from this rotational crops study was 338 days. The previously submitted cottonseed storage stability study (DP Barcode D195636, F. Fort, 1994) was not adequate due to the nonspecificity of the method and high background levels of thidiazuron; thus, it cannot be used to validate the stability of thidiazuron as stated in the summary of the submitted study.

Weather conditions were reported as typical throughout the duration of this study. Temperature and rainfall recordings were within the average historical values, and irrigation was used to supplement as needed for each site. However, high temperatures did effect the summer spinach crop planted in Texas, where temperature are typically high. Spinach planted at the 8 month plant back did not survive, but since leafy vegetable crops were not analyzed past the 1-month PBI, this was only a minor incident. Recorded weather and irrigation application data are presented in Appendix I.

Although the field trial residue data submitted was adequate, this study is unacceptable due to the inadequacy of the confined accumulation in rotational crops study. An unknown metabolite, accounting for 0.046 ppm (26.4 % TRR) in 14-DAT wheat straw and 0.010 ppm (8.8% TRR) in 120-DAT wheat straw, was *not* characterized during the confined study. According to OPPTS Guideline No. 860.1300, radioactive residues of 0.01-0.05 ppm must be characterized. The confined nor the field accumulation in rotational crops study can be approved with the toxicity and characterization of a metabolite *remaining unknown*.

D. CONCLUSION

The field accumulation in rotational crops study is adequate for quantifying thidiazuron, photo-thidiazuron, and thiadiazol urea residues in/on leafy vegetable, root, and small grain crops. The study had no major deficiencies and produced method validation and procedural recovery



Thidiazuron/120301/Bayer CropScience/

DACO 7.4.4/OPPTS 860.1900/OECD IIA 6.6.3, 6.8.7 and IIIA 8.6

Field Accumulation in Rotational Crops - Leafy vegetable, root, and small grain crops

residues within the acceptable limit of 70-120%. No meteorological abnormalities were reported during the conduct of the study. However, this study cannot be accepted because of the results from the confined accumulation in rotational crops study. The registrant must provide further characterization of the unknown metabolite, in addition to storage stability data, if the confined and field accumulation in rotational crop studies are to be supported.

E. DOCUMENT TRACKING

RDI: T. Jimerson (11/8/04)

DP Barcode(s): D294543

PC Code: 120201



Appendix I: Application Site Weather Reports



Pikeville, NC

**Table C-28 Trial 25819-04 North Carolina,
 Monthly Weather Data Summary**

Month/Year	Air Temperature (°C)		Soil Temperature (°C)				Precipitation (mm)	
	Max.	Min.	@50 mm		@200 mm		Observed	10-yr Avg
August 2000	35	12					134.1	133.0
September 2000	32	9					151.2	146.7
October 2000	31	1	28	8	29	15	0.0	94.6
November 2000	27	-5	22	1	22	7	65.3	68.8
December 2000	23	-9	15	0	15	4	23.1	61.1
January 2001	24	-10	15	0	14	3	25.0	112.2
February 2001	27	-4	19	2	18	7	63.7	63.3
March 2001	25	-4	20	2	19	7	146.4	108.7
April 2001	33	-1	31	6	28	11	15.5	66.5
May 2001	34	8	34	14	33	21	167.6	87.0
June 2001*	35	17	39	19	40	27	59.1	67.3
July 2001*	36	14	36	20	36	29	120.1	133.8
August 2001	37	15	36	21	36	30	137.8	133.0
September 2001	32	7	32	14	34	23	51.7	146.7

Climatological data were collected from an on site weather station.

**Table C-29 Trial 25819-04 North Carolina,
 Irrigation Applied**

Date	Amount (in)	Method	Source
2 Nov 2000	0.8	Sprinkler	Pond
3 May 2001	0.66	Sprinkler	Pond



Levelland, TX

**Table C-14 Trial 25819-02 Texas,
 Monthly Weather Data Summary**

Month/Year	Air Temperature (°F)		Soil Temperature (°F)				Precipitation (in)	
	Max.	Min.	@2 In		@8 In		Observed	10-yr Avg
May 2000	91	69	95	76	83	77	0.07	2.53
June 2000	84	64					6.26	3.68
July 2000	93	66	100	75	87	79	2.51	1.51
August 2000	94	65	103	77	89	81	0.80	1.90
September 2000	91	57	89	73	85	79	0.00	2.29
October 2000	78	50	78	60	69	64	4.72	1.06
November 2000	55	31	59	40	50	46	0.78	0.61
December 2000	48	23	44	35	43	40	0.38	0.86
January 2001	48	27	41	35	38	36	0.42	0.62
February 2001	59	31	52	40	46	42	0.35	0.66
March 2001	60	37	56	44	51	48	2.67	0.78
April 2001	67	49	60	59	66	61	0.02	1.77
May 2001	84	55	88	66	75	66	4.90	2.53
June 2001	94	64	107	79	95	86	0.30	3.68
July 2001	98	68	112	85	111	105	0.00	1.51
August 2001	92	65	100	77	99	92	3.05	1.90
September 2001	85	57	80	69	102	86	1.98	2.29
October 2001	77	44	79	59	68	63	0.21	1.06

Climatological data were collected on site from STAR HP-32 weather station, Levelland Texas. The data for June 2000 as well as the 10 year historical weather data from 1990 to 2000 were collected at NOAA Weather Station 72267, in Lubbock, Texas, located about 40 miles from the test site.

**Table C-15 Trial 25819-02 Texas,
 Irrigation Applied**

Date	Crops	Amount (~in)	Method/Source
28 Apr 2000	Cotton	2	Furrow/Well
23 May 2000	Cotton	5	Furrow/Well
20 Nov 2000	Leaf and Root Crops	2	Furrow/Well
1 Dec 2000	Leaf and Root Crops	1.5	Furrow/Well
11 Dec 2000	All Rotational Crops	2	Furrow/Well
20 Dec 2000	Leaf and Root Crops	2	Furrow/Well
5-8 Feb 2001	All Rotational Crops	6	Furrow/Well
20 Feb 2001	Leaf and Small Grain Crops	2	Furrow/Well
14 Mar 2001	Leaf Crop	3	Furrow/Well
4 Apr 2001	Leaf and Small Grain Crops	3	Furrow/Well
17 Apr 2001	Leaf and Small Grain Crops	3	Furrow/Well
22 May 2001	All Rotational Crops	2	Furrow/Well
8 Jun 2001	Leaf and Root Crops	3	Furrow/Well
16 Jun 2001	All Rotational Crops	3	Furrow/Well
22 Jun 2001	Leaf Crop	2	Furrow/Well
2 Jul 2001	Leaf Crop	2	Furrow/Well



Fresno, CA

**Table C-21 Trial 25819-03 California,
 Monthly Weather Data Summary**

Month/Year	Air Temperature (°C)		Soil Temperature (°C)				Precipitation (mm)	
	Max.	Min.	@60 mm		@200 mm		Observed	10-yr Avg
October 2000	33	6	29	12	26	13	67.7	16.43
November 2000	22	-2.5	21	6	18	7	0.2	16.69
December 2000	18	-4	18	5	15	6	4.2	22.36
January 2001	18	-3	16	4	12	4	51.7	64.17
February 2001	20	-3	20	5	15	6	45.1	47.50
March 2001	30	1	28	6	24	7	40.0	42.70
April 2001	32	1	30	9	26	9	27.8	12.34
May 2001	39	7	33	13	30	14	0.0	11.62
June 2001*	40	11					0.1	6.67
July 2001*	42	13					4.2	0.02
August 2001	40	13	35	19	31	20	0.0	0.00
September 2001	37	10	33	16	28	17	0.2	1.36
October 2001	36	7	30	14	26	15	9.1	18.43

Climatological data were collected from an on site weather station.

* Due to a weather station malfunction, the reported data were collected from a DowAgrosciences weather station approximately 1 mile southeast of the field site.



**Table C-22 Trial 25819-03 California,
 Irrigation Applied**

Date	Amount (in)	Method	Source
21 Jun 2000	5.0	Furrow	Well
10 Jul 2000	4.0	Furrow	Well
28 Jul 2000	4.0	Furrow	Well
2 Aug 2000	4.0	Furrow	Well
10 Aug 2000	3.0	Furrow	Well
15 Aug 2000	3.0	Furrow	Well
22 Aug 2000	3.5	Furrow	Well
5 Sep 2000	3.5	Furrow	Well
12 Sep 2000	3.5	Furrow	Well
19 Sep 2000	2.0	Furrow	Well
Cotton Harvested, Rotational Crops Planted			
17 Nov 2000	0.72	Sprinkler	Well
20 Nov 2000	0.38	Sprinkler	Well
21 Nov 2000	0.48	Sprinkler	Well
1 Dec 2000	0.65	Sprinkler	Well
9 Dec 2000	0.24	Sprinkler	Well
15 Dec 2000	0.48	Sprinkler	Well
21 Dec 2000	0.38	Sprinkler	Well
3 Jan 2001	0.38	Sprinkler	Well
19 Mar 2001	3.0	Furrow	Well
30 Mar 2001	4.0	Furrow	Well
18 Apr 2001	2.5	Furrow	Well
26 Apr 2001	3.0	Furrow	Well
3 May 2001	3.0	Furrow	Well
9 May 2001	3.5	Furrow	Well
17 May 2001	3.0	Furrow	Well
1 Jun 2001	3.0	Furrow	Well
8 Jun 2001	3.0	Furrow	Well
14 Jun 2001	3.0	Furrow	Well
26 Jun 2001	5.0	Furrow	Well
1 Jul 2001	0.60	Sprinkler	Well
3 Jul 2001	0.24	Sprinkler	Well
4 Jul 2001	0.24	Sprinkler	Well
5 Jul 2001	0.24	Sprinkler	Well
6 Jul 2001	0.24	Sprinkler	Well
7 Jul 2001	0.24	Sprinkler	Well
8 Jul 2001	0.24	Sprinkler	Well
13 Jul 2001	2.0	Furrow	Well
20 Jul 2001	2.5	Furrow	Well
26 Jul 2001	2.5	Furrow	Well
2 Aug 2001	2.0	Furrow	Well
10 Aug 2001	2.0	Furrow	Well
16 Aug 2001	2.0	Furrow	Well
24 Aug 2001	1.5	Furrow	Well
29 Aug 2001	1.0	Furrow	Well
5 Sep 2001	1.0	Furrow	Well
14 Sep 2001	1.0	Furrow	Well
20 Sep 2001	1.0	Furrow	Well
1 Oct 2001	2.0	Furrow	Well



Primary Evaluator Toiya Jimerson, Chemist, Date: 11/8/04

Reregistration Branch 1,
Health Effects Division

Reviewer

Felecia Fort, Chemist,
Reregistration Branch 1,
Health Effects Division

Whang Phang, Ph.D.,
Branch Senior Scientist
Reregistration Branch 1
Health Effects Division

STUDY REPORTS:

MRID No.: 46298402 Rolando Perez and Steven Perez (April 5, 2002) Independent Laboratory Validation of Aventis Method AW/01/01 Thidiazuron: Analytical Method for the Determination of Thidiazuron and its Metabolite Photo-Thidiazuron in Crop Matrices Using LC/MS/MS. Lab Project Number: B003830. Unpublished study prepared by Rolando Perez and Steven Perez. 74 pages.

EXECUTIVE SUMMARY:

Aventis CropScience Method AW/01/01 quantifies residues of thidiazuron (TDZ) and its metabolite photo-thidiazuron (P-TDZ) in crops using high performance liquid chromatography with a triple quadrupole mass spectrometer. The independent laboratory validation (ILV) study for method AW/01/01 uses corn forage as its experimental matrix, although mustard greens, and radish roots and tops were used as the crop matrixes in the residue analytical method study. The limit of quantification (LOQ) is 0.05 ppm (50 ppb) for this method.

Extractable residues of thidiazuron and photo-thidiazuron are removed from crops by blending with 80/20: acetonitrile/water. After blending the resulting extract is filtered, and diluted to a volume of 200 mL. A 250 μ L aliquot of the forementioned extract is diluted to a final volume of 10 mL with 20/80: acetonitrile/water. This solution is injected into an LC/MS/MS instrument. Quantitation was successfully completed in the second trial for field corn forage.

Corn forage samples were fortified at 50 ppb and 100 ppb. Average residue recoveries of TDZ and P-TDZ in/on corn forage were 76.8% and 72.0%, respectively, at the 50 ppb fortification level. Average residue recoveries at the 100 ppb fortification level were 70.4% and 77.6%, respectively, for thidiazuron and photo-thidiazuron. The submitted ILV trial for



analytical method AW/01/01 is adequate.

STUDY/WAIVER ACCEPTABILITY/DEFICIENCIES/CLARIFICATIONS:

Under the conditions and parameters used in the study, the ILV is classified as scientifically acceptable.

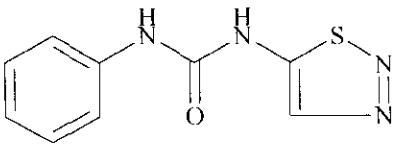
The acceptability of this study for regulatory purposes is addressed in the forthcoming U.S. EPA Residue Chemistry Summary Document.

COMPLIANCE:

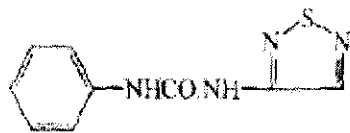
Signed and dated GLP, Quality Assurance and Data Confidentiality statements were provided. There were no reported deviations from regulatory practices during this study.

A. BACKGROUND INFORMATION

Thidiazuron has one registered use as a cotton defoliant. Currently, the combined residues of thidiazuron and its aniline containing metabolites are regulated (40 CFR180.403). The Metabolism Assessment Review Committee (9/29/95, F. Fort) has determined that the residue of concern in plant commodities consists of the parent thidiazuron only, and that the residues of concern in animal commodities consist of the parent thidiazuron, 4-hydroxythidiazuron, and phenyl urea. Tolerances are established in/on cottonseed (0.4 ppm), cottonseed hulls (0.8 ppm), eggs (0.1 ppm), and milk (0.05 ppm). Additional tolerances are set at 0.2 ppm for the fat, meat and meat by-products of cattle, goats, hogs, horses, poultry and sheep.

Compound	Chemical Structure 
Common name	Thidiazuron
IUPAC name	1-phenyl-3-(1,2,3-thiadiazol-5-yl)urea
CAS name	<u>N</u> -phenyl- <u>N'</u> -1,2,3-thiadiazol-5-ylurea
CAS #	51707-55-2



Compound	Chemical Structure
	
Common name	Photo-Thiazuron
IUPAC name	1-phenyl-3-(1,2,5-thiazolidin-3-yl)urea
CAS name	N-phenyl-N'-1,2,5-thiazolidin-5-ylurea
CAS #	71769-74-9

B. MATERIALS AND METHODS

B.1. Data-Gathering Method

B.1.1. Principle of the Method:

Residues of thiazuron and photo-thiazuron are extracted from crop matrices by blending with 80/20: acetonitrile/water. The extracts are filtered through a Buchner funnel and brought to a volume of 200mL with 80/20: acetonitrile/water in a graduated mixing cylinder. A 250 μ L aliquot of the extract is transferred to a 10 mL flask and diluted with 20/80: acetonitrile/water. Following the final dilution, a 75 μ L aliquot is injected into an LC/MS/MS instrument.

Method ID	AW/01/01
Analyte(s)	TDZ and P-TDZ
Extraction solvent/technique	Acetonitrile/Water by blending
Instrument/Detector	LC/MS/MS
Stability of std solutions	At least 3 months at 0 °C
Average Retention times (min.)	TDZ 8.32; P-TDZ 9.03

C. RESULTS AND DISCUSSION

Average residue recoveries of TDZ and P-TDZ in/on corn forage were 76.8% and 72.0%, respectively, at the 50 ppb fortification level. Average residue recoveries at the 100 ppb



fortification level were 70.4% and 77.6%, respectively, for thidiazuron and photo-thidiazuron (Tables C.1. and C.2.). HPLC analysis reported TDZ to have an average retention time of 8.32 minutes, and 9.03 minutes for P-TDZ in corn forage. Under the parameters of this study, the ILV trial is adequate.

Matrix	Spiking Level (ppb)	Recoveries Obtained (ppb)	Mean Percent Recovery \pm SD (CV)
Corn Forage	0	0	0
Corn Forage	50	38.40, 38.40	76.8 \pm 0.00
Corn Forage	100	70.40, 70.40	70.4 \pm 0.00

Matrix	Spiking Level (ppb)	Recoveries Obtained (ppb)	Mean Recovery \pm SD (CV)
Corn Forage	0	0	0
Corn Forage	50	35.20, 36.80	72.0 \pm 2.26
Corn Forage	100	76.80, 78.40	77.6 \pm 1.13

C.1. Independent Laboratory Validation

The ILV trial submitted for method AW/01/01 was conducted according to EPA guideline specifications. No major deficiencies were found in the method, and acceptable recoveries were obtained in the second trial. A 1:8 dilution was performed on the sample after the first trial produced low recoveries due to matrix effects, commonly called signal suppression. This change required technical communication between the performing laboratory and the study monitor. The dilution did not effect any other parameters of the study.

Suggestions from the performing laboratory include:

1. Increasing the injected volume from 75 μ L to 100 μ L in order to obtain a better signal to noise ratio.
2. Conditioning a new HPLC column with a control sample to obtain better instrument sensitivity.
3. Diluting the sample if matrix effects are encountered. A needle spike of a control sample can be made in order to determine if matrix effects are present.



D. CONCLUSION


The ILV trial for Aventis CropScience method AW/01/01 is adequate for quantifying thidiazuron and photo-thidiazuron residues in/on corn forage. The method had no major deficiencies and produced recovery residues within the acceptable limit of 70-120%.

E. DOCUMENT TRACKING

RDI: Toiya Jimerson (11/8/04)
DP Barcode(s): D294543
PC Code: 120301



Primary Evaluator Toiya Jimerson, Chemist, Date: 11/8/04
Reregistration Branch 1,
Health Effects Division

Reviewer Felecia Fort, Chemist, 
Reregistration Branch 1,
Health Effects Division

Whang Phang, Ph.D.,
Branch Senior Scientist
Reregistration Branch 1
Health Effects Division

STUDY REPORTS:

MRID No.: 46298403 Rolando Perez and Steven Perez (April 5, 2002) Independent Laboratory Validation of Aventis Method AW/02/01 Thidiazuron: Analytical Method for the Determination of 1,2,3-thidiazol-5-ylurea (AE F132345) Residues in Crop Matrices Using LC/MS/MS. Lab Project Number: B003831. Unpublished study prepared by Rolando Perez and Steven Perez. 62 pages.

EXECUTIVE SUMMARY:

Aventis CropScience Method AW/02/01 quantifies residues of 1,2,3-thidiazol-5-ylurea in crops using high performance liquid chromatography with a triple quadrupole mass spectrometer. The independent laboratory validation(ILV) study for method AW/02/01 uses corn forage as its experimental matrix, although mustard greens, wheat forage, and radish roots and tops were used as the crop matrices in the residue analytical method study. The limit of quantitation (LOQ) is 0.05 ppm (50 ppb) for this method.

Residues of 1,2,3-thidiazol-5-ylurea are extracted from crops by blending with 90/10: methanol/water. The resulting extract is then filtered, and diluted to a volume of 200 mL with 90/10: methanol/water. A 1 μ L aliquot of the forementioned extract is diluted to a final volume of 25 mL with 80/20: water/methanol. This solution is injected into an LC/MS/MS instrument. Quantitation was successfully completed in the first trial for field corn forage.

Corn forage samples were fortified at 50 ppb, 100 ppb, and 250 ppb. Overall recoveries of 1,2,3-thidiazol-5-ylurea in/on corn forage at the 50, 100, and 250 fortification levels were 80.5%, 79.8%, and 82.6%, respectively. The submitted ILV trial for analytical method AW/02/01 is adequate.



STUDY/WAIVER ACCEPTABILITY/DEFICIENCIES/CLARIFICATIONS:

Under the conditions and parameters used in the study, the field trial residue data are classified as scientifically acceptable.

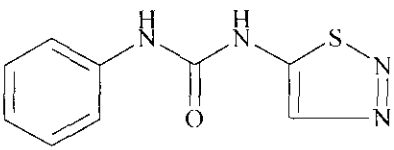
The acceptability of this study for regulatory purposes is addressed in the forthcoming U.S. EPA Residue Chemistry Summary Document.

COMPLIANCE:


Signed and dated GLP, Quality Assurance and Data Confidentiality statements were provided. There were no reported deviations from regulatory practices during this study.

A. BACKGROUND INFORMATION

Thidiazuron has one registered use as a cotton defoliant. Currently, the combined residues of thidiazuron and its aniline containing metabolites are regulated (40 CFR180.403). The Metabolism Assessment Review Committee (9/29/95, F. Fort) has determined that the residue of concern in plant commodities consists of the parent thidiazuron only, and that the residues of concern in animal commodities consist of the parent thidiazuron, 4-hydroxythidiazuron, and phenyl urea. Tolerances are established in/on cottonseed (0.4 ppm), cottonseed hulls (0.8 ppm), eggs (0.1 ppm), and milk (0.05 ppm). Additional tolerances are set at 0.2 ppm for the fat, meat and meat by-products of cattle, goats, hogs, horses, poultry and sheep.

Compound	Chemical Structure
	
Common name	Thidiazuron
IUPAC name	1-phenyl-3-(1,2,3-thiazol-5-yl)urea
CAS name	<u>N</u> -phenyl- <u>N</u> '-1,2,3-thiadiazol-5-ylurea
CAS #	51707-55-2



Compound	Chemical Structure 
Common name	Thiadiazol urea
IUPAC name	1,2,3-thiadiazol-5-ylurea
CAS name	1,2,3-thiadiazol-5-ylurea
CAS #	NA

B. MATERIALS AND METHODS

B.1. Data-Gathering Method

B.1.1. Principle of the Method:

Residues of 1,2,3-thiadiazol-5-ylurea are extracted from crops by blending with 90/10: methanol/water. The extracts are filtered through a Buchner funnel and brought to a volume of 200 mL with 90/10: methanol/water in a graduated mixing cylinder. A 1 μ L aliquot of the forementioned extract is diluted to a final volume of 25 mL with 80/20: water/methanol. Following the final dilution, a 50 μ L aliquot is injected into an LC/MS/MS instrument. Quantitation of 1,2,3-thiadiazol-5-ylurea was accomplished by high performance liquid chromatography with a triple quadruple mass spectrometer.

Method ID	AW/02/01
Analyte(s)	1,2,3-thiadiazol-5-ylurea
Extraction solvent/technique	Methanol/Water by blending
Instrument/Detector	LC/MS/MS
Stability of std solutions	At least 3 months at 0 °C
Average Retention time (min.)	5.98

C. RESULTS AND DISCUSSION

Overall recoveries of 1,2,3-thiadiazol-5-ylurea in/on corn forage at the 50, 100, and 250 fortification levels were 80.5%, 79.8%, and 82.6%, respectively (Table C.1.). HPLC analysis



showed the analyte to have an average retention time of 5.98 minutes in corn forage. Under the parameters of this study, the ILV trial is adequate.

Matrix	Spiking Level (ppb)	Recoveries Obtained (ppb)	Mean Percent Recovery \pm SD (CV)
Corn Forage	0	0, 0	0
Corn Forage	50	39.00, 41.50	80.50 \pm 3.54
Corn Forage	100	81.50, 78.00	79.8 \pm 2.47
Corn Forage	250	209.00, 204.00	82.60 \pm 1.41

C.1. Independent Laboratory Validation

The ILV trial submitted for method AW/02/01 was conducted according to EPA guideline specifications. No major deficiencies were found in the method, and acceptable recoveries were obtained in the first trial. A 1:2 dilution was performed on the 250 ppb fortified sample, however the results were not used because the method's original dilution produced adequate results.

Communication occurred between the performing laboratory and the study monitor prior to the start of the first trial on February 25, 2002. Two minor typographical errors were corrected in the method as a result of this communication.

Suggestions from the performing laboratory included:

1. Increasing the injected volume from 50 μ L to 75 μ L in order to obtain a better signal to noise ratio.
2. Conditioning a new HPLC column with a control sample to obtain better instrument sensitivity.
3. Incorporating additional chromatograms into the method including: one chromatogram per standard concentration level and one chromatogram per fortification level.

D. CONCLUSION

The ILV trial for Aventis CropScience method AW/02/01 is adequate for quantifying 1,2,3-thidiazol-5-ylurea residues in/on corn forage. The method had no major deficiencies and produced recovery residues within the acceptable limit of 70-120%.

E. DOCUMENT TRACKING



Thidiazuron/120301/Aventis CropScience
DACO 7.2.1, 7.2.2, and 7.2.3/OPPTS 860.1340/OECD IIA 4.2.5, 4.2.6 and 4.3
Residue Analytical Method- Plant

RDI: Toiya Jimerson (11/8/04)

DP Barcode(s): D294543

PC Code: 120301



Primary Evaluator Toiya Jimerson, Chemist, Date: 11/8/04
Reregistration Branch 1,
Health Effects Division

Reviewer Felecia Fort, Chemist,
Reregistration Branch 1,
Health Effects Division

Wang Phang, Ph.D.,
Branch Senior Scientist
Reregistration Branch 1
Health Effects Division

STUDY REPORTS:

MRID No.: 46298404 Lee E. Williams (November 18, 2002) Stability of Thidiazuron and its Metabolites 4-Hydroxy Thidiazuron and Phenyl Urea in Milk and Beef Tissues During Frozen Storage, USA. 1995 Lab Project Number: AW95R003. Unpublished study prepared by Lee E. Williams. 90 pages.

EXECUTIVE SUMMARY:

Samples of whole milk and beef muscles were spiked with thidiazuron, 99.4% ai, at levels of 0.10-0.25 ppm and were stored between -10 °C and -20 °C for a duration of at least 2 years. Under these conditions, residues of thidiazuron (TDZ) and its metabolite 4-hydroxy thidiazuron (4-OH-TDZ) showed good stability with most recoveries between 70-120% in milk and beef muscle. The half life values of TDZ and 4-OH-TDZ were found to be 6 and 14 years, respectively in beef muscle and whole milk.

Aged and freshly fortified whole milk and beef muscle samples were analyzed for 4-OH-TDZ and TDZ, respectively.

Extractable residues of thidiazuron and relevant metabolites were removed from milk by blending with acetonitrile. The acetonitrile extract was filtered, partitioned with hexane, and extracted with dichloromethane. The acetonitrile/dichloromethane extract was then rotary evaporated to dryness. After reconstitution in 1:1 acetonitrile/deionized water, 4-OH-TDZ was quantified by HPLC with ultraviolet detection at 290nm.

Thidiazuron and relevant metabolites were extracted from beef muscle by blending with methanol. The methanolic extract was filtered and diluted with acetonitrile. The acetonitrile/methanol extract was partitioned with hexane and rotary evaporated to dryness.



Following sample cleanup via solid phase extraction, thidiazuron residue was reconstituted in 1:1 acetonitrile/deionized water and quantified by HPLC with ultra violet detection at 290nm.

The experimental procedures correspond to the outlined analytical method with the following exceptions:

1. The determination of the stability of phenyl urea in beef liver was terminated. Two method procedural attempts of analyzing beef liver samples for phenyl urea residues were unsuccessful, so the storage stability study was aborted.
2. Sample evaporation could not be achieved at Step 5.2.5 of RAM AW/01/96 for the 0-Day beef tissue set.
3. The last sampling for each matrix was not analyzed at the storage intervals required in the protocol.
4. On day 752, the fresh fortification for whole milk was fortified at 0.04 ppm with 4-OH-TDZ instead of 0.10 ppm.

The data indicates that residues of thidiazuron in beef muscle and 4 hydroxy thidiazuron in whole milk are stable at -10 - -20 °C for 2 years. However, the study is unacceptable because no recoveries were obtained for the thidiazuron metabolite phenyl urea (PU). In addition, samples were fortified with either TDZ or 4-OH TDZ, but never all three residues of concern: TDZ, 4-OH TDZ, and PU.

STUDY/WAIVER ACCEPTABILITY/DEFICIENCIES/CLARIFICATIONS:

Under the conditions and parameters used in the study, the field trial residue data are classified as scientifically unacceptable because all three residues of concern, TDZ, 4-OH TDZ, and PU, were not collected simultaneously.

The acceptability of this study for regulatory purposes is addressed in the forthcoming U.S. EPA Residue Chemistry Summary Document.

COMPLIANCE:

Signed and dated GLP, Quality Assurance and Data Confidentiality statements were provided. Deviations from regulatory requirements included:

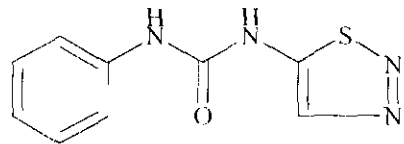
1. Some raw data entries were not made and/or corrected in accordance with 40CFR160.130(e).
2. Findings from one quality assurance inspector were not reported to management in a timely manner. However, there were no adverse finding during this inspection.

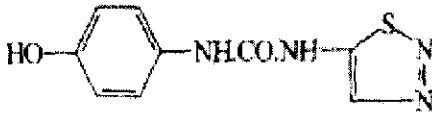


40 CFR 160.35(b)(4).

A. BACKGROUND INFORMATION

Thidiazuron has one registered use as a cotton defoliant. Currently, the combined residues of thidiazuron and its aniline containing metabolites are regulated (40 CFR 180.403). The Metabolism Assessment Review Committee (9/29/95, F. Fort) has determined that the residue of concern in plant commodities consists of the parent thidiazuron only, and that the residues of concern in animal commodities consist of the parent thidiazuron, 4-hydroxythidiazuron, and phenyl urea. Tolerances are established in/on cottonseed (0.4 ppm), cottonseed hulls (0.8 ppm), eggs (0.1 ppm), and milk (0.05 ppm). Additional tolerances are set at 0.2 ppm for the fat, meat and meat by-products of cattle, goats, hogs, horses, poultry and sheep.

Compound	Chemical Structure 
Common name	Thidiazuron
IUPAC name	1-phenyl-3-(1,2,3-thiazol-5-yl)urea
CAS name	N-phenyl-N'-1,2,3-thiazol-5-ylurea
CAS #	51707-55-2

Compound	Chemical Structure 
Common name	4-Hydroxy Thidiazuron
IUPAC name	1-(4-hydroxyphenyl)-3-(1,2,3-thiazol-5-yl)urea
CAS name	N-(4-hydroxyphenyl)-N'-1,2,3-(thiazol-5-yl)urea
CAS #	NA



B. EXPERIMENTAL DESIGN

B.1. Sample Preparation

Whole milk and beef muscles were both purchased at local grocers. No sample preparation was performed for whole milk. Beef tissues were ground in a Hobart meat grinder. The ground tissues were bottled and assigned a sample number for use as the bulk matrix.

Thirty-three representative sub-samples of beef tissue (20.0 g each) were weighed in 8 oz. Mason jars. Samples for aging beef muscle were fortified with thidiazuron at 0.25 ppm. Thirty-three representative sub-samples of whole milk (50.0 g each) were weighed in 8 oz. Mason jars. Samples for aging milk were fortified with 4-hydroxy thidiazuron at 0.10 ppm. Thirty-three untreated control samples for each matrix were also stored in Mason jars. All Mason jars were closed with aluminum screw caps, placed inside closed cardboard boxes, and stored in laboratory freezers.

B.2. Analytical Methodology

Aged and freshly fortified whole milk and beef muscle samples were analyzed for 4-hydroxy thidiazuron and thidiazuron, respectively. A sample set included one control, two freshly fortified recovery samples, and three aged samples of each matrix. The limit of quantification (LOQ) was found to be 0.01 ppm for 4-OH-TDZ in whole milk and 0.05 ppm for TDZ in beef muscle.

Extractable residues of thidiazuron and relevant metabolites were removed from milk by blending with acetonitrile. The acetonitrile extract was filtered, partitioned with hexane, and extracted with dichloromethane. The acetonitrile/dichloromethane extract was then rotary evaporated to dryness. After reconstitution in 1:1 acetonitrile/deionized water, 4-OH-TDZ residue was quantified by HPLC with ultraviolet detection at 290nm.

Thidiazuron and relevant metabolites were extracted from beef muscle by blending with methanol. After extraction, the methanolic extract was filtered and diluted with acetonitrile. The acetonitrile/methanol extract was partitioned with hexane and rotary evaporated to dryness. Following sample cleanup via solid phase extraction, thidiazuron residue was reconstituted in 1:1 acetonitrile/deionized water and quantified by HPLC with ultra violet detection at 290nm.

The experimental procedures correspond to the outlined analytical method with the following exceptions:

1. The determination of the stability of phenyl urea in beef liver was terminated.
2. Sample evaporation could not be achieved at Step 5.2.5 of RAM AW/01/96 for the 0-day



- beef tissue set.
3. The last sampling for each matrix was not analyzed on the storage intervals required in the protocol.
 4. Mile analysis at day 752, the fresh fortification was fortified at 0.04 ppm with 4-OH-TDZ instead of 0.10 ppm.

C. RESULTS AND DISCUSSION

Whole milk recoveries for 4 hydroxy thidiazuron ranged between 92-102%, also showing good stability (Table C.1.). However, control values for 4-OH TDZ were variable. With an LOQ of 0.01 ppm, 0-day and 752-day milk had residue values of 0.0228 and 0.0101 ppm, respectively. Data for thidiazuron in beef muscle showed good stability with the exception of 751-day beef which had a recovery of only 64%, 0.1601/0.25 ppm, (Table C.2.). Control values for beef were all less than the LOQ (0.05ppm).

No data was collected for the recoveries of phenyl urea during this study. Two method procedural attempts of analyzing beef liver samples for PU were unsuccessful, and the storage stability study for this analyte was aborted. In addition, samples were fortified with TDZ and either 4-OH TDZ, but never all three residues of concern: TDZ, 4-OH TDZ, and PU. For these reasons, the analytical method for this study could not be accepted.

TABLE C.1. Summary of Concurrent Recoveries of 4-Hydroxy-Thidiazuron from Whole Milk

Matrix	Analyte	Spike level (ppm)	Storage Interval (days)	Recovered residues (ppm) ¹	Recoveries (%)
Whole milk	4-OH-TDZ	0.10	0	0.0987	98.7
Whole milk	4-OH-TDZ	0.10	92	0.1097	109.7
Whole milk	4-OH-TDZ	0.10	172	0.1013	101.3
Whole milk	4-OH-TDZ	0.10	256	0.0919	91.9
Whole milk	4-OH-TDZ	0.10	366	0.0858	85.8
Whole milk	4-OH-TDZ	0.10	550	0.1023	102.3
Whole milk	4-OH-TDZ	0.10	752	0.0919	91.9

¹ Average determined residue for triplicate analyses.

TABLE C.2. Summary of Concurrent Recoveries of Thidiazuron from Beef Muscle

Matrix	Analyte	Spike level (ppm)	Storage Interval (days)	Recovered residues (ppm) ¹	Recoveries (%) ²
Beef muscle	TDZ	0.25	0	0.2291	91.6
Beef muscle	TDZ	0.25	92	0.2190	87.6
Beef muscle	TDZ	0.25	182	0.2015	80.6



Beef muscle	TDZ	0.25	273	0.1956	78.2
Beef muscle	TDZ	0.25	365	0.1786	71.4
Beef muscle	TDZ	0.25	547	0.2306	92.2
Beef muscle	TDZ	0.25	751	0.1601	64.0

¹ Average determined residue for triplicate analyses.

² Thidiazuron result corrected for recovery value of 79.14 %.

D. CONCLUSION

The submitted storage stability study is inadequate because acceptable residue recoveries could not be obtained for phenyl urea in beef tissues. In addition, samples were fortified with either thidiazuron or 4 hydroxy thidiazuron, but never all three residues of concern: TDZ, 4-OH TDZ, and PU. The registrant must demonstrate an analytical method capable of determining recovery residues in each matrix fortified simultaneously for all three residues of concern in order for the Agency to accept this study.

E. DOCUMENT TRACKING

RDI: Toiya Jimerson (11/8/04)
DP Barcode(s): D294543
PC Code: 120301

DATA PACKAGE BEAN SHEET

Date: 21-Jul-2004

Page 1 of 3

*** Registration Information ***

Registration: RED-4092-19149 - Thidiazuron RED

Company: -

Risk Manager: RM 51 - Susan Lewis - (703) 308-8009 Room# CM-2 604P

Risk Manager Reviewer: Stephanie Plummer SPLUMMER

Sent Date: _____ Calculated Due Date: _____ Edited Due Date: _____

Type of Registration: Project

Action Desc: _____

Ingredients: 120301, Thidiazuron

*** Data Package Information ***

Expedite: Yes No Date Sent: 21-Jul-2004 Due Back: _____

DP Ingredient: 120301, Thidiazuron

DP Title: Thidiazuron residue chemistry studies

CSF Included: Yes No Label Included: Yes No Parent DP #: _____

Assigned To	Date In	Date Out	
Organization: <u>HED / RRB1</u>	_____	_____	Administrative Due Date: _____
Team Name: _____	_____	_____	Negotiated Due Date: <u>30-Sep-2004</u>
Reviewer Name: _____	_____	_____	Projected Completion Date: _____
Contractor Name: _____	_____	_____	

*** Studies Sent for Review ***

Printed on Page 2

*** Additional Data Package for this Decision ***

Printed on Page 3

*** Data Package Instructions ***

Attn: Felicia Fort
 Please review the following thidiazuron residue chemistry studies: MRID 462984-01, -02, -03, and -04. Thank you. CRM is Stephanie Plummer, 305-0076.

MDL	Study Title	Decision
46298400	Bayer CropScience LP (2004) Submission of Residue and Fate Data in Support of the Reregistration of Thidiazuron. Transmittal of 4 Studies.	
46298401	Norris, F. (2002) Thidiazuron: Magnitude of Residues in Rotational Crops Following Cotton Treated with Dropp. Project Number: 00AW25819, B003793, 25819/02. Unpublished study prepared by Aventis CropScience and South Texas Ag. Research. 248 p.	860.1900/Field accumulation in ro
46298402	Perez, R.; Perez, S. (2002) Independent Laboratory Validation of Aventis Method AW/01/01 Thidiazuron: Analytical Method for the Determination of Thidiazuron and its Metabolite Photo-Thidiazuron in Crop Matrices Using LC/MS/MS: Final Report. Project Number: B003830, 02AW27947, ADPEN/2K2AV/0131/02. Unpublished study prepared by Adpen Labs. 74 p.	860.1340/Residue analytical meth
46298403	Perez, R.; Perez, S. (2002) Independent Laboratory Validation of Aventis Method AW/02/01 Thidiazuron: Analytical Method for the Determination of 1,2,3-thidiazol-5-ylurea (AE F132345) Residues in Crop Matrices Using LC/MS/MS: Final Report. Project Number: 02AW27947U, ADPEN/2K2AV/0131/01, B003831. Unpublished study prepared by Adpen Labs. 62 p.	860.1340/Residue analytical meth
46298404	Williams, L. (2002) Stability of Thidiazuron and its Metabolites 4-Hydroxy Thidiazuron and Phenyl Urea in Milk and Beef Tissues During Frozen Storage, USA, 1995. Project Number: AW95R003, B03987, AW/95R/03. Unpublished study prepared by Bayer CropScience LP. 90 p.	860.1380/Storage stability data



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

June 25, 2004

OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

BAYER CROPSCIENCE LP
2 T.W. ALEXANDER DRIVE
RESEARCH TRIANGLE PARK, NC 27709

Report of Analysis for Compliance with PR Notice 86-5

Thank you for your submittal of 06-JUN-04. Our staff has completed a preliminary analysis of the material. The results are provided as follows:

Your submittal was found to be in full compliance with the standards for submission of data contained in PR Notice 86-5. A copy of your bibliography is enclosed, annotated with Master Record ID's (MRIDs) assigned to each document submitted. Please use these numbers in all future references to these documents. Thank you for your cooperation. If you have any questions concerning this data submission, please raise them with the cognizant Product Manager, to whom the data have been released.



Bayer CropScience

June 18, 2004

Document Processing Desk (7504C)
Office of Pesticide Programs
U.S. Environmental Protection Agency
Room 266A, Crystal Mall 2
1921 Jefferson Davis Highway
Arlington, Virginia 22202

Attention: Ms. Stephanie Plummer, SRRD

Dear Ms. Plummer,

Re: **Thidiazuron; Active Ingredient No. 120301**
Submission in Support of the Reregistration of Thidiazuron

An electronic copy of the attached study reports will be submitted once MRID numbers are assigned to the studies. Therefore, only 2 paper copies of each report are enclosed in this submission.

Bayer CropScience
2 T.W. Alexander Drive
Research Triangle Park, NC 27709
Phone: 919 549-2000

Enclosed several residue chemistry studies in support of the reregistration of Thidiazuron. As the Agency requested during the Thidiazuron SMART meeting of June 2003, we are submitting these studies to upgrade the existing data base on file at the Agency for this product.

Please let me know if we can assist in any way during the reregistration process for Thidiazuron. You can reach me by phone at (919) 549-2718 or by e-mail at: Danielle.Laroche@bayercropscience.com.

Sincerely,

Danielle A. Larochelle
Registration Product Manager
February 13, 2004
Ms. Stephanie Plummer

Attachments: Document Nos. B003793, B003830, B003831, B003987

Corr. # daL056-04

Bayer CropScience



VOLUME 1 of 5

TRANSMITTAL DOCUMENT

Thidiazuron
Active Ingredient Number 120301

**Submission of Residue Chemistry Studies in Support of the
Reregistration Eligibility Decision for Thidiazuron**

Bayer CropScience
2 T.W. Alexander Drive
Research Triangle Park, NC 27709
Phone: 919 549-2000

Transmittal Date

June 18, 2004

Company Official:

Danielle A. Larochelle

Company Name:

Bayer CropScience

Company Contact:

Danielle A. Larochelle

Telephone

(919) 549-2718

Bayer CropScience
P.O. Box 12014
2 T.W. Alexander Drive
Research Triangle Park, NC 27709

Bayer CropScience
P.O. Box 12014
2 T.W. Alexander Drive
Research Triangle Park, NC 27709

BIBLIOGRAPHY OF SUBMITTED DATA

Volume 1 of 5: Transmittal Document

Volume 2 of 5: Norris, F. (2002) THIDIAZURON: Magnitude of Residues in Rotational Crops Following Cotton Treated with DROPP. EPA Guideline 860.1900. Aventis CropScience. Study No. 00AW25819. Document No. B003793. March 28, 2002. 248pp.

46298401

MRID #: _____

Volume 3 of 5: Perez, R. and S. Perez. (2002) Independent Laboratory Validation of Aventis Method AW/01/01 – Thidiazuron: Analytical Method for the Determination of Thidiazuron and its Metabolite Photo-Thidiazuron in Crop Matrices Using LC/MS/MS. EPA Guideline 860.1340. ADPEN Laboratories, Inc. Study No. 02AW27947. Document No. B003830. April 5, 2002. 74pp.

MRID #: **46298402**

Volume 4 of 5: Perez, R. and S. Perez. (2002) Independent Laboratory Validation of Aventis Method AW/02/01 – Thidiazuron: Analytical Method for the Determination of 1,2,3-thiadiazol-5-ylurea (AE F132345) Residues in Crop Matrices Using LC/MS/MS. EPA Guideline 860.1340. ADPEN Laboratories, Inc. Study No. 02AW27947U. Document No. B003831. April 5, 2002. 62pp.

MRID #: **46298403**

Volume 5 of 5: Williams, L.E. (2002) Stability of Thidiazuron and its Metabolites 4-Hydroxy Thidiazuron and Phenyl Urea in Milk and Beef Tissues During Frozen Storage, USA, 1995. EPA Guideline 860.1380. Bayer CropScience. Study No. AW-95R-03. Document No. B003987. November 18, 2002. 90pp.

46298404



13544

R122696

Chemical: Thidiazuron

**PC Code:
120301**

HED File Code: 61200 SRRD CDC

Memo Date: 11/8/2004

File ID: 00000000

Accession #: 412-06-0197

**HED Records Reference Center
7/25/2006**