UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
OFFICE OF PESTICIDE PROGRAMS
ENVIRONMENTAL CHEMISTRY LABORATORY
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March 10, 2009

MEMORANDUM

SUBJECT: AE1170437 - ECM0245S1-S6 DP # D356962

FROM: Joseph Ferrario, Branch Chief
BEAD/Environmental Chemistry Laboratory

TO: Margaret Ervin ECM Gatekeeper
OPP/Environmental Fate and Effects Division
EISB 7507P

The EFED/Environmental Fate and Effects Division has requested an Environmental Chemistry Method Review of a method for the determination of AE1170437 and its metabolites in soil and sediment (MRID No. 47443276). The method was submitted by Bayer CropScience- in accordance with the registration of the above mentioned analytes. The method validation data was reviewed and the conclusions included in the attached Environmental Chemistry Method Review Report.

The following report includes an overview of the method and the method completeness, statements of adherence to EPA regulations, a presentation of results and a discussion of problems found in the registrant method and those discovered by the independent laboratory. A statement of method acceptability is also included.

If you have any questions concerning this report, please contact Elizabeth Flynt at (228) 688-2410 or me at (228) 688-3212.

Attachments
cc: Dr. Christian Byrne, QA Officer
BEAD/Environmental Chemistry Laboratory

Elizabeth C. Flynt
BEAD/ECL

AE1170437 in Soil and Sediment/680818/
Bayer CropScience/264

ENVIRONMENTAL CHEMISTRY METHOD REVIEW REPORT

Data Requirement: PMRA Data Code: NA
EPA DP Barcode: 356962
OECD Data Point: NA
EPA Guideline: ECM Method Review

Test material:
Common name: AE1170437
Chemical name: N-[(1R,2S)-2,3-Dihydro-2,6-dimethyl-1H-inden-1-yl]-6-[(1R)-1-fluoroethyl]-1,3,5-triazine-2,4-diamine

IUPAC name: N-[(1R,2S)-2,3-dihydro-2,6-dimethyl-1H-inden-1-yl]-6-(1-fluoroethyl)-1,3,5-triazine-2,4-diamine

Primary Evaluator: _____ **Date:** 1/12/09
Dr. Jeremy Stewart, Chemist

Peer Reviewer: _____ **Date:** 1/12/09
Charles Kennedy, Chemist

QA Officer: _____ **Date:** 03/09/09
Dr. Christian Byrne, QA Officer

ANALYTICAL METHOD: 47443276, Tiambo Xu, June 16, 2008, *"Method of Analysis for the Determination of Residues of AE1170437 and its metabolites AE1170437 Carboxylic Acid (AE2158969), AE1170437 Triazine-indanone (AE2158968), AE1170437 Hydroxyethyl (AE2300077), AE1170437 Olefin (BCS-AA10201), and AE1170437 Diaminotriazine (1-Fluoroethyl triazinediamine) in Soil and Sediment Using LC/MS/MS"*, The unpublished method was developed by Bayer CropScience, Stilwell, Kansas. Pages 1-55. The study no. is DH-002-S06-02.

EXECUTIVE SUMMARY

The method is applicable for the quantitative determination of residues of AE1170437 and its metabolites in soil and sediment. The method was created by Bayer CropScience, and claims exemption from EPA's Good Laboratory Practice Standards, Title 40 Code of Federal Regulations Part 160; stating "According to 40 CFR Part 160.3, this is not an experimental study. Therefore, the rules and regulations set forth under 40 CFR Part 160 do not apply." After a thorough review, ECB found that this method met many of the criteria for a scientifically valid method. This method is acceptable with minor revisions.

AE1170437 in Soil and Sediment/680818/
Bayer CropScience/264

ENVIRONMENTAL CHEMISTRY METHOD REVIEW REPORT

Method Summary

A 15 g soil or sediment sample is extracted with an acetonitrile/water mixture using microwave digestion, and an aliquot of the supernatant is centrifuged and diluted with water. It then undergoes ESI-LC/MS/MS analysis. The limits of quantitation (LOQ) were reported to be 1.5 µg/kg for all analytes.

METHOD ACCEPTABILITY/DEFICIENCIES/CLARIFICATIONS

This method meets most requirements for a scientifically valid method. However, the method and final report have several deficiencies:

On page 50 of the report, the chromatogram for the parent compound, AE1170437, in unfortified California soil shows a significant peak matching the R_f for AE1170437. The presence of the peak suggests the soil was contaminated (in the field or lab), a microbe found in the soil manufactures AE1170437, or the LC-MS instrument was contaminated from a previous analysis. Furthermore, the "native" concentration was not added to the formula mentioned in the above paragraph for the calculation of the MDL and LOQ. An examination of the chromatograms for the unfortified soil sample and the 1.5 ppb fortification experiments revealed that the signal-to-noise ratios ranged from 95-165, with one outlier at 14 corresponding to the standard mentioned earlier in this paragraph. Assuming error in the amount of AE1170437 present in the unfortified soil sample, the LOQ could be reduced by at least a factor of 9 to achieve a 10:1 signal-to-noise ratio. However, determining the actual LOQ was not attempted.

Matrix characterization was not present in the registrant's method.

This method was determined to be acceptable with minor revisions.

COMPLIANCE

Signed and dated statements that this method was exempt from being conducted in accordance with the requirements for Good Laboratory Practice Standards, 40 CFR 160 were present. The claim was "According to 40 CFR Part 160.3, this is not an experimental study. Therefore, the rules and regulations set forth under 40 CFR Part 160 do not apply." A statement of non-confidentiality on the basis of the method falling within the scope of FIFRA Section 10 (d)(1)(A), (B), or (C) was present.

A. BACKGROUND INFORMATION

AE1170437 is a non-selective residual herbicide currently being developed by Bayer CropScience

TABLE A.1. Test Compound Nomenclature for AE1170437	
Compound -	Chemical Structure ***See Appendix B for the chemical structure information**
Common name	AE1170437
Company experimental name	AE1170437
IUPAC name	N-[(1R,2S)-2,3-dihydro-2,6-dimethyl-1H-inden-1-yl]-6-(1-fluoroethyl)-1,3,5-triazine-2,4-diamine
CAS Name	N-[(1R,2S)-2,3-Dihydro-2,6-dimethyl-1H-inden-1-yl]-6-[(1R)-1-fluoroethyl]-1,3,5-triazine-2,4-diamine
CAS #	950782-86-2

TABLE A.2. Physicochemical Properties of the Technical Grade Test Compound	
Parameter	Value
Melting point/range	Not available
pH	Not available
Density	Not available
Water solubility (25 °C)	Not available
Solvent solubility (mg/ml at 20 °C)	Not available
Vapour pressure	Not available
Dissociation constant (pK_a)	Not available
Octanol/water partition coefficient	Not available
UV/visible absorption spectrum	Not available

MATERIALS AND METHODS

B.1. Principle of Method

A 15 g soil or sediment sample is extracted with an acetonitrile/water mixture using microwave digestion, and an aliquot of the supernatant is centrifuged and diluted with water. It then undergoes ESI-LC/MS/MS analysis.

TABLE B.1.1.	**Summary Parameters for the Analytical Method Used for the Quantitation of Chemical Residues in Matrices Studied**
Method ID	ECM0245S1-S6
Analyte(s)	AE1170437 (AE1170437), AE2158969, AE2158968, AE2300077, BCS-AA10201, and AE1170437 Diaminotriazine (1-Fluoroethyl triazinediamine)
Extraction solvent/technique	Soil or sediment sample extracted with acetonitrile/water mixture by microwave digestion.
Cleanup strategies	Centrifugation and dilution
Instrument/Detector	LC/MS/MS – Schimadzu HPLC + PE Sciex API 4000 Mass Analyzer

C. RESULTS AND DISCUSSION

C.1.Recovery Results Summary

TABLE C.1.1. Recovery Results from Method Validation of Soil and Sediment Samples			
Matrix	Spiking Level (conc. units)	Mean% Recoveries	Relative Standard Deviation
***See Appendix A**			

C.1.2. Method Characteristics

TABLE C.1.2. Method Characteristics	
Analyte	AE1170437 (AE1170437), AE2158969, AE2158968, AE2300077, BCS-AA10201, and AE1170437 Diaminotriazine (1-Fluoroethyl triazinediamine)
Limit of Quantitation	Estimated at 1.5 µg/kg
Limit of Detection (LOD)	Calculated at 0.3 µg/kg
Accuracy/Precision at LOQ	***See Appendix A**
Reliability of the Method/ [ILV]	An ILV was performed for this method. MRID No. 47443278
Linearity	Linear curves were prepared for each analyte. The correlation of coefficient for all metabolites were $r > 0.999$
Specificity	The method is very specific due to the use of MS-MS and isotopic internal standards, which is the most highly specific method for detection of residues at low conc. Two transitional ions were also suggested during analysis for additional peak confirmation.

C.2. Independent Laboratory Validation (ILV)

TABLE C.2.1. Recovery Results Obtained by an Independent Laboratory Validation of the Method for the Determination of AE1170437 and its Metabolites in Soil and Sediment

Matrix	Spiking Level (conc. units)	Recoveries
*** See Appendix C**		

D. CONCLUSION

This is a well documented method that was confirmed by an independent laboratory validation. This method is acceptable with minor revisions to the final report and method.

APPENDIX A

Mean Recovery Data for AE1170437 and its metabolites AE1170437 Carboxylic Acid (AE2158969), AE1170437 Triazine-indanone (AE2158968), AE1170437 Hydroxyethyl (AE2300077), AE1170437 Olefin (BCS-AA10201), and AE1170437 Diaminotriazine (1-Fluoroethyl triazinediamine) in Soil.

Compound	Spike Level, ppb	Individual Recoveries, %	Mean ± stdev (RSD)%
AE1170437	1.5	108, 109, 107, 108, 116, 105, 108, 109, 107, 103, 126, 107, 109, 109	109 ± 5.6 (5.1)
	15	112, 109, 109, 110, 109, 110	110 ± 1.2 (1.1)
AE1170437 Diaminotriazine	1.5	104, 103, 100, 101, 102, 104, 104, 103, 105, 99, 103, 107, 103, 104	103 ± 2.1 (2.0)
	15	106, 105, 103, 103, 102, 102	104 ± 1.6 (1.5)
AE1170437 Hydroxyethyl	1.5	94, 96, 94, 92, 94, 91, 93, 93, 95, 95, 98, 98, 98, 95	94.6 ± 2.2 (2.3)
	15	103, 102, 102, 106, 103, 104	103 ± 1.5 (1.5)
AE1170437 Carboxylic Acid	1.5	97, 96, 92, 95, 96, 98, 98, 93, 95, 92, 97, 101, 99, 100	96.2 ± 2.8 (2.9)
	15	103, 101, 101, 100, 98, 100	100 ± 1.8 (1.8)
AE1170437 Triazine-indanone	1.5	101, 100, 102, 102, 102, 99, 101, 102, 103, 97, 101, 104, 103, 106	102 ± 2.3 (2.3)
	15	111, 104, 105, 105, 105, 103	106 ± 2.8 (2.7)
AE1170437 Olefin	1.5	95, 95, 93, 97, 92, 92, 96, 98, 99, 100, 96, 105, 102, 99	97.0 ± 3.8 (3.9)
	15	105, 100, 100, 105, 102, 102	102 ± 2.3 (2.2)

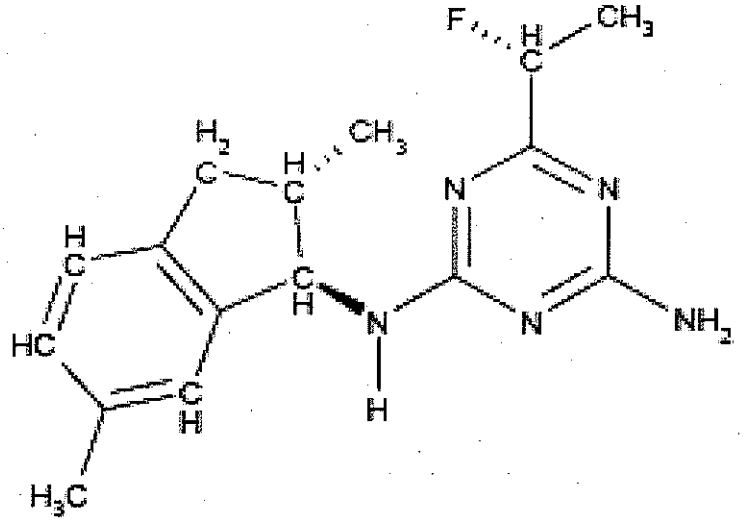
APPENDIX A (continued)

Mean Recovery Data for AE1170437 and its metabolites AE1170437 Carboxylic Acid (AE2158969), AE1170437 Triazine-indanone (AE2158968), AE1170437 Hydroxyethyl (AE2300077), AE1170437 Olefin (BCS-AA10201), and AE1170437 Diaminotriazine (1-Fluoroethyl triazinediamine) in Sediment.

Analyte	Spike Level, ppb	Individual Recoveries, %	Mean ± stdev (RSD)%
AE1170437	1.5	106, 107, 107, 105, 105, 105, 108	106 ±1.2 (1.1)
	15	112, 110, 109	110 ±1.5 (1.4)
AE1170437 Diaminotriazine	1.5	100, 102, 99, 100, 98, 101, 103	101 ±1.6 (1.6)
	15	103, 103, 100	102 ±2.0 (2.0)
AE1170437 Hydroxyethyl	1.5	92, 96, 93, 91, 90, 90, 93	92.0 ±2.4 (2.6)
	15	102, 100, 99	100 ±1.5 (1.5)
AE1170437 Carboxylic Acid	1.5	96, 98, 95, 94, 92, 95, 98	95.4 ± 2.1 (2.2)
	15	104, 101, 97	101 ±3.4 (3.4)
AE1170437 Triazine-indanone	1.5	102, 104, 101, 101, 102, 100, 103	102 ±1.3 (1.3)
	15	106, 106, 105	106 ±0.6 (0.6)
AE1170437 Olefin	1.5	90, 97, 94, 96, 93, 97, 96	94.6 ±2.6 (2.7)
	15	105, 101, 99	102 ±3.1 (3.0)

APPENDIX B

Chemical Structure of AE1170437



APPENDIX C

Independent Laboratory Validation Results for the Recoveries of AE1170437 and Its Metabolites from Two German Soils, Höfchen (silt loam) and Laacher Hof (sandy loam), Using Method DH-002-S06-01.

Substance	Höfchen		Laacher Hof	
	Overall mean [%]	RSD [%]	Overall mean [%]	RSD [%]
AE1170437	96	6.2	96	5.8
AE1170437 Carboxylic Acid	95	4.8	95	4.4
AE1170437 Triazine-indanone	97	4.8	97	5.2
AE1170437 Hydroxyethyl	96	5.6	96	3.0
AE1170437 Olefin	96	6.3	96	5.8
AE1170437 Diaminotriazine	96	4.2	95	5.5