

US EPA ARCHIVE DOCUMENT

Data Evaluation Report on the aerobic biotransformation of BAS 510 F in water-sediment system

PMRA Submission Number {.....}

EPA MRID Number 45405214

Data Requirement: PMRA Data Code:
EPA DP Barcode: D278387
OECD Data Point:
EPA Guideline: 162-4

Test material:

Common name: BAS 510 F.

Chemical name

IUPAC: 2-Chloro-*N*-(4-chlorobiphenyl-2-yl)-nicotinamide.
CAS name: 2-Chloro-*N*-(4-chloro[1,1-biphenyl]-2-yl)-3-pyridinecarboxamide.
CAS No: 188425-85-6.
synonyms: 2-Chloro-*N*-(4'-chlorobiphenyl-2-yl)-nicotinamide.
Nicobifen.
~~BAS 516 02 F.~~

SMILES string:

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EXECUTIVE SUMMARY:

The aerobic biotransformation of [diphenyl-U-¹⁴C]- and [pyridine-3-¹⁴C]-labeled 2-chloro-*N*-(4'-chlorobiphenyl-2-yl)-nicotinamide (BAS 510 F) was studied in a water/German loamy sand sediment system (water pH 8.50, organic carbon 12.0-14.4%; sediment pH in CaCl₂ 6.8, organic carbon 0.8%) from a pond and a water/German loam sediment system (water pH 8.10, organic carbon 8.6-15.2%; sediment pH in CaCl₂ 7.5, organic carbon 4.1-4.3%) from a pond-like area adjacent to a river, both located in Germany, for 100 days in darkness at 20 ± 2°C. Both labels were applied at the rate of 0.24 mg a.i./L water. Sediment:water ratios used were 1:1.5 (190 g wet wt. sediment:290 mL water) for the water/loamy sand sediment system and 1:2.1 (140 g wet wt. sediment:290 mL water) for the water/loam sediment system. This study was conducted in accordance with USEPA Subdivision N Guideline §162-4 (1982), BBA Guidelines (Part IV, 5-1), and SETAC Europe Procedures for Assessing the Environmental Fate and Ecotoxicity of Pesticides (Part 8.2) and in compliance with compliance with Federal Republic of Germany GLP Regulations (Chemikaliengesetz, Appendix 1-19a, 1994) and met the requirements of USEPA GLP Standards (40 CFR, Part 160, 1989). The test system consisted of sealed flasks containing treated water/sediment incubated in darkness in a temperature-controlled chamber; each flask was connected to a flow-through system for the continuous collection of CO₂ and volatile organics. Water and sediment were pre-incubated for 22 days, then, following treatment, water and sediment samples were analyzed after 0, 1, 2, 7, 14, 29, 59 and 100 days of incubation. Water samples were analyzed directly. Sediment samples were sequentially extracted with acetonitrile:water (1:1, v:v) and acetonitrile. Water and sediment extracts were analyzed by reverse-phase HPLC; procedures for identification of parent [¹⁴C]BAS 510 F were not specified. HPLC results were confirmed using normal-phase one-dimensional HPTLC.

Aerobic conditions were maintained in the water layer of the water/sediment systems, but the sediment layer remained anaerobic throughout the study. During the 100-day study, the overall material balance (both labels and systems) averaged ($n = 32$) 97.1 ± 1.2% (range 92.5-100.2%) of the applied radioactivity. Total [¹⁴C]residues of [¹⁴C]BAS 510 F (both labels) gradually partitioned from the water layer to the loamy sand sediment with distribution ratios (water:sediment) of *ca.* 4:1 at day 1, 1:1 at 7-14 days, 1:2 at 29 days and 1:5 at 100 days, but partitioned from the water layer to the loam sediment at a faster rate with distribution ratios of *ca.* 2:1 at day 1, 1:1 at 2 days, 1:2 at 7 days, 1:7 at 29 days and 1:15 at 100 days.

In water/loamy sand sediment systems, [¹⁴C]BAS 510 F (both labels) in the total system gradually decreased from 96.1-97.0% of the applied at day 0 to 84.1-86.0% at 100 days. [¹⁴C]BAS 510 F in the water layer decreased 96.1-97.0% at day 0 to 51.7-55.2% at 7 days, 30.9-31.1% at 29 days and was 17.3-17.4% at 100 days, while in the sediment [¹⁴C]BAS 510 F increased from 19.8-20.1% at 1 day to 48.1-53.2% at 14 days and was 66.8-68.6% at 100 days.

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In sterile water/loamy sand sediment at 101 days, [¹⁴C]BAS 510 F (both labels) comprised 14.3-15.0% of the applied in the water layer and 70.6-72.4% in the sediment.

In water/loam sediment systems, [¹⁴C]BAS 510 F (both labels) in the total system decreased from 95.9-96.9% of the applied radioactivity at day 0 to 85.7-86.3% at 100 days (minimum 83.9% at 59 days). [¹⁴C]BAS 510 F in the water layer decreased 95.9-96.9% at day 0 to 50.2-54.2% at 2 days, 21.4-23.0% at 14 days 12.1% (both labels) at 29 days and was 6.1-6.2% at 100 days, while in the sediment [¹⁴C]BAS 510 F increased from 31.7-32.4% at 1 day to 60.4-61.5% at 7 days and was 76.5-80.2% at 29-100 days. In sterile water/loam sediment systems at 101 days, [¹⁴C]BAS 510 F (both labels) comprised 7.0-7.6% of the applied in the water layer and 83.1-84.8% in the sediment.

For both labels and systems, no major transformation products of [¹⁴C]BAS 510 F were detected in the water layers or sediment extracts. Up to four minor unidentified [¹⁴C]compounds were detected (each at ≤0.4% of the applied) in the water layers of both systems, but were not detected in sediment extracts.

For both labels, extractable [¹⁴C]residues increased from 0.6-0.7% of the applied in both sediments at day 0 to 66.8-68.6% at 100 days in the loamy sand sediment and to 76.5-80.2% at 29-100 days in the loam sediment, while non-extractable [¹⁴C]residues in both sediments increased from ≤0.1% at day 0 to 10.3-13.4% at 100 days. In extracted 59- and 100-day sediment samples (both labels and sediments), 1.6-6.3% of the applied was associated with fulvic acids, 2.2-3.9% with humic acids and 0.6-2.3% with humins. Volatilization of ¹⁴CO₂ and organic volatiles was not significant for either label, totaling ≤0.7% of the applied at study termination in both systems.

DT₅₀ and DT₉₀ values for the dissipation of [¹⁴C]BAS 510 F (results from both labels combined for calculation) from the water layer to the loamy sand sediment were 9 and 133 days, respectively, and from the water layer to the loam sediment were 3 and 43 days, respectively. Half-life values for the dissipation of [¹⁴C]BAS 510 F (both labels calculated individually) from the water layer to the sediment, based on first-order kinetics and linear regression over the study duration (0-100 days), were 44-46 days in water/loamy sand sediment systems and 27-28 days in water/loam sediment systems. Degradation of both labels appeared bi-phasic with initial (0-14 days) and secondary (14-100 days) half-life values for the dissipation of [¹⁴C]BAS 510 F from the water to loamy sand sediment of 12-14 and 66-76 days, respectively. Initial (0-7 days) and secondary (7-100 days) half-life values for the dissipation of [¹⁴C]BAS 510 F from the water to loam sediment were 5 and 41 days, respectively.

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BAS 510 F was found to be stable in two aerobic water/sediment systems with dissipation of parent compound from the water layer to the sediment. Half-lives were not calculated for the total systems since degradation of the parent was negligible in the water/sediment systems.

Results Synopsis:

Test system: Water/loamy sand sediment from pond in Germany.
Water/loam sediment from pond-like area off Rhine river in Germany.

Dissipation of parent BAS 510 F *from water to loamy sand sediment:*

Half-life values:

[Diphenyl-U-¹⁴C]BAS 510 F:

- overall (0-100 days): 45.5 days ($r^2 = 0.836$).
- initial (0-14 days): 12.4 days ($r^2 = 0.931$).
- secondary (14-100 days): 75.6 days ($r^2 = 0.980$).

[Pyridine-3-¹⁴C]BAS 510 F:

- overall (0-100 days): 43.5 days ($r^2 = 0.847$).
- initial (0-14 days): 13.8 days ($r^2 = 0.871$).
- secondary (14-100 days): 66.1 days ($r^2 = 0.911$).

DT₅₀ (both labels): 9 days

DT₉₀ (both labels): 133 days

Dissipation of parent BAS 510 F *from water to loam sediment:*

Half-life values:

[Diphenyl-U-¹⁴C]BAS 510 F:

- overall (0-100 days): 27.4 days ($r^2 = 0.777$).
- initial (0-7 days): 5.0 days ($r^2 = 0.902$).
- secondary (7-100 days): 41.1 days ($r^2 = 0.841$).

[Pyridine-3-¹⁴C]BAS 510 F:

- overall (0-100 days): 28.1 days ($r^2 = 0.789$).
- initial (0-7 days): 5.4 days ($r^2 = 0.845$).
- secondary (7-100 days): 40.9 days ($r^2 = 0.850$).

DT₅₀ (both labels): 3 days.

DT₉₀ (both labels): 43 days.

Major transformation products: None.

Minor transformation products: Four unidentified [¹⁴C]compounds.

Study Acceptability: This study is classified as supplemental and cannot be used to satisfy the guideline requirement for an aerobic biotransformation study in a water/sediment system. The test system was flooded and incubated prior to treatment with the parent compound. For a valid

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study, the test system should be flooded and treated simultaneously to ensure that the primary microbial population is aerobic at the initiation of the study. Additionally, a foreign soil was utilized. However, since the compound was stable in the test system, the study does provide some useful information and an additional study is not required at this time. It is noted that the reported half-lives are not true aerobic transformation in water/sediment half-lives, as they do not represent the degradation/transformation of the parent compound in the whole test system, but reflect only the dissipation of the parent compound from the water phase to the sediment phase.

I. MATERIALS AND METHODS

GUIDELINE FOLLOWED: This study was conducted in accordance with USEPA Subdivision N Guideline §162-4 (1982), BBA Guidelines (Part IV, 5-1), and SETAC Europe Procedures for Assessing the Environmental Fate and Ecotoxicity of Pesticides (Part 8.2, p. 14). No deviations affected the validity of the study.

COMPLIANCE: This study was conducted in compliance with Federal Republic of Germany GLP Regulations (Chemikaliengesetz, Appendix 1-19a, 1994) and met the requirements of USEPA GLP Standards (40 CFR, Part 160, 1989, p. 3). Signed and dated Data Confidentiality, GLP, Quality Assurance, and study Certification statements were provided (pp. 2-6).

A. MATERIALS:

1. Test Materials: [Diphenyl-U-¹⁴C]- and [pyridine-3-¹⁴C]-labeled 2-chloro-*N*-(4'-chlorobiphenyl-2-yl)-nicotinamide.

Chemical Structure:

Description: Technical, solid. (pp. 15, 16, 54-56).

Purity:

[Diphenyl-U-¹⁴C]-labeled: Radiochemical purity: >99% (pp. 54, 64). Batch No.: 641-1018. Specific activity: 5.23 MBq/mg (314,000 dpm/μg).

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[Pyridine-3-¹⁴C]-labeled: Radiochemical purity: >97% (pp. 55, 65). Batch No.: 640-1026. Specific activity: 5.81 MBq/mg (349,000 dpm/μg).

Unlabeled: Purity: 99.4% (p. 56). Lot No.: 01174-236.

Storage conditions of test chemicals:

At low temperature (unspecified) in darkness (pp. 54, 55).

Table 1: Physico-chemical properties of BAS 510 F.

Parameter	Values	Comments
Molecular weight:	343.22 g/mol.	
Water solubility:	4.64 mg/L.	In deionized water at 20°C.
Vapor pressure/volatility:	Not reported.	
UV absorption:	Not reported.	
pK _a :	Not reported.	
K _{ow} /log K _{ow} :	Not reported.	
Stability of compound at room temperature:	Expected stability ≥2 years.	

Data obtained from pp. 15, 56 of the study report.

2. Water/sediment collection, storage and properties:

Table 2: Description of water/sediment collection and storage.

Description	Details
Geographic location of water and sediment for System A:	Kellmetschweiher pond located in a forested area of Kastenberghede west of Schifferstadt in south-western Germany.
Geographic location of water and sediment for System B:	Pond-like arm of Rhine river, referred to as Berghäuser Altrhein, located in forested area south of Speyer in south-western Germany.
Pesticide use history at the collection sites:	Not reported.
Sampling date:	June 2, 1998.
Collection procedures:	water: Not reported.
	sediment: Collected from upper 2-4 inches using a shovel.

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Description		Details
Sampling depth:	water:	Not reported.
	sediment:	ca. 40 cm for Kellmetschweiher sediment. ca. 15 cm for Berghäuser Altrhein sediment.
Storage conditions:		Not reported.
Storage length:		Not reported.
Preparation:	water:	0.2-mm sieved.
	sediment:	2-mm sieved.

Data obtained from pp. 16, 17, 57, 58 of the study report.

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Table 3: Properties of the waters.

Property	Details for Kellmetschweiher water		Details for Berghäuser Altrhein water	
Temperature (°C):	22.6		20.2	
pH:	8.50		8.10	
Redox potential (mV):	Initial:	Final:	Initial:	Final:
	237		211	
Oxygen concentration (mg/L):	Initial:	Final:	Initial:	Final:
	9.6		3.7	
Dissolved organic carbon (%):	Not determined.		Not determined.	
Total organic carbon (%):	Initial:	Final:	Initial:	Final:
	12.0	14.4	15.2	8.6
Hardness (mmol/L):	1.958		1.308	
Electrical conductivity:	Not determined.		Not determined.	
Biomass (CFU/mL):	Bacteria:	6.5 x 10 ²		1.0 x 10 ³
	Actinomycetes:	6		18
	Fungi:	4		10

Data obtained from pp. 16, 17, 57, 58 of the study report.

Table 4: Properties of the sediments.

Property	Details for Kellmetschweiher sediment	Details for Berghäuser Altrhein sediment
Textural classification:	Loamy sand.	Loam.
% sand:	84	49
% silt:	6	41
% clay:	10	10
pH (in CaCl ₂):	6.8	7.5
Organic carbon (%):	0.8	4.1
CEC:	11(meq/100 g)	18 (mVal/100 g)

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Property		Details for Kellmetschweiher sediment		Details for Berghäuser Altrhein sediment	
Redox potential (mV):		Initial:	Final:	Initial:	Final:
		-176		-50	
Bulk density (g/cm ³):		Not determined.		Not determined.	
Water content (%):		22.25		57.94	
ATP ¹ enzyme capacity (µg/kg soil):		13		1605	
Biomass (CFU/g):	Bacteria:	3.9 x 10 ⁷		5.3 x 10 ⁷	
	Actinomycetes:	2.5 x 10 ⁶		2.3 x 10 ⁶	
	Fungi:	7.7 x 10 ⁴		1.6 x 10 ⁶	

¹ATP = adenosine tri-phosphate.

Data obtained from pp. 57, 58 of the study report.

B. EXPERIMENTAL CONDITIONS:

1. Preliminary experiments: None.

2. Experimental conditions:

Table 5: Study design.

Criteria		Details for Kellmetschweiher system (System A)	Details for Berghäuser Altrhein system (System B)
Duration of the test:		100 days.	
Water: Filtered/unfiltered water: Type and size of filter used:		Filtered. 0.2-mm sieved.	
Water/sediment per replicate:	water:	ca. 290 mL.	ca. 290 mL.
	sediment:	ca. 190 g wet wt.	ca. 140 g wet wt.
Sediment:water ratio:		ca. 1:1.5 (w:v).	ca. 1:2.1 (w:v).
Nominal application rate:		0.24 mg a.i./L water. Each system treated with [diphenyl-U- ¹⁴ C]- or [pyridine-3- ¹⁴ C]-labeled BAS 510.	
Control conditions, if used:	Sterile:	Sterile (120°C for 30 minutes) water/sediment systems were treated with [diphenyl-U- ¹⁴ C]- or [pyridine-3- ¹⁴ C]-labeled BAS 510 F.	

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Criteria		Details for Kellmetschweiher system (System A)		Details for Berghäuser Altrhein system (System B)	
	Non-sterile:	Non-sterile water/sediment systems were treated with unlabeled BAS 510 F.			
No. of Replications:	Controls, if used:	Single sterile and non-sterile systems taken at study termination.			
	Treatments:	Single system taken at each collection interval.			
Test apparatus (type/material/volume):		Flasks (material/volume not specified) containing the water/sediment were pre-incubated for 22 days in a temperature-controlled chamber prior to treatment. The water/sediment systems were incubated in sealed flasks equipped with inlet/outlet ports for the collection of volatiles. A hanging magnetic stirrer was used to agitate the water layer in each flask.			
Details of traps for CO ₂ and organic volatiles, if any:		Humidified, CO ₂ -free air was continuously drawn (flow rate not specified) through the headspace in each non-sterile treated flask, then sequentially through ethylene glycol (one trap) and 0.5 N sodium hydroxide (two traps).			
If no traps were used, is the system closed/open?		Volatiles traps were used for non-sterile treated systems. Sterile treated systems were incubated closed.			
Identity and concentration of co-solvent:		Acetonitrile, final concentration 0.03% based on water layer.			
Test material application:	Vol. of test solution used/treatment:	0.1 mL of 722 µg/mL test solution.			
	Application method:	Applied to surface of water layer via pipet.			
Any indication of the test material adsorbing to the walls of the test apparatus?		Not indicated.			
Microbial biomass (CFU/mL) of control water ¹ :		Initial:	Final:	Initial:	Final:
	Bacteria:	6.5 x 10 ²	4.7 x 10 ²	1.0 x 10 ³	2.6 x 10 ³
	Actinomycetes:	6	20	18	0
	Fungi:	4	0	10	0
Microbial biomass (CFU/mL) of control sediment ¹ :		Initial:	Final:	Initial:	Final:
	Bacteria:	3.9 x 10 ⁷	4.0 x 10 ⁷	5.3 x 10 ⁷	3.4 x 10 ⁸
	Actinomycetes:	2.5 x 10 ⁶	1.5 x 10 ⁶	2.3 x 10 ⁶	2.8 x 10 ⁶
	Fungi:	7.7 x 10 ⁴	1.2 x 10 ⁵	1.6 x 10 ⁶	5.7 x 10 ⁵

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Criteria		Details for Kellmetschweiher system (System A)		Details for Berghäuser Altrhein system (System B)	
Microbial biomass/ population of treated:		Initial:	Final:	Initial:	Final:
	water:	ND ²	ND	ND	ND
	sediment:	ND	ND	ND	ND
Experimental conditions:	Temperature (°C):	20 ± 2°C.			
	Continuous darkness:	Yes.			
Other details, if any:		None.			

¹See Reviewer's Comment No. 8.

²ND = Not determined.

Data obtained from pp. 17, 18, 57-59 of the study report.

3. Aerobic conditions: Humidified, CO₂-free air continuously drawn (flow rate not specified) through the headspace of each flask with the water layer agitated via a hanging magnetic stirrer (p. 59). The dissolved oxygen content and redox potential of the water/sediment systems were determined at 2- to 7-day intervals during the 22-day pre-incubation period and at each sampling interval following treatment (pp. 60-63).

4. Supplementary experiments: To facilitate the identification of possible biotransformation products, additional water/sediment systems were treated with [diphenyl-U-¹⁴C]- or [pyridine-3-¹⁴C]-labeled BAS 510 F at the higher nominal rate of 0.72 mg a.i./L water; incubation conditions were as described above with duplicate systems sampled at 62 and 105 days after treatment (pp. 17, 18).

5. Sampling:

Table 6: Sampling details.

Criteria		Details
Sampling intervals:	Controls:	
	Sterile:	101 days.
	Non-sterile:	105 days.
	Treated:	0, 1, 2, 7, 14, 29, 59 and 100 days.
Sampling method:		Duplicate water/sediment systems were collected at each interval.
Method of collection of CO ₂ and volatile organic compounds:		Trapping solutions were collected and replaced at each sampling interval.

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Criteria	Details
Sampling intervals/times for: Sterility check, if sterile controls are used: Aerobicity:	Sterility was not verified. Oxygen concentration of the water layer and redox potential of the water and sediment were determined at each sampling interval.
Sample storage before analysis:	Water layers and sediment were separated upon sampling. All non-sterile treated water layers were analyzed the same day collected. Sediment extracts were analyzed within 1 week of collection. Water layers and sediment extracts were stored refrigerated or frozen (temperatures not specified).
Other observations, if any:	None.

Data obtained from pp. 18, 19, 61, 63, 66 of the study report.

C. ANALYTICAL METHODS:

Separation of water and sediment: Upon sampling, the temperature, pH and oxygen concentration of the water layer and the redox potential of the water layer and sediment were determined for each water/sediment system (pp. 18, 61, 63). Water and sediment were then separated by decanting the water and transferring the sediment to a centrifuge tube.

Extraction/clean up/concentration methods: Water layers were analyzed directly by HPLC. For high performance TLC (HPTLC) analysis, an aliquot (typically 20 mL) of the water layer was evaporated under vacuum and the residue re-dissolved in acetonitrile:water (1:1, v:v, 5 mL; p. 18). Sediment samples were extracted three times each with acetonitrile:water (1:1, v:v) and acetonitrile; extraction volumes were 50-70 mL (pp. 18-20). For each extraction, the sediment in extraction solution was shaken on a rotary shaker for 15 minutes at 180 rpm, and the sediment and extract were separated by centrifugation. Extracts were combined prior to HPLC and HPTLC analyses.

Total ¹⁴C measurement: For all sampling intervals, aliquots (volume not specified) of the separated water layer, sediment extracts, and trapping solutions were analyzed for total radioactivity by LSC (pp. 29, 30, 70-73). Extracted sediment was air dried at room temperature, then aliquots (amount not specified) were analyzed for total radioactivity by LSC following combustion.

Non-extractable residue determination: Aliquots (ca. 25 g) of extracted 59- and 100-day sediment samples were further extracted three times with 0.5 M sodium hydroxide solution followed by a water rinse (p. 19). Sodium hydroxide extracts were combined and acidified to pH 1 with concentrated hydrochloric acid, then the resulting precipitate (humic acids) was removed

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by centrifugation (7,250-10,400 x g, 10 minutes). The precipitate was re-dissolved in 0.5 M sodium hydroxide and analyzed by LSC. Remaining acidified extract (fulvic acids) was analyzed by LSC, then partitioned three times with ethyl acetate; organic phases were combined and analyzed by HPLC. [¹⁴C]Residues remaining in the extracted sediment (humins) were quantified by LSC following combustion.

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

Identification and quantification of parent compound: Water and sediment extracts were analyzed by reverse-phase HPLC under the following conditions: Waters Spherisorb ODS-2, SNC column (8 x 250 mm, 10 μm particle size), Waters Spherisorb ODS-2, SPC guard column (8 x 33 mm, particle size not specified), gradient mobile phase combining (A) water:acetonitrile:formic acid (95:5:0.1, v:v) and (B) water:acetonitrile:formic acid (5:95:0.1, v:v) [percent A:B at 0 min. 100:0 (v:v), 15 min. sample injected, 20 min. 100:0, 55 min. 0:100, 58 min. 0:100, 60 min. 100:0], injection volume not specified, flow rate 2.5 mL/minute, UV (wavelength not specified) and radioactivity detection (pp. 21, 38-45). It was not specified how parent [diphenyl-U-¹⁴C]- and [pyridine-3-¹⁴C]-labeled BAS 510 F were identified in the water layers and sediment extracts.

Water layers and sediment extracts were also analyzed using normal-phase one-dimensional HPTLC on silica gel plates developed with isopropyl ether:methanol:formic acid (80:20:1, v:v; pp. 20, 21, 46-51). Unlabeled reference compound was chromatographed with the samples. Following development, areas of radioactivity were detected using a Fuji BAS 1000 Imager and unlabeled reference compounds were detected under UV light (254 nm).

Identification and quantification of transformation products: No major transformation products were detected in water layers or sediment extracts.

Identification of radioactivity recovered in the 100-day sodium hydroxide trapping solutions was confirmed as ¹⁴CO₂ by two methods (p. 19). An aliquot of the trapping solution was acidified with concentrated hydrochloric acid, purged with nitrogen to release dissolved ¹⁴CO₂, then the purged solution was analyzed for any remaining radioactivity by LSC. The presence of ¹⁴CO₂ in the sodium hydroxide trapping solutions was also confirmed by barium chloride precipitation.

Detection limits (LOD, LOQ) for the parent compound and transformation products: Detection limits for the LSC, HPLC and HPTLC analyses were not reported.

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II. RESULTS AND DISCUSSION:

A. TEST CONDITIONS: During the pre-incubation period and following treatment, aerobicity, temperature and other environmental conditions were maintained in the water layer, but anaerobic conditions existed in the sediment layer of the water/sediment systems.

Water/loamy sand sediment systems (system A): During the 22-day pre-incubation period, temperatures were 19.0-20.8°C, pH values were 7.16-8.40, oxygen concentrations were 6.5-8.7 mg/L (72-94%), and redox potentials were 198 to 315 mV in the water layer and -115 to -152 in the sediment (p. 60). During the 100-day study following treatment with [¹⁴C]BAS 510 F (both labels), temperatures were 19.3-20.0°C, pH values were 7.30-8.72, oxygen concentrations were 5.7-8.6 mg/L (65-94%), and redox potentials were 125 to 295 mV in the water layer and -100 to -146 in the sediment (p. 61). In sterile water/loamy sand sediment systems treated with [¹⁴C]BAS 510 F (both labels) at 101 days posttreatment, temperatures were 20.2-20.5°C, pH values were 7.66-7.67, oxygen concentrations were 6.7-7.0 mg/L (75-79%), and redox potentials were 120 to 124 mV in the water layer and -94 to -98 in the sediment. In control non-sterile water/loamy sand sediment systems treated with unlabeled BAS 510 F at 105 days posttreatment, temperatures were 19.3-19.7°C, pH values were 6.69-8.07, oxygen concentrations were 3.6-8.0 mg/L (40-86%), and redox potentials were 173 to 217 mV in the water layer and -95 to -99 in the sediment.

Water/loam sediment systems (system B): During the 22-day pre-incubation, temperatures were 19.2-21.4°C, pH values were 7.17-8.54, oxygen concentrations were 4.4-8.3 mg/L (48-93%), and redox potentials were 195 to 310 mV in the water layer and -130 to -205 in the sediment (p. 62). During the 100-day study following treatment with [¹⁴C]BAS 510 F (both labels), temperatures were 19.5-21.6°C, pH values were 7.21-8.54, oxygen concentrations were 1.1-8.5 mg/L (12-94%), and redox potentials were 120 to 260 mV in the water layer and -118 to -210 in the sediment (p. 63). In sterile water/loamy sand sediment systems treated with [¹⁴C]BAS 510 F (both labels) at 101 days posttreatment, temperatures were 20.1-20.2°C, pH values were 6.91-7.36, oxygen concentrations were 2.4-6.8 mg/L (26-76%), and redox potentials were 125 to 138 mV in the water layer and -88 to -105 in the sediment. In control non-sterile water/loamy sand sediment systems treated with unlabeled BAS 510 F at 105 days posttreatment, temperatures were 20.0-20.3°C, pH values were 7.85-8.22, oxygen concentrations were 8.0-8.1 mg/L (89-90%), and redox potentials were 135 to 140 mV in the water layer and -166 to -175 in the sediment.

B. MATERIAL BALANCE: During the 100-day incubation, overall recovery of radiolabeled material (both labels) for both water/sediment systems averaged $97.1 \pm 1.2\%$ ($n = 32$, range 92.5-100.2%) of the applied. Recoveries of radiolabeled material in the water/loamy sand sediment averaged $97.9 \pm 1.1\%$ ($n = 8$, range 96.4-100.2%) of the applied in [diphenyl-U-¹⁴C]BAS 510 F-

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treated systems and $97.3 \pm 0.9\%$ (96.5-99.3%) in [pyridine-3-¹⁴C]BAS 510 F-treated systems and in water/loam sediment averaged $96.4 \pm 1.6\%$ (92.5-97.6%) of the applied in [diphenyl-U-¹⁴C]BAS 510 F-treated systems and $97.0 \pm 0.5\%$ (96.0-97.5%) in [pyridine-3-¹⁴C]BAS 510 F-treated systems (pp. 70-73).

Table 7: Biotransformation of [diphenyl-U-¹⁴C]BAS 510 F, expressed as percentage of applied radioactivity (n = 1), in water/loamy sand sediment systems (system A) under aerobic conditions.

Compound		Sampling times (days)							
		0	1	2	7	14	29	59	100
BAS 510 F	water	97	75.8	69.5	55.2	40	30.9	25.3	17.4
	sediment	NA ¹	20.1	26.5	38.6	53.2	60.4	63.9	68.6
Total CO ₂	entire system	NA	0.0	0.0	0.0	0.1	0.0	0.2	0.4
Total volatile organics	entire system	NA	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Non-extractable residues	sediment	0.1	0.7	1.6	2.5	4.6	6.9	10.8	12.4
Total % recovery		97.4	96.6	97.7	96.4	97.9	98.2	100.2	98.8

¹Not analyzed.

Data obtained from pp. 70, 74 of the study report.

Table 8: Biotransformation of [pyridine-3-¹⁴C]BAS 510 F, expressed as percentage of applied radioactivity (n = 1), in water/loamy sand sediment systems (system A) under aerobic conditions.

Compound		Sampling times (days)							
		0	1	2	7	14	29	59	100
BAS 510 F	water	96.1	76	71	51.7	44.5	31.1	21.4	17.3
	sediment	NA ¹	19.8	24.7	42.2	48.1	60.1	64.5	66.8
Total CO ₂	entire system	NA	0.0	0.0	0.0	0.1	0.1	0.3	0.7
Total volatile organics	entire system	NA	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Non-extractable residues	sediment	0.0	0.8	1.4	2.9	3.8	6.0	13.1	13.4
Total % recovery		96.7	96.6	97.1	96.8	96.5	97.3	99.3	98.1

¹Not analyzed.

Data obtained from pp. 71, 75, 76 of the study report.

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Table 9: Biotransformation of [diphenyl-U-¹⁴C]BAS 510 F, expressed as percentage of applied radioactivity (n = 1), in water/loam sediment systems (system B) under aerobic conditions.

Compound		Sampling times (days)							
		0	1	2	7	14	29	59	100
BAS 510 F	water	96.9	63.7	54.2	32.3	21.4	12.1	7.4	6.1
	sediment	NA ¹	32.4	41.4	61.5	69.2	78.4	76.5	80.2
Total CO ₂	entire system	NA	0.0	0.0	0.0	0.1	0.2	0.5	0.5
Total volatile organics	entire system	NA	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Non-extractable residues	sediment	0.0	1.5	1.6	2.9	4.9	6.2	8.1	10.5
Total % recovery		97.5	97.6	97.2	96.7	95.6	97.0	92.5	97.3

¹Not analyzed.

Data obtained from pp. 72, 77 of the study report.

Table 10: Biotransformation of [pyridine-3-¹⁴C]BAS 510 F, expressed as percentage of applied radioactivity (n = 1), in water/loam sediment systems (system B) under aerobic conditions.

Compound		Sampling times (days)							
		0	1	2	7	14	29	59	100
BAS 510 F	water	95.9	63.0	50.2	33.9	23	12.1	8.4	6.2
	sediment	NA ¹	31.7	44.7	60.4	69.8	79.1	80.1	79.5
Total CO ₂	entire system	NA	0.0	0.0	0.0	0.0	0.3	0.6	0.5
Total volatile organics	entire system	NA	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Non-extractable residues	sediment	0.1	1.3	2.1	3.1	4.4	5.9	8.2	10.3
Total % recovery		96.7	96.0	97.0	97.5	97.1	97.5	97.4	96.5

¹Not analyzed.

Data obtained from pp. 73, 78, 79 of the study report.

C. TRANSFORMATION OF PARENT COMPOUND: In water/loamy sand sediment systems (system A), [¹⁴C]BAS 510 F (both labels) in the total system gradually decreased from 96.1-97.0% of the applied radioactivity at day 0 to 84.1-86.0% at 100 days posttreatment (pp. 74-76). [¹⁴C]BAS 510 F in the water layer decreased 96.1-97.0% at day 0 to 51.7-55.2% at 7 days, and 30.9-31.1% at 29 days, and was 17.3-17.4% at 100 days. In the sediment, [¹⁴C]BAS 510 F increased from 19.8-20.1% at 1 day to 48.1-53.2% at 14 days, and was 66.8-68.6% at 100

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days. Following application of [¹⁴C]BAS 510 F to the surface of the water layer, [¹⁴C]residues gradually partitioned into the sediment with distribution ratios (water:sediment) of *ca.* 4:1 at day 1, 1:1 at 7-14 days, 1:2 at 29 days and 1:5 at 100 days. In sterile water/loamy sand sediment systems at 101 days, [¹⁴C]BAS 510 F (both labels) comprised 14.3-15.0% of the applied in the water layer and 70.6-72.4% in the sediment.

In water/loam sediment systems (system B), [¹⁴C]BAS 510 F (both labels) in the total system decreased from 95.9-96.9% of the applied radioactivity at day 0 to 85.7-86.3% at 100 days posttreatment (minimum of 83.9% at 59 days, pp. 77-79). [¹⁴C]BAS 510 F in the water layer decreased from 95.9-96.9% at day 0 to 50.2-54.2% at 2 days, 21.4-23.0% at 14 days, and 12.1% (both labels) at 29 days, and was 6.1-6.2% at 100 days. In the sediment, [¹⁴C]BAS 510 F increased from 31.7-32.4% at 1 day to 60.4-61.5% at 7 days and was 76.5-80.2% at 29-100 days. Following application of [¹⁴C]BAS 510 F to the surface of the water layer, [¹⁴C]residues gradually partitioned into the sediment with distribution ratios (water:sediment) of *ca.* 2:1 at day 1, 1:1 at 2 days, 1:2 at 7 days, 1:7 at 29 days and 1:15 at 100 days. In sterile water/loam sediment systems at 101 days, [¹⁴C]BAS 510 F (both labels) comprised 7.0-7.6% of the applied in the water layer and 83.1-84.8% in the sediment.

HALF-LIFE/DT₅₀: DT₅₀ and DT₉₀ values for the dissipation of [¹⁴C]BAS 510 F from the water layer to the sediment were determined from non-linear degradation curves calculated by ModelMaker version 3.0.4 software (pp. 27, 52, 53). Results from both labels were combined per respective system for the graphical determination of DT₅₀ and DT₉₀ values. Half-life values for the dissipation of [diphenyl-U-¹⁴C]- and [pyridine-3-¹⁴C]-BAS 510 F from the water layer to the sediment were also determined using least squares linear regression analysis (assuming dissipation followed first-order kinetics) calculated by Corel Quattro Pro 8 software; dissipation appeared bi-phasic for both label studies. It is noted that the reported half-lives are not true aerobic transformation in water/sediment half-lives, as they do not represent the degradation/transformation of the parent compound in the whole test system, but reflect only the dissipation of the parent compound from the water phase to the sediment phase.

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Table 11: Half-life ($t_{1/2}$)/DT₅₀ values of dissipation BAS 510 F from water layer to sediment in aerobic water/sediment systems.

System	¹⁴ C label	First order half-life				Regression equation	DT ₅₀ ¹ (days)	DT ₉₀ (days)
		half-life ¹ (days)						
		initial	r ²	secondary	r ²			
water/loamy sand sediment (system A)	diphenyl	12.4	0.931	75.6	0.98	Linear form $y = mx + b$ as $\ln C = -kt + \ln C_0$; $\ln C_0$ is initial concentration ($b = y$ intercept), $\ln C$ is concentration at time t (y), k is the slope (m), t is time (x) or $kt = \ln C_0 - \ln C$. Half-life ($t_{1/2}$) = $-(\ln/k)$.	ND ³	ND
	pyridine	13.8	0.871	66.1	0.911		ND	ND
	diphenyl + pyridine	ND	ND	ND	ND		9	133
water/loam sediment (system B)	diphenyl	5	0.902	41.1	0.841		ND	ND
	pyridine	5.4	0.845	40.9	0.85		ND	ND
	diphenyl + pyridine	ND	ND	ND	ND		3	43

¹Determined by the reviewer.

²Determined by the registrant (pp. 27, 52, 53).

³Not determined.

TRANSFORMATION PRODUCTS: No major transformation products of [diphenyl-U-¹⁴C]- or [pyridine-3-¹⁴C]-BAS 510 F were detected in the water layers or sediment extracts from either system. Up to four minor unidentified [¹⁴C]compounds were detected (each at ≤0.4% of the applied radioactivity) in the water layers of the two systems, but were not detected in the sediment extracts (pp. 80-85).

NON-EXTRACTABLE AND EXTRACTABLE RESIDUES: In water/loamy sand sediment systems (system A), extractable [¹⁴C]residues (both labels) increased from 0.4-0.6% of the applied at day 0 to 66.8-68.6% at 100 days; non-extractable [¹⁴C]residues increased from ≤0.1% at day 0 to 10.8-13.4% at 59-100 days (pp. 70, 71). In extracted 59- and 100-day sediment samples (both labels), 4.8-6.3% of the applied was associated with fulvic acids, 2.2-2.4% with humic acids and 0.6-0.7% with humins (p. 37).

In water/loam sediment systems (system B), extractable [¹⁴C]residues (both labels) increased from 0.6-0.7% of the applied at day 0 to 78.4-79.1% at 29 days and were 76.5-80.2% at 59-100 days; non-extractable [¹⁴C]residues increased from ≤0.1% at day 0 to 10.3-10.5% at 100 days (pp. 72, 73). In extracted 100-day sediment samples (both labels), 1.6-1.7% of the applied was associated with fulvic acids, 3.4-3.9% with humic acids and 2.2-2.3% with humins (p. 37).

VOLATILIZATION: Volatilization of ¹⁴CO₂ was not significant for either [diphenyl-U-¹⁴C]- or [pyridine-3-¹⁴C]-BAS 510 F, totaling ≤0.7% of the applied at study termination in both

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water/sediment systems; organic volatiles were not detected (<0.1% of applied) at any sampling interval (pp. 70-73).

TRANSFORMATION PATHWAY: BAS 510 F was found to be stable in two aerobic water/sediment systems with gradual dissipation of parent compound from the water layer to the sediment.

Table 12: Chemical names for identified transformation products of BAS 510 F in aerobic water/sediment systems.

Registrant's Code Name	Cas Number	CAS and/or IUPAC Chemical Name(s)	Chemical formula	Molecular weight	SMILES string
None					

D. SUPPLEMENTARY EXPERIMENT-RESULTS: None reported.

III. STUDY DEFICIENCIES: This study is acceptable and can be used to fulfill Subdivision N Guideline §162-4 data requirements.

IV. REVIEWER'S COMMENTS:

1. Anaerobic conditions existed in the sediment layers of the water/sediment systems during the pre-incubation period and following treatment, but aerobicity was maintained in the water layers throughout the entire study. Given the stability of BAS 510 F in the water/sediment systems over the 100-day incubation, as well as the relatively slow degradation of the compound in aerobic soil systems (*cf.* MRID 45405208), it is unlikely that the anaerobic conditions in the sediment layers had any impact on the study results. The reviewer notes, however, that the reported half-lives are not true aerobic transformation in water/sediment half-lives, as they do not represent the degradation/transformation of the parent compound in the whole test system, but reflect only the dissipation of the parent compound from the water phase to the sediment phase.
2. The registrant averaged results from the two different [¹⁴C]-labels ([diphenyl-U-¹⁴C]- and [pyridine-3-¹⁴C]-) used to treat each water/sediment system (loamy sand and sand sediments) based on the conclusion that the differently labeled test substances degraded similarly within a given system (pp. 24, 25, 29-36). In this DER, the reviewer presented the individual results for each [¹⁴C]-label (pp. 70-85). In future studies, it is necessary that replicate data be obtained and reported for each label/sediment type combination utilized.

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3. For HPLC analyses, it was not specified how parent [diphenyl-U-¹⁴C]- and [pyridine-3-¹⁴C]-labeled BAS 510 F were identified in water layers and sediment extracts. The registrant should specify whether parent [¹⁴C]BAS 510 F in the water layers and sediment extracts was identified by co-chromatography with labeled reference standard and/or comparison to the retention time of an unlabeled reference standard, and provide supporting chromatograms. For HPTLC analyses, the registrant indicated that unlabeled reference compounds were used, but, again, it could not be determined if [¹⁴C]BAS 510 F was identified by co-chromatography or R_f value, and supporting chromatograms were not provided.
4. HPLC column recoveries were not reported.
5. Detection limits for the LSC, HPLC and HPTLC analyses were not reported.
6. The registrant reported that the presence of ¹⁴CO₂ in the 100-day sodium hydroxide trapping solutions was confirmed by two methods, but supporting results were not provided. Formation of ¹⁴CO₂ and organic volatiles was insignificant, totaling <1% of applied at study termination for both labels in the water/sediment systems.
7. The material and volume of the incubation flasks used to contain the water/sediment systems were not specified.
8. Biomass, as plate counts of bacteria, actinomycetes and fungi; and enzyme capacity, as presence of ATP, of the sediment and water were determined at the beginning and end of incubation (pp. 57, 58). It was unclear as to exactly which samples (treated/untreated) were analyzed. The registrant reported that the water/sediment systems treated with unlabeled BAS 510 F at 0.24 mg a.i./L water were used for parameter characterization at the end of the incubation period. It is assumed that this included the total nitrogen, phosphorus and total organic carbon results; ATP concentration, and biomass determinations that were reported in Appendices 4 and 5 (pp. 18, 57, 58). The registrant should clarify this issue.
9. The registrant reported that the application rate used in this study (0.24 mg a.i./L water) was calculated as equivalent to the concentration expected when the field application rate for BAS 510 F of 700 g a.i./ha was equally distributed in water to a depth of 30 cm (p. 24). That application rate would yield a concentration of 233 µg a.i./L or 70 µg a.i. per incubation flask based on the volume of water used (ca. 300 mL).
10. BAS 510 F chemical name 2-chloro-*N*-(4'-chlorobiphenyl-2-yl)-nicotinamide, as presented in the study report, was identified as the IUPAC name by the Compendium of Pesticide Common Names (<http://www.hclrss.demon.co.uk/nicobifen.html>). The CAS name 2-chloro-*N*-(4-chloro[1,1-biphenyl]-2-yl)-3-pyridinecarboxamide was also obtained from the Compendium of Pesticide Common Names. The following BAS 510 F synonyms were

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obtained from USEPA/OPP Chemical Databases (<http://www.cdpr.ca.gov/cgi-bin/epa/chemidtriris.pl?pccode=128008> and (http://www.cdpr.ca.gov/cgi-bin/mon/bycode.pl?p_chemcode=5790): 2-chloro-*N*-(4'-chlorobiphenyl-2-yl)-nicotinamide, nicobifen, and BAS 516 02 F.

V. REFERENCES: The following reference was cited in the study:

1. Walker, A. and N. Crout. 1997. ModelMaker, User Manual, Version 3. Cherwell Scientific Publishing Limited, Oxford.

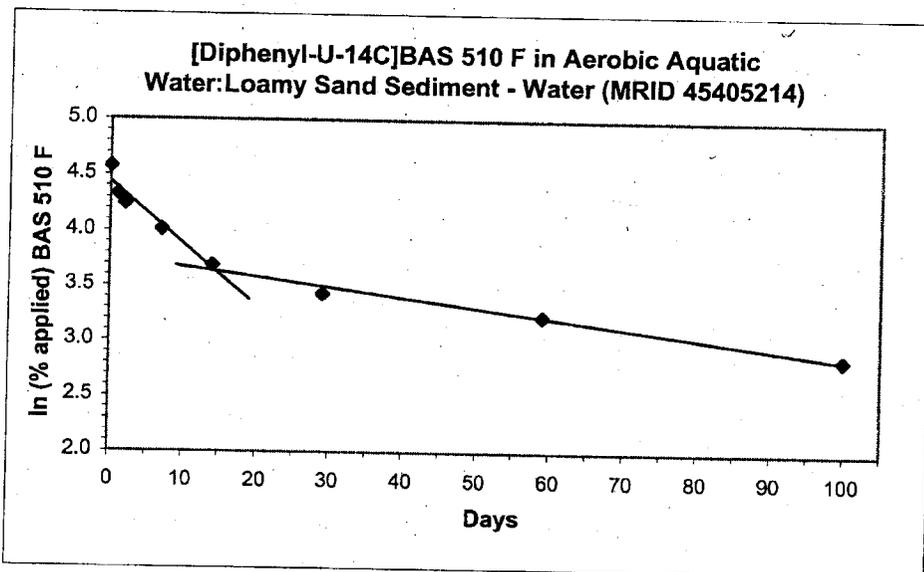
Aerobic Aquatic Metabolism of [14C]BAS 510 F in Water:Sediment
 MRID 45405214

[Diphenyl-U-14C]BAS 510 F in water-loamy sand sediment
 Half-life; dissipation from water layer to sediment
 all intervals

Day	BAS 510 F %AR	Ln(%AR)
0	97	4.574711
1	75.8	4.328098
2	69.5	4.241327
7	55.2	4.010963
14	40	3.688879
29	30.9	3.430756
59	25.3	3.230804
100	17.4	2.85647

Regression Output:

Constant	4.199
Std Err of Y Est	0.26
R Squared	0.836
No. of Observations	8
Degrees of Freedom	6
X Coefficient(s)	-0.015223
Std Err of Coef.	0.002749



half-life 45.52379 days

*AR = Applied Radioactivity
 Linear regression analysis performed
 using Corel Quattro.Pro 8.
 Results from p. 74 of study report.

[Pyridine-3-14C]BAS 510 F in water-loamy sand sediment (system A)

Half-life; dissipation from water layer to sediment

All intervals

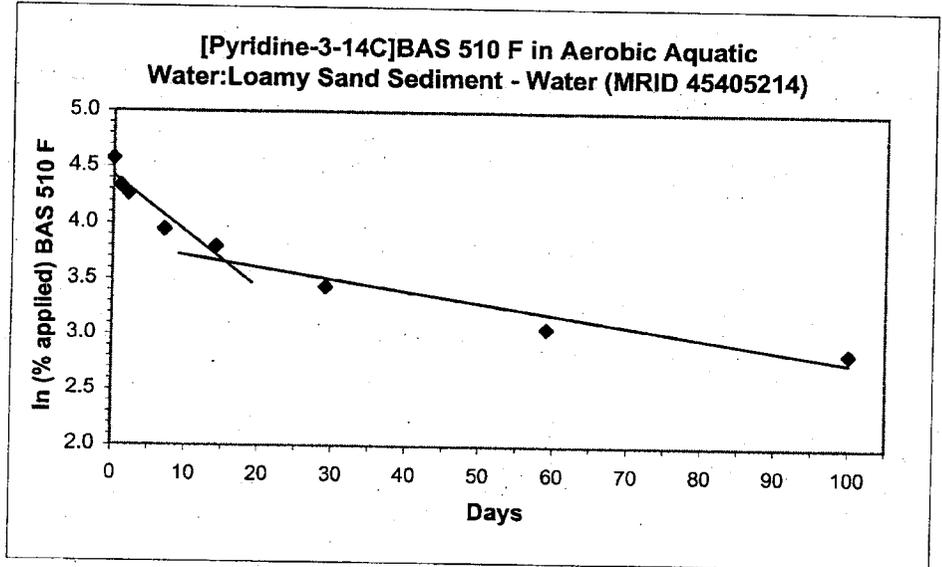
Day	BAS 510 F %AR	Ln(%AR)
0	97	4.574711
1	76	4.330733
2	71	4.26268
7	51.7	3.945458
14	44.5	3.795489
29	31.1	3.437208
59	21.4	3.063391
100	17.3	2.850707

Regression Output:

Constant	4.205
Std Err of Y Est	0.262
R Squared	0.847
No. of Observations	8
Degrees of Freedom	6
X Coefficient(s)	-0.015948
Std Err of Coef.	0.002772

half-life 43.45351 days

*AR = Applied Radioactivity
 Linear regression analysis performed
 using Corel Quattro Pro 8.
 Results from p. 75 of study report.



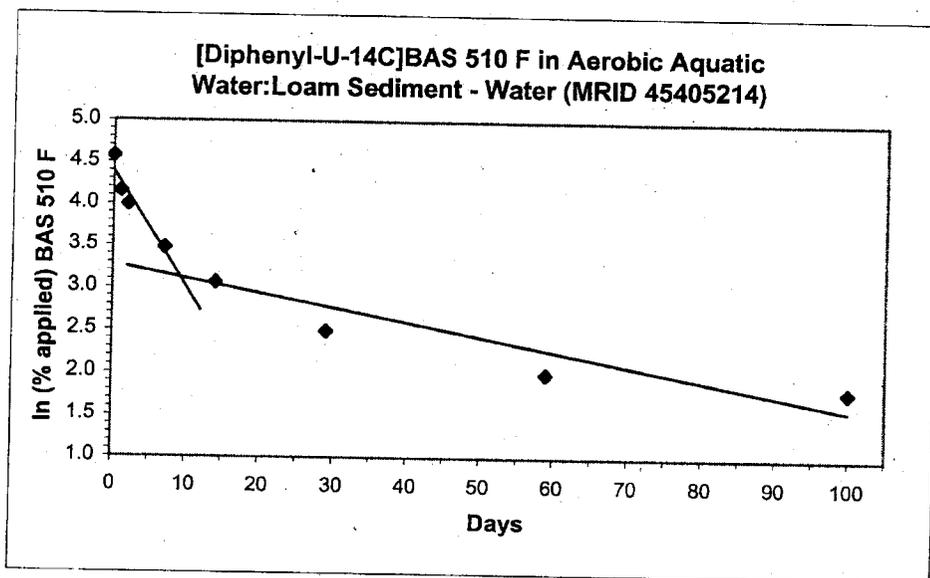
Aerobic Aquatic Metabolism of [14C]BAS 510 F in Water:Sediment
 MRID 45405214

[Diphenyl-U-14C]BAS 510 F in water-loam sediment (system B)
 Half-life; dissipation from water layer to sediment
 All intervals

Day	BAS 510 F %AR	Ln(%AR)
0	96.9	4.57368
1	63.7	4.154185
2	54.2	3.992681
7	32.3	3.475067
14	21.4	3.063391
29	12.1	2.493205
59	7.4	2.00148
100	6.1	1.808289

Regression Output:

Constant	3.867
Std Err of Y Est	0.524
R Squared	0.777
No. of Observations	8
Degrees of Freedom	6
X Coefficient(s)	-0.025332
Std Err of Coef.	0.005537



half-life 27.35694 days

*AR = Applied Radioactivity
 Linear regression analysis performed
 using Corel Quattro Pro 8.
 Results from p. 77 of study report.

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[Pyridine-3-14C]BAS 510 F in water-loam sediment (system B)
Half-life; dissipation from water layer to sediment

All intervals

Day	BAS 510 F %AR	Ln(%AR)
0	95.9	4.563306
1	63	4.143135
2	50.2	3.916015
7	33.9	3.523415
14	23	3.135494
29	12.1	2.493205
59	8.4	2.128232
100	6.2	1.824549

Regression Output:

Constant	3.87
Std Err of Y Est	0.493
R Squared	0.789
No. of Observations	8
Degrees of Freedom	6
X Coefficient(s)	-0.024672
Std Err. of Coef.	0.005208

half-life 28.08824 days

*AR = Applied Radioactivity
Linear regression analysis performed
using Corel Quattro Pro 8.
Results from p. 78 of study report.

