

Data Evaluation Report on the anaerobic biotransformation of thidiazuron in a water-sediment system

PMRA Submission Number {.....}

EPA MRID Number 46119602

Data Requirement: PMRA Data Code:
EPA DP Barcode: D298740
OECD Data Point:
EPA Guideline: 162-3

Test material:

Common name: Thidiazuron.

Chemical name

IUPAC: 1-Phenyl-3-(1,2,3-thiadiazol-5-yl)urea.

CAS name: N-Phenyl-N'-1,2,3-thiadiazol-5-ylurea.

CAS No: 51707-55-2.

Synonyms: Dropp, AE B049537.

SMILES string: O=C(Nc1cccc1)Nc1cnns1.

Primary Reviewer: Dana Worcester
Dynamac Corporation

Signature:

Date:

QC Reviewer: Kathleen Ferguson
Dynamac Corporation

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Secondary Reviewer: Alex Clem
EPA

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Date:

12/7/04

Company Code:

Active Code:

Use Site Category:

EPA PC Code: 120301

CITATION: Liu, A., and P. Abbott. 2002. The degradation of [¹⁴C-thiadiazol]-thidiazuron in one soil under laboratory anaerobic conditions at 20°C. Unpublished study performed and submitted by Bayer CropScience, Durham, NC. Study Identification: 603AW. Experiment start date May 1, 2002, and completion date November 13, 2002 (p. 6). Final report issued November 13, 2002.



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EXECUTIVE SUMMARY:

The biotransformation of [thiadiazol-¹⁴C]1-phenyl-3-(1,2,3-thiadiazol-5-yl)urea (thidiazuron) was studied in water-sandy loam soil (deionized water; soil pH (water) 7.7, organic matter 5.4%) from Abington, England, for 120 days under anaerobic conditions in darkness at $20 \pm 1^\circ\text{C}$.

[¹⁴C]Thidiazuron was applied at a rate of 0.22 mg a.i./kg, which was estimated to be equivalent to approximately 196 g a.i./ha. The soil:water ratio was approximately 2:1 (50 g:25 mL); the soil was flooded to a depth of ca. 1 cm. The study was conducted in compliance with the USEPA GLP

This experiment was cited as being submitted under USEPA Subdivision N Guideline §162-4 for *aerobic* aquatic metabolism, but this must have been an error since the study was conducted under *anaerobic* conditions, as stated in the title. In fact, the soil:water system was anaerobic at the time of treatment and throughout the study (redox potentials ranged from -316 mV to -147 mV) and the samples were incubated under a nitrogen atmosphere. Therefore, as conducted, it was more typical of §162-3 (Anaerobic Aquatic Metabolism), except that a soil rather than a sediment was used. Thus, the study also had an aspect of §162-2 (Anaerobic Soil Metabolism), except that the system was not kept aerobic for an initial period and subsequently made anaerobic.

Even though the study is a hybrid among USEPA protocols and has other deficiencies, it has validity, and was adequate to demonstrate that thidiazuron, for our purposes, was effectively stable under anaerobic conditions in a water:soil system. A similar result was obtained from a submission (MRID 42666101) for §162-3 (Anaerobic Aquatic Metabolism), in which a sediment was used. This is sufficient evidence to confirm the persistence of thidiazuron under anaerobic conditions.

The test system consisted of Erlenmeyer-type flasks (14 cm x 7 cm i.d) containing treated flooded soil that were connected to individual flow-through volatile trapping systems. Nitrogen gas was continually forced through the samples, then through tubes of ethylene glycol and ethanolamine. Duplicate flasks were collected at 0, 7, 13, 29, 55, 90 and 120 days posttreatment. The samples were centrifuged, and the water layer was decanted and analyzed directly. Soil samples were extracted by shaking three times with acetonitrile:water (4:1, v:v) at ambient temperatures, then were Soxhlet extracted using acetonitrile. The water layers, soil extracts, extracted soil, and trapping solutions were analyzed for total radioactivity using LSC. The water (0 and 7 days only) and the soil extracts were analyzed for [¹⁴C]thidiazuron and its transformation products by reverse-phase HPLC. [¹⁴C]Compounds were identified by co-chromatography with unlabeled reference standards and LC/MS/MS analysis.

Conditions in the test system were reducing to strongly reducing throughout the study, with redox potentials ranging from -316 mV to -147 mV. The pH ranged from 8.54 to 9.57 on days 5-118. Daily temperatures averaged $20 \pm 1^\circ\text{C}$.

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Overall recoveries of radiolabeled material averaged $94.69 \pm 4.29\%$ (range 87.38-100.49%) of the applied. Recoveries decreased with time and were 92.09-92.61% at study termination.

[¹⁴C]Residues moved quickly from the water to the soil phase, so that [¹⁴C]residue concentrations in the water were <5% at and after 7 days. The ratio of [¹⁴C]residues in the soil and water was approximately 1:7 at day 0 and approximately 25:1 at and after 7 days.

In the total system, [¹⁴C]thidiazuron decreased from an average of 96.52% of the applied at 0 days posttreatment to 83.81% at 29 days and 76.72% at 120 days (study termination). In the water layer, [¹⁴C]thidiazuron averaged 86.50% of the applied at 0 days posttreatment and 4.01% at 7 days; [¹⁴C]residues in the water were not characterized after 7 days. In sediment extracts, [¹⁴C]thidiazuron increased from an average 10.02% of the applied at 0 days posttreatment to a maximum of 91.92% at 13 days, and ranged from 66.9-76.72% at 55-120 days.

In the water layer, no major or minor transformation products were isolated. In the soil, no major transformation products were isolated. The only minor transformation product that was identified, 1-(3-hydroxyphenyl)-3-(1,2,3-thidiazol-5-yl)urea (3-hydroxy-thidiazuron, AE F147706), averaged a maximum of 2.59% of the applied at 29 days posttreatment. In addition, a very polar compound (Rt 4 minutes) averaged a maximum of 4.34% of the applied at 90 days. The concentration of extractable [¹⁴C]residues in the soil increased to 77.04% of the applied at 120 days posttreatment, and nonextractable residues totaled 13.66-15.14% at 55-120 days. At 120 days posttreatment, [¹⁴C]residues trapped in the ethanolamine solutions (presumed to be CO₂) totaled 0.275% of the applied and [¹⁴C]residues trapped in the ethylene glycol solutions totaled 0.005%.

Based on first order linear regression analysis (Excel 2000), thidiazuron degraded with half-lives of 289 days in the entire system and 385 days in the soil. A half-life in water could not be calculated because data was only available for two sampling intervals; the observed DT50 was <7 days. The half-lives are of uncertain value because they are extrapolated well beyond the duration of the study and assume that degradation continues to follow a first order model.

A transformation pathway was proposed by the study author. Thidiazuron degrades to 3-hydroxy-thidiazuron via hydroxylation, and 3-hydroxy-thidiazuron further degrades into polar compounds.

Results Synopsis:

Test system used: water-sandy loam soil.

Half-lives:

Water: <7 days (observed DT50).

Soil: (13 to 120-day data): 385.1 days ($r^2 = 0.2876$).

Entire system (0 to 120-day data): 288.8 days ($r^2 = 0.5198$).

Major transformation products:

None.

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Minor transformation products:

1-(3-Hydroxyphenyl)-3-(1,2,3-thiazol-5-yl)urea (3-hydroxy-thidiazuron, AE F147706).
CO₂.

Study Acceptability: This study is classified as **supplemental**. It is scientifically valid but cannot be used to meet the requirement for either an aerobic or anaerobic aquatic metabolism study. It fails to meet the requirement for an aerobic aquatic metabolism study (§162-4) because the soil:water system was anaerobic at the time of treatment and throughout the study (redox potentials ranged from -316 mV to -147 mV) and the samples were incubated under a nitrogen atmosphere. It fails to meet the requirement for an anaerobic aquatic metabolism study (§162-3) because a soil rather than a sediment was used and the study was terminated after only 120 days, when 76-77% of the applied thidiazuron remained undegraded. However, based on this study and evidence from other studies, it is reasonably clear for risk assessment purposes that thidiazuron is persistent under anaerobic conditions, and that there would be little value added by the submission of additional anaerobic data.

I. MATERIALS AND METHODS

GUIDELINE FOLLOWED: This study was conducted in accordance with SETAC- Europe Procedures for Assessing the Environmental Fate and Ecotoxicology of Pesticides, European Economic Community Annex 1 Commission Directive 95/36/EC (July 1995), OECD 91/414/EEC (pp. 1, 15, 27). USEPA Subdivision N Guideline §162-4 (Aerobic Aquatic Metabolism) was also cited, but this may have been an error since the study protocol (nitrogen atmosphere) was more typical of §162-3 (Anaerobic Aquatic Metabolism). The following significant deviation from Subdivision N Guideline 162-4 was noted:

The soil:water system was anaerobic at the time of treatment and throughout the study (redox potentials ranged from -316 mV to -147 mV) and the samples were incubated under a nitrogen atmosphere. This does not affect the validity of the study.

The following significant deviation from Subdivision N Guideline §162-3 (Anaerobic Aquatic Metabolism) was noted:

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The study was terminated after 120 days, at which point 76.18-77.26% of the applied thidiazuron remained undegraded. This does not affect the validity of the study.

COMPLIANCE: This study was conducted in compliance with USEPA Good Laboratory Practice Standards (40 CFR Part 60; p. 3). Signed and dated Data Confidentiality, GLP, Quality Assurance, and Certification of Authenticity statements were provided (pp. 2-5).

A. MATERIALS:

1. Test Material: [Thidiazol-¹⁴C]thidiazuron (p. 11).

Chemical Structure: See DER Attachment.

Description: Not reported.

Purity: Radiochemical purity: 100% (HPLC, Figure 2, p. 39).
 Batch No.: GAR 2026/6 (p. 15).
 Analytical purity: >97%.
 Specific activity: 83.8 µCi/mg.
 Location of radiolabel: Thidiazol ring.

Storage conditions of test chemical: Not reported.

Table 1: Physico-chemical properties of thidiazuron.

Parameter	Values	Comments
Molecular weight:	Not reported.	
Molecular formula:	Not reported.	
Water solubility:	Not reported.	
Vapor pressure:	Not reported.	
UV absorption:	Not reported.	
pK _a :	Not reported.	
K _{ow} /log K _{ow} :	Not reported.	
Stability of compound at room temperature:	Not reported.	

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2. Water-soil collection, storage and properties:

Table 2: Description of water-soil collection and storage.

Description		Details
Geographic location:		Soil was obtained from Abington, Cambridgeshire, England The water was deionized water.
Collection date:		March 1, 2002.
Pesticide use history at the collection site:		Not reported.
Collection procedures:	Water:	Not applicable.
	Soil:	Not reported.
Sampling depth:	Water:	Not applicable.
	Soil:	Not reported.
Storage conditions:		Soil was stored at <i>ca.</i> 4-10°C.
Storage length:		Approximately 1 month.
Preparation:	Water:	Deionized water was used.
	Soil:	Sieved (2.0-mm).

Data obtained from pp. 19-20 of the study report.

Table 3: Properties of the water.*

Property	Details
Temperature (°C):	Not reported.
pH:	Not reported.
Redox potential:	Not reported.
Oxygen concentration:	Not reported.
Dissolved organic carbon (DOC):	Not reported.
Total organic carbon (TOC):	Not reported.
Hardness:	Not reported.
Electrical conductivity (µmhos):	Not reported.
Biomass (mg microbial C/100 g or CFU or other):	Not reported.

* Deionized water was used in the study.

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Table 4: Properties of the soil.

Property		Details
Textural classification:		Sandy loam.
% Sand:		73
% Silt:		20
% Clay:		7
pH:	Water (1:1)	7.7
	CaCl ₂	7.2
Organic carbon (%):		3.14
Organic matter (%):		5.4
CEC (meq/100 g soil):		19.4
Redox potential:		Not reported.
% Moisture at:	0 bar	65.4
	1/10 bar	23.2
	1/3 bar	17.9
	15 bar	14.3
Bulk density (disturbed, g/cm ³):		1.20.
Biomass (µg/g):	Bacterial	295.5
	Fungal	91.0
Taxonomic classification:		Not reported.
Other:		Not reported.

Data obtained from Tables 1-2, pp. 28-29 of the study report. The particle size distribution was based on the USDA Textural Classification system.

B. EXPERIMENTAL CONDITIONS:

1. **Preliminary experiments:** No preliminary experiments were reported.

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2. Experimental conditions:

Table 5: Study design.

Parameter		Details
Duration of the test:		120 days.
Water: Filtered/unfiltered water: Type and size of filter used, if any:		Deionized water. Not applicable.
Amount of soil and water per treatment:	Water:	ca. 25 mL. The soil was flooded with water to a depth of ca. 1 cm.
	Soil:	50 g (dry wt. equivalent).
Water/soil ratio:		1:2 (25 mL water:50 g soil).
Nominal application rate (mg a.i./L):		0.2 mg a.i./kg.
Actual application rate (mg a.i./L):		0.223 mg a.i./kg, equivalent to 196 g a.i./ha assuming a soil depth of 7.5 cm.
Control conditions, if used:		No sterile controls were used.
No. of Replications:	Controls, if used:	No sterile controls were used.
	Treatments:	Duplicate flasks were collected at each sampling interval.
Test apparatus (Type/material/volume):		Erlenmeyer type flasks (14 cm x 7.2 bottom i.d.) containing treated flooded soil (25 mL water, 50 g soil) were attached to a flow-through volatile trapping system (nitrogen gas) and incubated in the dark at a controlled temperature.
Details of traps for CO ₂ and organic volatiles, if any:		Humidified nitrogen gas was continually forced through the sample flasks (6.1 mL/minute), then through single tubes of ethylene glycol and ethanolamine (30 mL/trap) trapping solutions.
If no traps were used, is the system closed/open?		Volatiles traps were used.
Identity and concentration of co-solvent:		Acetonitrile, ca. 0.1% by volume.

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Parameter		Details
Test material application:	Volume of test solution used/treatment:	216 μ L/50 g soil.
	Application method:	The stock solution was applied to the water surface using a Gilson pipette.
Any indication of the test material adsorbing to the walls of the test apparatus?		None.
Biomass of controls:		Controls were not used.
Biomass of treated soil (μ g/g soil dry wt):	initial	295.5
	Final	91.0
Experimental conditions:	Temperature ($^{\circ}$ C):	$20 \pm 1^{\circ}$ C
	Continuous darkness:	Yes.
Other details, if any:		None.

Data obtained from pp. 20, 21, 23; Table 2, p. 29; Figure 1, p. 38; Appendix 2, pp. 54-55; Appendix 4, p. 70; and Appendix 5, p. 74 of the study report.

3. Aerobic/Anaerobic conditions: Samples were maintained under a steady stream of nitrogen during the study. Redox potentials in the surface water were -177 to -162 one day prior to treatment and ranged from -316 mV to -147 mV throughout the study (reducing to strongly reducing; Table 3, p. 30).

4. Supplementary experiments: No supplementary experiments were reported.

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5. Sampling:

Table 6: Sampling details.

Criteria	Details
Sampling intervals:	0, 7, 13, 29, 55, 90 and 120 days.
Sampling method:	Duplicate flasks were collected at each interval.
Method of collection of CO ₂ and volatile organic compounds:	The trapping solutions were collected at each sampling interval.
Sampling intervals/times for: Sterility check, if sterile controls are used:	Sterile controls were not used.
Redox potential/dissolved oxygen/pH of water layer and redox potential of soil:	The redox potential and pH of the water layer were measured at -1, 5, 12, 29, 54, 89 and 118 days. The redox potential of the soil was not measured.
Sample storage before analysis:	Samples were extracted on the day of collection. The water, soil extracts, and volatile trapping solutions were analyzed by LSC on the day of collection, except for the 7 day sample which was analyzed the day after collection. HPLC analysis was conducted within 30 days. Samples were stored at <i>ca.</i> -10°C until HPLC or MS analysis.
Other observations, if any:	None.

Data obtained from pp. 23, 25 and Table 3, p. 30 of the study report.

C. ANALYTICAL METHODS:

Separation of the soil and water: The water layer was “carefully” decanted into a polypropylene centrifuge tube and centrifuged (p. 22).

Extraction/clean up/concentration methods: The water layer was analyzed directly by LSC and HPLC, without modification or manipulation.

The soil was transferred to centrifuge bottles and extracted three times by shaking on a wrist-action shaker with acetonitrile:water (4:1, v:v, 50-75 mL; p. 22) for 20 minutes/extraction at ambient temperatures. After each extraction, the samples were centrifuged and the supernatant decanted. The extracts were pooled and aliquots were analyzed using LSC. Samples collected after 0 days posttreatment were then Soxhlet extracted using acetonitrile. Following extraction, the samples were centrifuged and the supernatant decanted, and aliquots were analyzed using LSC. The pooled ambient extracts and the Soxhelt extract were separately were concentrated using rotary evaporation, and aliquots of the concentrate were analyzed by LSC and HPLC. Recoveries following concentration averaged 91.22% (Appendix 7.6, p. 84).

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Nonextractable residue determination: The extracted soil was air-dried, homogenized, and ground, and portions were analyzed for total radioactivity by LSC following combustion (p. 22).

In order to characterize the unextracted residues in the 55-day sample, a portion of the extracted soil (*ca.* 40 g) was shaken for *ca.* 1 hour with 0.25M HCl (150 mL), then centrifuged (p. 22, Figure 10, p. 47). The acidic extract was analyzed using LSC ("acidic extract"). The acid-extracted soil pellet was shaken for *ca.* 24 hours with 0.5M NaOH (150 mL), then centrifuged. The supernatant was acidified (*ca.* pH 2) with concentrated HCl, and the supernatant (fulvic acid) and resulting precipitate (humic acid) were analyzed using LSC. The soil pellet (humins) was dried and analyzed by LSC following combustion.

Volatile residue determination: Triplicate aliquots (1 mL) of the ethylene glycol and ethanolamine solutions were analyzed by LSC (p. 21).

Total ¹⁴C measurement: Total [¹⁴C]residues were determined by summing the concentrations of residues measured in the water layer, soil extracts, extracted soil, and volatile trapping solutions (Appendix 4, pp. 71-72).

Derivatization method, if used: A derivatization method was not employed.

Identification and quantification of parent compound: The samples were analyzed using reverse-phase HPLC under the following conditions (p. 18): PerkinElmer RP-18 Guard column (C18 cartridge); Polaris C8-Ether column (4.6 mm x 250 mm, 5 μ), a gradient mobile phase consisting of (A) water (B) acetonitrile [percent A:B (v:v) 0 minutes, 90:10; 7 minutes, 90:10; 12 minutes, 80:20; 16 minutes, 70:30; 20 minutes, 60:40; 35 minutes, 30:70; 38 minutes, 0:100; 45 minutes, 0:100; 46 minutes, 90:10; 60 minutes, 90:10]; and a flow rate of 1 mL/minute, with UV (254 nm) and radioactive flow detection. Thidiazuron was identified by comparison with an unlabeled reference standard (purity not reported, *Rt* = 28.31; p. 16, Figure 3, p. 40).

The identification of thidiazuron in a 120 day posttreatment sample was confirmed using LC/MS in positive electrospray mode (Appendix 3, pp. 60-61).

Identification and quantification of transformation products: Transformation products were separated, quantified and identified by reverse-phase HPLC as described for the parent (p. 18). The reference standards used were thidiazolurea (AE F132345, purity not reported, *Rt* = 7.21 minutes), 3-hydroxy-thidiazuron (AE F147706, purity not reported, *Rt* = 25.41 minutes), and 4-hydroxy-thidiazuron (AE F132346; purity not reported, *Rt* = 24.16 minutes; p. 16, Figure 3, p. 40). An attempt to confirm the identification of 3-hydroxy-thidiazuron was unsuccessful, possibly because of the very low concentration of this compound in the sample (Appendix 3, p. 61).

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Detection limits (LOD, LOQ) for the parent compound: The LSC Limit of Detection was any concentration greater than background (30 dpm, 1.6 ng of thidiazuron) and the HPLC LOD was 0.002% of the applied (p. 23, Table 7, pp. 35-37). The LSC Limit of Quantification was twice background (60 dpm; 0.16 ng); the HPLC LOQ was 300 dpm.

Detection limits (LOD, LOQ) for transformation products: The LOD and LOQ were the same as described for the parent.

II. RESULTS AND DISCUSSION:

A. TEST CONDITIONS: The test systems were reducing to strongly reducing throughout the study, ranging from -316 to -147 mV throughout the study (Table 3, p. 30). The pH gradually increased during the study from 7.99-8.59 at one day prior to treatment to 9.02-9.57 at 89 and 118 days. The incubation temperature was maintained at $20 \pm 1^\circ\text{C}$ (Appendix 5, p. 74).

B. MATERIAL BALANCE: Overall recoveries of radiolabeled material averaged $94.69 \pm 4.29\%$ (range 87.38-100.49%; Table 6, p. 33). Recoveries totaled 94.04-100.49% of the applied through 29 days posttreatment, were 87.38-89.75% at 55 days, and increased to 92.09-92.61% at 120 days. [^{14}C]Residues moved quickly from the water to the soil phases. The ratio of [^{14}C]residues in the water and water decreased from 7:1 immediately posttreatment to $\geq 1:25$ at and after 7 days.

Table 7: Biotransformation of [¹⁴C]thiazuron, expressed as percentage of applied radioactivity, in water/sandy loam soil under anaerobic conditions (mean ± SD, n = 2).*

Compound	Sampling times (days)								
	0	7	13	29	55	90	120		
Thiazuron	Water	86.50 ± 3.41	4.01 ± 0.64	NA	NA	NA	NA	NA	NA
	Soil ¹	10.02 ± 2.83	88.70 ± 1.64	91.92 ± 0.42	83.81 ± 1.68	66.90 ± 2.21	69.80 ± 11.57	76.72 ± 0.76	
	System ²	96.52 ± 0.58	92.71 ± 1.00	91.92 ± 0.42	83.81 ± 1.68	66.90 ± 2.21	69.80 ± 11.57	76.72 ± 0.76	
3-OH thiazuron	Water	ND	ND	NA	NA	NA	NA	NA	NA
	Soil ¹	0.23 ± 0.33	0.58 ± 0.81	1.22 ± 0.19	2.59 ± 0.70	1.92 ± 1.63	2.27 ± 0.87	0.32 ± 0.45	
	System ²	0.23 ± 0.33	0.58 ± 0.81	1.22 ± 0.19	2.59 ± 0.70	1.92 ± 1.63	2.27 ± 0.87	0.32 ± 0.45	
Unknown (Rt 4 minutes)	Water	ND	ND	NA	NA	NA	NA	NA	NA
	Soil	ND	ND	ND	ND	3.25 ± 2.88	4.34 ± 4.62	ND	
Total extractable soil residues ³		10.25 ± 2.50	89.28 ± 0.83	93.14 ± 0.62	86.40 ± 0.98	72.06 ± 0.96	76.41 ± 6.08	77.04 ± 1.22	
Ethanolamine (presumed ¹⁴ CO ₂)		NA	0.060 ± 0.002	0.096 ± 0.002	0.129 ± 0.004	0.199 ± 0.001	0.283 ± 0.008	0.275 ± 0.064	
Ethylene glycol		NA	0.009 ± 0.004	0.006 ± 0.001	0.000 ± 0.000	0.005 ± 0.001	0.003 ± 0.003	0.005 ± 0.002	
Nonextractable soil residues		2.09 ± 0.47	3.32 ± 0.25	4.30 ± 0.03	7.24 ± 0.63	15.14 ± 0.74	13.66 ± 1.23	14.15 ± 0.88	
	Water	86.50 ± 3.41	4.01 ± 0.64	2.40 ± 0.14	1.49 ± 0.10	1.17 ± 0.03	0.83 ± 0.01	0.89 ± 0.02	
	Soil	12.34 ± 2.97	92.60 ± 0.58	97.44 ± 0.64	93.63 ± 1.61	87.20 ± 1.70	90.07 ± 4.85	91.19 ± 0.33	
System	98.84 ± 0.44	96.68 ± 0.06	99.94 ± 0.78	95.25 ± 1.70	88.57 ± 1.68	91.18 ± 4.86	92.35 ± 0.37		

* Means and standard deviations calculated by the reviewer using data obtained from Table 6, p. 33 and Table 7, pp. 34-37 of the study report.

1 Soil; reviewer-calculated by summing the ambient and Soxhlet extractions.

2 Entire system; reviewer-calculated by summing the reported concentrations in the water and soil.

3 Total extractable; reviewer-calculated by summing ambient and Soxhlet extractions.

ND = Not detected; <0.002% of the applied.

NA = Not analyzed. The water phase was not analyzed by HPLC after 7 days posttreatment.

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C. TRANSFORMATION OF PARENT COMPOUND: In the total system, [¹⁴C]thidiazuron decreased from an average 96.52% of the applied at 0 days posttreatment to 83.81% at 29 days and was 76.72% at 120 days (reviewer calculated from Table 7, pp. 34-37). In the water layer, [¹⁴C]thidiazuron averaged 86.50% of the applied at 0 days posttreatment and 4.01% at 7 days; [¹⁴C] residues in the water were not characterized after 7 days. In sediment extracts, [¹⁴C]thidiazuron increased from an average 10.02% of the applied at 0 days posttreatment to a maximum of 91.92% at 13 days and was 76.72% at 120 days.

HALF-LIFE/DT50: Based on first order linear regression analysis (Excel 2000) and using data for individual samples, thidiazuron degraded with half-lives of 289 days in the entire soil/water system and 385 days in the soil (reviewer-calculated from Table 7, pp. 34-37). The half-lives are of uncertain value because they are extrapolated well beyond the duration of the study and assume that degradation continues to follow a first order model. A half-life in water could not be calculated because data was only available for two sampling intervals; the observed DT 50 was <7 days. The total system value calculated by the reviewer is longer than the 185 days calculated by the study author using Excel 97 with a curve fitting add-in (Appendix 6, pp. 76-77).

Half-lives/DT50s

Medium	First-order Linear			DT50 (days)	DT90 (days)
	Half-life (days)	Regression equation	r ²		
Water	Could not be calculated.			NR	NR
Soil	13- to 120-day data: 385.08	y = -0.0018x + 4.4551	0.2876	NR	NR
Total system	1- to 120-day data: 288.81	y = -0.0024x + 4.5141	0.5198	185.57	616.44

Half-life values were calculated by the reviewer using data obtained from Table 7, pp. 34-37 of the study report. DT50 and DT90 values were calculated by the study author and are presented in Figure 12, p. 49, and Appendix 6, p. 77, of the study report.

NR = Not reported. The study author determined DT50 and DT90 values only for the entire system.

TRANSFORMATION PRODUCTS: No major transformation products were isolated from either the water or soil (Table 7, pp. 34-37). No minor transformation products were isolated from the water at 0 and 7 days posttreatment; at and after 13 days, water residues (0.82-2.5% of the applied) were not characterized (Table 6, p. 33). In the soil, the only minor transformation product identified was 1-(3-hydroxyphenyl)-3-(1,2,3-thiazol-5-yl)urea (3-hydroxy-thidiazuron, AE F147706), which was primarily associated with the ambient extracts and averaged a maximum of 2.59% of the applied at 29 days posttreatment. Also, one HPLC peak (Rt 4 minutes, "very polar") associated with the Soxhlet extract averaged a maximum of 4.34% of the applied at 90 days.

NONEXTRACTABLE AND EXTRACTABLE RESIDUES: The concentration of extractable [¹⁴C]residues in the soil increased from an average 10.25% of the applied at 0 days posttreatment to 77.04% of the applied at 120 days posttreatment (Table 6, p. 33). Nonextractable [¹⁴C]residues

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were a maximum averaged of 15.14% of the applied at 55 days posttreatment and were 14.15% at 120 days. At 55 days posttreatment, the distribution of the nonextractable residues was 10.58% of the applied as humin, 1.37% as humic acid, and 2.56% as fulvic acid (Figure 10, p. 47).

VOLATILIZATION: At 120 days posttreatment, [¹⁴C]residues trapped in the ethanolamine solutions (presumed to be CO₂) totaled 0.275% of the applied and [¹⁴C]residues trapped in the ethylene glycol solutions totaled 0.005% (Table 6, p. 33).

TRANSFORMATION PATHWAY: A transformation pathway was proposed by the study author (p. 26; Figure 11, p. 48). Thidiazuron degrades to 3-hydroxy-thidiazuron via hydroxylation, and 3-hydroxy-thidiazuron further degrades into an unidentified polar compound.

Table 8: Chemical names for transformation products of thidiazuron.

Applicant's code	CAS Number	Chemical Name	Chemical formula	Molecular weight	SMILES string
3-Hydroxy-thidiazuron, AE F147706		1-(3-Hydroxyphenyl)-3-(1,2,3-thiazol-5-yl)urea			

Data obtained from p. 16 of the study report.

D. SUPPLEMENTARY EXPERIMENT-RESULTS: No supplementary experiments were reported.

III. STUDY DEFICIENCIES: This study was submitted under USEPA Subdivision N Guideline §162-4 (Aerobic Aquatic Metabolism). However, this study cannot be used as an aerobic aquatic metabolism study because the soil:water environment was anaerobic at the time of treatment and throughout the study, with redox potentials of -177 to -162 one day prior to treatment and ranging from -316 mV to -147 mV throughout the study (reducing to strongly reducing; Table 3, p. 30). In addition, the samples were incubated under a flow-through nitrogen atmosphere.

The study does not meet Subdivision N Guideline §162-3 (Anaerobic Aquatic Metabolism) data requirement because the study was terminated at 120 days posttreatment. At this time, 76.72% of the applied thidiazuron remained undegraded in the water-sandy loam system. Therefore, the half-lives for thidiazuron in the were extrapolated well beyond the final sampling interval. The accuracy of the half-lives are therefore highly uncertain because they are based on the assumption that the pattern of dissipation remains linear throughout the process.

IV. REVIEWER'S COMMENTS:

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1. The overall recovery of radioactivity was low in both samples at 55 days (87.38 and 89.75% of the applied) and in one sample (87.74%) at 90 days; recoveries increased to 92.09 and 92.61% at 120 days (Table 6, p. 33). The low recoveries correspond to the low concentrations of thidiazuron (61.62-68.46% of the applied at 55-90 days) in the system (Table 7, p. 34). In the one sample at 90 days and the two samples at 120 days where recoveries are >90%, the concentrations of thidiazuron are 76.18-77.98% of the applied.
2. Up to 15.14% of the applied radioactivity was not extracted from the soils (Table 6, p. 33). However, it is unlikely that harsher extraction procedures would have resulted in significant amounts of identifiable residues.
3. The study author's assumed but did not confirm the identification of [¹⁴C]residues in the ethanolamine trapping solution as CO₂. Since the concentration of residues in the trapping solutions was <1% of the applied, identifications were not required.
4. The volume of water used to flood the soil was not reported in the body of the report; the study author's only reported that the soil was flooded to a depth of 1 cm (p. 20). In one of the sample calculations (p. 70), the study author's report the total volume of the surface water as 25 mL. Logically, it would seem that >25 mL of water would be required to cover 50 g of soil; however, this value (25 mL) is also in line with the acetonitrile cosolvent concentration of approximately 0.1% by volume reported by the study author (p. 21) and was therefore assumed by the reviewer to be accurate. It would have been preferred if the study author had clearly stated in the text the volume of water used to flood the soil.
5. The test soil used in the study was from the UK. The soil was fully characterized according to the USDA Soil Textural Classification system, but was not formally compared to a soil from the US (Table 1, p. 28). The physical properties of the soil appear to be typical of US soils.
6. Physico-chemical properties of thidiazuron such as water solubility, vapor pressure, and UV absorption were not reported.
7. The daily incubation temperature was reported as 20 ± 1°C (Appendix 5, p. 74). More detailed information was not provided. The minimum, maximum, and mean temperatures should have been included in the study report.
8. The source history and collection procedures for the soil prior to use were not reported. Also, the water was reported as deionized water, no other details were reported.

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9. The study author reported that the nominal application rate chosen for this study was based on a single field application rate of 196 g a.i./ha based on a soil bulk density of 1.3 g/cm³ and a soil depth of 7.5 cm (p. 20).
10. According to N. Wolfe, *et al.* (see reference below), redox potentials in the range of +400 to +800 mV are considered strongly oxidizing, +200 to +400 mV moderately oxidizing, -50 to +200 mV moderately reducing, -200 to -50 mV reducing, and -400 to -200 mV strongly reducing.

V. REFERENCES:

1. U.S. Environmental Protection Agency. 1982. Pesticide Assessment Guidelines, Subdivision N, Chemistry: Environmental Fate, Section 162-3, Anaerobic Aquatic Metabolism Studies. Office of Pesticide and Toxic Substances, Washington, DC. EPA 540/9-82-021.
2. U.S. Environmental Protection Agency. 1989. FIFRA Accelerated Reregistration, Phase 3 Technical Guidance. Office of the Prevention, Pesticides, and Toxic Substances, Washington, DC. EPA 540/09-90-078.
3. U.S. Environmental Protection Agency. 1993. Pesticide Registration Rejection Rate Analysis - Environmental Fate. Office of the Prevention, Pesticides, and Toxic Substances, Washington, DC. EPA 738-R-93-010.
4. Wolfe, N., *et al.* 1990. Abiotic transformations in water, sediments and soil. *In Pesticides in the Soil Environment*, Soil Science Society of America, pp. 103-110.

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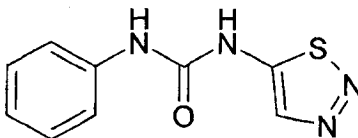
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**ATTACHMENT
Chemical Structure of Thidiazuron**

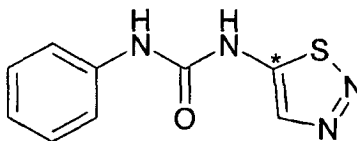
Thidiazuron (AE B049537; DROPP; SN 49537)

IUPAC name: 1-Phenyl-3-(1,2,3-thiadiazol-5-yl)urea.
CAS name: *N*-Phenyl-*N'*-1,2,3-thiadiazol-5-ylurea.
CAS No: 51707-55-2 (330-54-1 also found).
SMILES string: O=C(Nc1ccccc1)Nc1cnns1

Unlabeled



[Thiadiazol-¹⁴C]thidiazuron or [¹⁴C]Thidiazuron



* Position of the radiolabel