Test Material: Spinetoram (XDE-175)

MRID: 46695143

Title: Method Validation Report for the Determination of XDE-175 and its Metabolites in Soil and Sediment using Dow AgroSciences Methods GRM 05.01 and GRM 05.02

MRID: 46695144

Title: Independent Laboratory Validation of Dow AgroSciences Method GRM 05.02 - Determination of Residues of XDE-175 and its Metabolites in Soil and Sediment by On-Line Solid Phase Extraction and Liquid Chromatography with Tandem Mass Spectrometry

EPA PC Code: 110009

OCSPP Guideline: 850.6100

For CDM Smith

Primary Reviewer: Lisa Muto **Signature:** Lisa Muto

Date: 9/8/15

Secondary Reviewer: Kathleen Ferguson **Signature:** Kathleen P. Ferguson

Date: 9/8/15

QC/QA Manager: Joan Gaidos **Signature:**

Date: 9/8/15

Analytical method for spinetoram [XDE-175 (XDE-175-J and XDE-175-L)] and its transformation products, XDE-175-N-demethyl-J and XDE-175-N-demethyl-L, in soil and sediment

Reports: ECM: EPA MRID No.: 46695143. Hastings, M.J. 2005. Method Validation Report for the Determination of XDE-175 and its Metabolites in Soil and Sediment using Dow AgroSciences Methods GRM 05.01 and GRM 05.02. Laboratory Study ID: 041020. Report prepared, sponsored and submitted by Regulatory Laboratories – Indianapolis Lab, Dow AgroSciences LLC, Indianapolis, Indiana; 127 pages. Final report and Methods dated May 26, 2005 (pp. 1, 26, 71).

ILV: EPA MRID No. 46695144. Kalvan, H.C. 2005. Independent Laboratory Validation of Dow AgroSciences Method GRM 05.02 - Determination of Residues of XDE-175 and its Metabolites in Soil and Sediment by On-Line Solid Phase Extraction and Liquid Chromatography with Tandem Mass Spectrometry. Mogi Mirim Regulatory Laboratory Report No.: GHB-P 1116. Dow AgroSciences PTR No.: 10000943-5008-1 and Study No.: 050036. Report prepared by Mogi Mirim Regulatory Laboratory, Dow AgroSciences Ind. Ltda., Mogi Mirim, Brazil, and sponsored and submitted by Dow AgroSciences LLC, Indianapolis, Indiana; 145 pages. Final report issued September 12, 2005.

Document No.: MRIDs 46695143 & 46695144

Guideline: 850.6100

Statements: ECM: The study was conducted in accordance with USEPA FIFRA and OECD Good Laboratory Practice (GLP) standards (1998; p. 3 of MRID 46695143). Signed and dated No Data Confidentiality, GLP, and Quality Assurance statements were provided (pp. 2-4). A statement of the authenticity of the study report was included with the quality assurance statement (p. 4).

ILV: The study was conducted in accordance with USEPA and OECD GLP standards (1998; p. 3 of MRID 46695144). Signed and dated No Data Confidentiality, GLP, Quality Assurance and Authenticity statements were provided (pp. 2-5).

Classification: This analytical method is classified as supplemental. Method GRM 05.01 was not attempted or validated by the ILV; only GRM 05.02 was validated. The soil matrices were insufficiently characterized in the ECM and ILV; it could not be determined if the ILV was provided with the most difficult soil type with which to validate the methods. In the ECM, the number of samples was insufficient for all analyses, and three of the individual recoveries in the ECM were <70%. ECM representative chromatograms were inadequate to support the methods. Sample recoveries were corrected in the ILV.

PC Code: 110009

Reviewer:

Larry Liu Liu

Date: 5/3/17

All page numbers refer to those listed in the upper right-hand corner of the MRIDs.

Executive Summary

The analytical methods, Dow AgroSciences Methods GRM 05.01 and GRM 05.02, are designed for the quantitative determination of XDE-175 (XDE-175-J and XDE-175-L) and its transformation products (XDE-175-N-demethyl-J and XDE-175-N-demethyl-L) in soil and sediment at the LOQ of 0.005 μg/g using LC/MS/MS. Analytical method GRM 05.02 also used on-line SPE purification. Both methods GRM 05.01 and GRM 05.02 employed a quantitative and a confirmatory HPLC analysis. The LOQ is less than the lowest toxicological level of concern in soil for all analytes. Method GRM 05.02 was validated by the ILV in the first trial; method GRM 05.01 was not attempted. The soil matrices were insufficiently characterized in the ECM and ILV; it could not be determined if the ILV was provided with the most difficult soil type with which to validate the methods. The ILV validated the method GRM 05.02 with “a loamy soil and a clay sediment”. In the ECM, the number of samples was insufficient for all analyses, and representative chromatograms from only one matrix per HPLC analysis were included to support each of the methods.

Table 1. Analytical Method Summary

Analyte(s) by Pesticide	MRID		EPA Review	Matrix	Method Date (dd/mm/yyyy)	Registrant	Analysis	Limit of Quantitation (LOQ)
	Environmental Chemistry Method	Independent Laboratory Validation						
XDE-175-J	46695143 (GRM 05.01)	None[1]		Soil[2,3]	26/05/2005	Dow AgroSciences LLC	LC/MS/MS[4]	0.005 µg/g
XDE-175-L								
XDE-175-N-demethyl-J								
XDE-175-N-demethyl-L								
XDE-175-J	46695143 (GRM 05.02)	46695144					LC/MS/MS with on-line SPE[4]	
XDE-175-L								
XDE-175-N-demethyl-J								
XDE-175-N-demethyl-L								

1 The submitted ILV only validated method GRM 05.02; GRM 05.01 was not performed in the ILV.

2 For ECM Methods GRM 05.01 and GRM 05.02, loam soil, loamy sand soil, clay loam soil, silty clay loam soil, loamy sand sediment and sandy clay loam sediment were minimally characterized (p. 10 of MRID 46695143). USDA classifications were reported, but particle percentages were not reported. Organic carbon percentages were reported.

3 The matrices of the ILV were loamy soil and clay sediment (p. 14 of MRID 46695144). Characterization for two unspecified soil matrices were provided (texture classification was not specified as USDA; soil origin Mogi Mirim Experimental Station; Appendix B, pp. 121-123). Reported soil characterization data were 20.32-21.04% clay and 11.54-15.06 g/dm^3 organic matter. It could not be determined if the characterization data applied to the soil matrices of the ILV validation.

4 Methods GRM 05.01 and GRM 05.02 both used liquid chromatography with positive-ion APCI tandem mass spectrometry analysis; however, GRM 05.02 also used on-line SPE purification (pp. 13-14 of MRID 46695143).

I. Principle of the Method

Samples (5.0 ± 0.05 g) of soil in amber 40-mL vials were fortified, as necessary, then extracted twice with methanol:0.1N sodium hydroxide (90:10, v:v; 20 mL then 15 mL) via shaking on a flat-bed shaker for 60 minutes at *ca*. 180 excursions/minute (p. 13; Appendix A, p. 35; Appendix B, pp. 80-81 of MRID 46695143). The method noted that samples should be protected from light either with amber glass vials or aluminum foil-wrapped vials. Solids and solvent were separated using centrifugation (5 minutes at 2000 rpm). The volume of the combined extracts was adjusted to 40 mL using methanol:0.1N sodium hydroxide (90:10, v:v). Separate aliquots of the extraction solvent were removed for further processing via GRM 05.01 or GRM 05.02.

ECM Method GRM 05.01 – suitable for post-registration residue monitoring

An aliquot (1.0 mL) of the extraction solvent was diluted with 10 mL of 10% sodium chloride then partitioned with 5.0 mL methyl tert-butyl ether (MTBE; p. 13; Appendix A, pp. 35-36 of MRID 46695143). After shaking on a flat-bed shaker for 5 minutes at *ca*. 180 excursions/minute, the sample was centrifuged for 5 minutes at 2000 rpm. The MTBE layer was isolated and mixed with 100 µL of methanol:glycerine (80:20, v:v; the method noted that none of the aqueous layer should be transferred with the MTBE layer). The MTBE was evaporated to near dryness (*ca*. 20 µL of glycerine remaining) using a dry block heater set at 40°C under nitrogen. The method noted that a Turbovap evaporator should not be used and that the extract should not be evaporated to dryness. The residue was reconstituted in 980 µL of methanol:acetonitrile:water (35:35:30, v:v:v). The final solution was analyzed by liquid chromatography with positive-ion atmospheric pressure chemical ionization (APCI) tandem mass spectrometry.

Samples were analyzed for XDE-175-J, XDE-175-L, XDE-175-N-demethyl-J and XDE-175-N-demethyl-L using a MDS/SCIEX API 4000 LC/MS/MS (p. 14; Appendix A, pp. 33-35 of MRID 46695143). The instrumental conditions consisted of a YMC ODS-AM column (4.6 x 50 mm, 5-µm; column temperature ambient), a mobile phase gradient of (A) acetonitrile:methanol (1:1, v:v) containing 10 mM ammonium acetate and (B) water containing 10 mM ammonium acetate [percent A:B (v:v) at 0:01 min. 70:30, 5:00-5:30 min. 100:0, 5:45-7:00 min. 70:30] and MS/MS detection in positive APCI ionization mode (temperature, 425°C), and injection volume 30 µL. One parent-daughter ion transition was monitored per analyte: *m/z* 748.6 → 142.2 for XDE-175-J, *m/z* 760.9 → 142.2 for XDE-175-L, *m/z* 734.9 → 128.2 for XDE-175-N-demethyl-J, and *m/z* 746.7 → 128.2 for XDE-175-N-demethyl-L. Confirmatory HPLC analysis was performed using a Synergi Polar RP column (4.6 x 75 mm, 4-µm). Retention times for XDE-175-J, XDE-175-L, XDE-175-N-demethyl-J and XDE-175-N-demethyl-L were observed at 5.3, 5.6, 4.0 and 4.3 min., respectively, for quantitative HPLC and at 4.7, 4.8, 4.4 and 4.5 min., respectively, for confirmatory HPLC (Appendix A, Figures 13-20, pp. 62-69).

ECM Method GRM 05.02 – developed for pre-registration data generation (field dissipation analysis)

An aliquot (250 µL) of the extraction solvent (see above) was transferred to a 96-well plate (pp. 13-14; Appendix B, pp. 78-81; Appendix B, Appendix 1, pp. 121-127 of MRID 46695143). 25 µL of the 0.1 µg/mL mixed XDE-175 and metabolites stable isotope internal standard solution was added to each sample. The mixture was diluted with 250 µL of water:glacial acetic acid (99.5:0.5, v:v). The samples were purified by on-line solid phase extraction using a strong cation exchange (SCX) cartridge (40-90 µm). The SPE cartridge was pre-conditioned with water (3 mL), methanol (2 mL) and methanol:acetonitrile:water (40:40:20, v:v:v) containing 0.1M ammonium acetate (0.2 mL). The analytes were eluted with 0.2 mL of methanol:acetonitrile:water (4:4:2, v:v:v) containing 0.1M ammonium acetate onto the LC analytical column. XDE-175 and its metabolites were analyzed by liquid chromatography with positive-ion APCI tandem mass spectrometry.

Samples were analyzed for XDE-175-J, XDE-175-L, XDE-175-N-demethyl-J and XDE-175-N-demethyl-L and their internal stable isotope standards (IS) using a MDS/SCIEX API 4000 LC/MS/MS (p. 14; Appendix B, pp. 78-80 of MRID 46695143). The instrumental conditions consisted of a YMC ODS-AM column (4.6 x 50 mm, 5-µm; column temperature ambient), a mobile phase gradient of (A) acetonitrile:methanol (1:1, v:v) containing 10 mM ammonium acetate and (B) water containing 10 mM ammonium acetate [percent A:B (v:v) at 0:01-1:05 min. 70:30, 3:05-5:00 min. 100:0, 5:15-6:15 min. 70:30] and MS/MS detection in positive APCI ionization mode (temperature, 425°C), and injection volume 30 µL. One parent-daughter ion transition was monitored per analyte or IS: *m/z* 748.6 → 142.2 for XDE-175-J, *m/z* 760.9 → 142.2 for XDE-175-L, *m/z* 734.9 → 128.2 for XDE-175-N-demethyl-J, and *m/z* 746.7 → 128.2 for XDE-175-N-demethyl-L; and *m/z* 757.9 → 146.2 for XDE-175-J IS, *m/z* 769.9 → 146.2 for XDE-175-L IS, *m/z* 739.9 → 128.2 for XDE-175-N-demethyl-J IS, and *m/z* 751.7 → 128.2 for XDE-175-N-demethyl-L IS. Confirmatory HPLC analysis was performed using a Synergi Polar RP column (4.6 x 75 mm, 4-µm). Retention times for XDE-175-J, XDE-175-L, XDE-175-N-demethyl-J and XDE-175-N-demethyl-L were observed at 4.2 (4.1), 4.3, 3.6 and 3.7-3.8 (3.7) min., respectively, for quantitative HPLC and at 3.8, 3.9, 3.6 and 3.7 min., respectively, for confirmatory HPLC (RT for the IS was reported in parenthesis if it differed from the RT of the analyte; Appendix B, Figures 21-28, pp. 113-120).

ILV

In the ILV, only Method GRM 05.02 was performed; Method GRM 05.01 was not included in the ILV (pp. 13-14 of MRID 46695144). Method GRM 05.02 was performed exactly as written (pp. 16-20). Samples were analyzed for XDE-175-J, XDE-175-L, XDE-175-N-demethyl-J and XDE-175-N-demethyl-L and their internal stable isotope standards (IS) using a MDS/SCIEX API 4000 LC/MS/MS with on-line SPE. The LC instrument and instrumental conditions were the same as that of the ECM, except that the injection volume was 10 µL (partial loopfull – flexibility mode). One parent-daughter ion transition was monitored per analyte or IS: *m/z* 748.5 → 142.2 for XDE-175-J, *m/z* 760.5 → 142.2 for XDE-175-L, *m/z* 734.5 → 128.2 for XDE-175-N-demethyl-J, and *m/z* 746.5 → 128.2 for XDE-175-N-demethyl-L; and *m/z* 757.5 → 146.2 for XDE-175-J IS, *m/z* 769.6 → 146.3 for XDE-175-L IS, *m/z* 739.6 → 128.2 for XDE-175-N-demethyl-J IS, and *m/z* 751.4 → 128.3 for XDE-175-N-demethyl-L IS. These monitored transitions were very similar to those of the ECM. Confirmatory HPLC analysis was also performed using a Synergi Polar RP column (4.6 x 75 mm, 4-µm), as in the ECM. Retention times for XDE-175-J, XDE-175-L, XDE-175-N-demethyl-J and XDE-175-N-demethyl-L were observed at *ca*. 4.0-4.1, 4.2, 3.4-3.5 and 3.6-3.7 min., respectively, for quantitative HPLC and at *ca*. 3.8, 3.8-3.9, 3.5-3.6 and 3.6-3.7 min., respectively, for confirmatory HPLC (IS and analyte;

Figures 3-20, pp. 44-61). The study author noted that the "samples must be protected from bright light during extraction" using one of the methods described in GRM 05.01 and GRM 05.02 (p. 26).

LOQ/LOD

The LOQ for all analytes was the same in the ECM Methods and ILV at 0.005 µg/g (pp. 13, 20-21; Appendix A, Table 7, p. 53; Appendix B, Table 7, p. 100 of MRID 46695143; pp. 13, 25; Table 11, p. 40 of MRID 46695144). The LOD for all analytes was 0.0015 µg/g in the ECM Methods. The LOD was not reported in the ILV, but values for LOD were calculated.

II. Recovery Findings

ECM (MRID 46695143: Methods GRM 05.01 and GRM 05.02): Individual recoveries from GRM 05.01 and GRM 05.02 were 70-120% for analysis of XDE-175-J, XDE-175-L, XDE-175-N-demethyl-J and XDE-175-N-demethyl-L in four soils (loam, loamy sand, clay loam and silty clay loam) and two sediments (sandy clay loam and loamy sand) at fortification levels of 0.005 µg/g (LOQ), 0.05 µg/g (10×LOQ), 0.50 µg/g (100×LOQ) and 1.00 µg/g (200×LOQ), except for two recoveries of XDE-175-J in silty clay loam soil (GRM 05.01, 68%; GRM 05.02, 66%) and one recovery of XDE-175-L in silty clay loam soil (GRM 05.02, 63%; Appendix A, Tables 2-5, pp. 45-52; Appendix B, Tables 2-5, pp. 92-99). The number of samples (n = 2) was insufficient for all analyses in both methods. Mean recoveries and relative standard deviations (RSDs) of statistical significance could not be calculated. Performance data (recovery results) were only provided for quantitative HPLC analysis; recovery results from the confirmatory HPLC analysis were not reported. Confirmation of residue identity was performed by comparing the retention times of the analytes in the samples to the analytes in the calibration standards; the method noted that the confirmatory HPLC analysis could be performed, if necessary, since only one ion transition was monitored per analyte (pp. 21-22). Both Methods GRM 05.01 and GRM 05.02 allowed for recovery data to be corrected for residues found in the control samples; however, residues were not quantified in any of the control samples (Appendix A, pp. 37-39; Appendix A, Tables 2-5, pp. 45-52; Appendix B, pp. 82-85; Appendix B, Tables 2-5, pp. 92-99). Recoveries from samples fortified at 0.0015 µg/g (LOD) ranged (methods and matrices combined) from 80-120% for XDE-175-J, 87-127% for XDE-175-L, 87-133% for XDE-175-N-demethyl-J and 80-120% for XDE-175-N-demethyl-L (n = 1 for each matrix/analyte; DER Attachment 2). The soil/sediment matrices were minimally characterized; the sources were not reported (p. 10). USDA classifications were reported, but particle percentages were not reported. Organic carbon percentages were 0.6%, 1.3%, 5.0%, 1.2%, 2.4% and 2.8% for loam soil, loamy sand soil, clay loam soil, silty clay loam soil, loamy sand sediment and sandy clay loam sediment, respectively.

ILV (MRID 46695144: Method GRM 05.02 only): Mean recoveries and relative standard deviations (RSDs) were within guidelines for analysis of XDE-175-J, XDE-175-L, XDE-175-N-demethyl-J and XDE-175-N-demethyl-L in loamy soil and clay sediment at fortification levels of 0.005 µg/g (LOQ) and 0.05 µg/g (10×LOQ) using the quantitative and confirmatory HPLC analyses (Tables 1-2, pp. 30-31). Performance data (recovery results) from quantitative HPLC analyses yielded higher mean recoveries than confirmatory HPLC analyses, but RSDs were comparable between the quantitative and confirmatory HPLC analyses. The study author determined that a “carryover effect for the quantitation column” was responsible for the “differences in the recovery ranges for the quantitation and the confirmation injections” (pp. 25-26). Calculations allowed for recovery data to be corrected for residues found in the control samples (pp. 22-24; Tables 3-10, pp. 32-39). Residues ranged 0.0000-0.0008 µg/g in the control samples of the quantitative HPLC analysis and 0.000-0.001 µg/g in the control samples of the confirmatory HPLC analysis. “A loamy soil and a clay sediment” were used for validation (p. 14). Characterization for two unspecified soil matrices were provided (texture classification was not specified as USDA; soil origin Mogi Mirim Experimental Station; Appendix B, pp. 121-123). Reported soil characterization data were 76.24% sand, 2.72% silt, 21.04% clay; 11.54 g/dm^3 organic matter for Sample 051329 Soil and 77.68% sand, 2.00% silt, 20.32% clay; 15.06 g/dm^3 organic matter for Sample 051330 Soil. It could not be determined if this characterization data applied to the matrices of the ILV validation. Method GRM 05.02 was validated in the first trial; Method GRM 05.01 was not attempted (pp. 13-14, 24).

Table 2. Initial Validation Method Recoveries for XDE-175 and Its Transformation Products in Four Soils and Two Sediments[1,2]

Analyte	Fortification Level (µg/g)	Number of Tests	Recovery Range (%)	Mean Recovery (%)	Standard Deviation (%)	Relative Standard Deviation (%)
GRM 05.01 - Quantitative HPLC						
	Loam Soil					
XDE-175-J	0.0015 (LOD)	1	93	--	--	--
	0.005 (LOQ)	2	83, 89	--	--	--
	0.05	2	88, 98	--	--	--
	0.50	2	88, 89	--	--	--
	1.00	2	89, 104	--	--	--
XDE-175-L	0.0015 (LOD)	1	93	--	--	--
	0.005 (LOQ)	2	88, 92	--	--	--
	0.05	2	83, 92	--	--	--
	0.50	2	89	--	--	--
	1.00	2	87, 95	--	--	--
XDE-175-N-demethyl-J	0.0015 (LOD)	1	93	--	--	--
	0.005 (LOQ)	2	89, 91	--	--	--
	0.05	2	91	--	--	--
	0.50	2	91, 94	--	--	--
	1.00	2	91, 99	--	--	--
XDE-175-N-demethyl-L	0.0015 (LOD)	1	93	--	--	--
	0.005 (LOQ)	2	85, 91	--	--	--
	0.05	2	87, 90	--	--	--
	0.50	2	93, 96	--	--	--
	1.00	2	86, 98	--	--	--
	Loamy Sand Soil					
XDE-175-J	0.0015 (LOD)	1	93	--	--	--
	0.005 (LOQ)	2	89, 95	--	--	--
	0.05	2	96, 98	--	--	--
	0.50	2	94, 116	--	--	--
	1.00	2	95, 98	--	--	--
XDE-175-L	0.0015 (LOD)	1	93	--	--	--
	0.005 (LOQ)	2	89, 91	--	--	--
	0.05	2	85, 90	--	--	--
	0.50	2	91, 107	--	--	--
	1.00	2	84, 90	--	--	--
XDE-175-N-demethyl-J	0.0015 (LOD)	1	87	--	--	--
	0.005 (LOQ)	2	91, 92	--	--	--
	0.05	2	95, 97	--	--	--
	0.50	2	98, 112	--	--	--
	1.00	2	92, 94	--	--	--

Analyte	Fortification Level (µg/g)	Number of Tests	Recovery Range (%)	Mean Recovery (%)	Standard Deviation (%)	Relative Standard Deviation (%)
XDE-175-N-demethyl-L	0.0015 (LOD)	1	93	--	--	--
	0.005 (LOQ)	2	93, 95	--	--	--
	0.05	2	92	--	--	--
	0.50	2	91, 111	--	--	--
	1.00	2	93, 96	--	--	--
	Sandy Clay Loam Sediment					
XDE-175-J	0.0015 (LOD)	1	87	--	--	--
	0.005 (LOQ)	2	78, 91	--	--	--
	0.05	2	83, 84	--	--	--
	0.50	2	81, 90	--	--	--
	1.00	2	78, 82	--	--	--
XDE-175-L	0.0015 (LOD)	1	93	--	--	--
	0.005 (LOQ)	2	77, 84	--	--	--
	0.05	2	77, 81	--	--	--
	0.50	2	80, 87	--	--	--
	1.00	2	75, 82	--	--	--
XDE-175-N-demethyl-J	0.0015 (LOD)	1	93	--	--	--
	0.005 (LOQ)	2	85, 86	--	--	--
	0.05	2	84	--	--	--
	0.50	2	87, 95	--	--	--
	1.00	2	85, 86	--	--	--
XDE-175-N-demethyl-L	0.0015 (LOD)	1	87	--	--	--
	0.005 (LOQ)	2	83, 95	--	--	--
	0.05	2	83, 85	--	--	--
	0.50	2	86, 90	--	--	--
	1.00	2	82, 87	--	--	--
	Loamy Sand Sediment					
XDE-175-J	0.0015 (LOD)	1	107	--	--	--
	0.005 (LOQ)	2	92, 99	--	--	--
	0.05	2	97, 99	--	--	--
	0.50	2	96, 102	--	--	--
	1.00	2	105, 116	--	--	--
XDE-175-L	0.0015 (LOD)	1	107	--	--	--
	0.005 (LOQ)	2	92, 93	--	--	--
	0.05	2	97, 114	--	--	--
	0.50	2	101, 102	--	--	--
	1.00	2	102, 104	--	--	--
XDE-175-N-demethyl-J	0.0015 (LOD)	1	100	--	--	--
	0.005 (LOQ)	2	95, 98	--	--	--

Analyte	Fortification Level (µg/g)	Number of Tests	Recovery Range (%)	Mean Recovery (%)	Standard Deviation (%)	Relative Standard Deviation (%)
	0.05	2	96, 99	--	--	--
	0.50	2	103,107	--	--	--
	1.00	2	95, 101	--	--	--
XDE-175-N-demethyl-L	0.0015 (LOD)	1	80	--	--	--
	0.005 (LOQ)	2	92, 93	--	--	--
	0.05	2	94, 98	--	--	--
	0.50	2	98, 101	--	--	--
	1.00	2	93	--	--	--
	Clay Loam Soil					
XDE-175-J	0.0015 (LOD)	1	113	--	--	--
	0.005 (LOQ)	2	97, 108	--	--	--
	0.05	2	93, 101	--	--	--
	0.50	2	111, 112	--	--	--
	1.00	2	98, 100	--	--	--
XDE-175-L	0.0015 (LOD)	1	100	--	--	--
	0.005 (LOQ)	2	95, 104	--	--	--
	0.05	2	106, 115	--	--	--
	0.50	2	93, 104	--	--	--
	1.00	2	102, 103	--	--	--
XDE-175-N-demethyl-J	0.0015 (LOD)	1	100	--	--	--
	0.005 (LOQ)	2	93, 102	--	--	--
	0.05	2	97, 101	--	--	--
	0.50	2	102, 104	--	--	--
	1.00	2	101, 102	--	--	--
XDE-175-N-demethyl-L	0.0015 (LOD)	1	100	--	--	--
	0.005 (LOQ)	2	100, 102	--	--	--
	0.05	2	95, 98	--	--	--
	0.50	2	101, 102	--	--	--
	1.00	2	94, 97	--	--	--
	Silty Clay Loam Soil					
XDE-175-J	0.0015 (LOD)	1	113	--	--	--
	0.005 (LOQ)	2	88	--	--	--
	0.05	2	83, 87	--	--	--
	0.50	2	84	--	--	--
	1.00	2	**68**, 82	--	--	--
XDE-175-L	0.0015 (LOD)	1	87	--	--	--
	0.005 (LOQ)	2	80, 82	--	--	--
	0.05	2	78, 104	--	--	--
	0.50	2	71, 75	--	--	--
	1.00	2	70	--	--	--

Analyte	Fortification Level (µg/g)	Number of Tests	Recovery Range (%)	Mean Recovery (%)	Standard Deviation (%)	Relative Standard Deviation (%)
XDE-175-N-demethyl-J	0.0015 (LOD)	1	93	--	--	--
	0.005 (LOQ)	2	91, 92	--	--	--
	0.05	2	91, 95	--	--	--
	0.50	2	89, 93	--	--	--
	1.00	2	73, 78	--	--	--
XDE-175-N-demethyl-L	0.0015 (LOD)	1	80	--	--	--
	0.005 (LOQ)	2	88, 92	--	--	--
	0.05	2	89, 90	--	--	--
	0.50	2	83, 87	--	--	--
	1.00	2	73, 74	--	--	--
GRM 05.02 – Quantitative HPLC						
	Loam Soil					
XDE-175-J	0.0015 (LOD)	1	107	--	--	--
	0.005 (LOQ)	2	97, 103	--	--	--
	0.05	2	99, 101	--	--	--
	0.50	2	102, 104	--	--	--
	1.00	2	104, 108	--	--	--
XDE-175-L	0.0015 (LOD)	1	127	--	--	--
	0.005 (LOQ)	2	97, 105	--	--	--
	0.05	2	96	--	--	--
	0.50	2	98, 103	--	--	--
	1.00	2	102	--	--	--
XDE-175-N-demethyl-J	0.0015 (LOD)	1	120	--	--	--
	0.005 (LOQ)	2	94, 101	--	--	--
	0.05	2	82, 87	--	--	--
	0.50	2	83, 91	--	--	--
	1.00	2	89, 93	--	--	--
XDE-175-N-demethyl-L	0.0015 (LOD)	1	107	--	--	--
	0.005 (LOQ)	2	88, 96	--	--	--
	0.05	2	85, 86	--	--	--
	0.50	2	85, 86	--	--	--
	1.00	2	95, 100	--	--	--
	Loamy Sand Soil					
XDE-175-J	0.0015 (LOD)	1	113	--	--	--
	0.005 (LOQ)	2	103, 103	--	--	--
	0.05	2	102, 106	--	--	--
	0.50	2	100, 101	--	--	--
	1.00	2	100, 119	--	--	--
XDE-175-L	0.0015 (LOD)	1	120	--	--	--

Analyte	Fortification Level (µg/g)	Number of Tests	Recovery Range (%)	Mean Recovery (%)	Standard Deviation (%)	Relative Standard Deviation (%)
	0.005 (LOQ)	2	97, 106	--	--	--
	0.05	2	99, 103	--	--	--
	0.50	2	93, 104	--	--	--
	1.00	2	104, 118	--	--	--
XDE-175-N-demethyl-J	0.0015 (LOD)	1	133	--	--	--
	0.005 (LOQ)	2	95, 101	--	--	--
	0.05	2	87, 93	--	--	--
	0.50	2	85, 92	--	--	--
	1.00	2	95, 104	--	--	--
XDE-175-N-demethyl-L	0.0015 (LOD)	1	120	--	--	--
	0.005 (LOQ)	2	98, 102	--	--	--
	0.05	2	87, 92	--	--	--
	0.50	2	86, 90	--	--	--
	1.00	2	93, 109	--	--	--
	Sandy Clay Loam Sediment					
XDE-175-J	0.0015 (LOD)	1	120	--	--	--
	0.005 (LOQ)	2	95, 103	--	--	--
	0.05	2	91, 92	--	--	--
	0.50	2	91, 97	--	--	--
	1.00	2	92, 101	--	--	--
XDE-175-L	0.0015 (LOD)	1	100	--	--	--
	0.005 (LOQ)	2	88, 103	--	--	--
	0.05	2	85, 91	--	--	--
	0.50	2	88, 99	--	--	--
	1.00	2	88, 93	--	--	--
XDE-175-N-demethyl-J	0.0015 (LOD)	1	120	--	--	--
	0.005 (LOQ)	2	91, 101	--	--	--
	0.05	2	77, 80	--	--	--
	0.50	2	80, 89	--	--	--
	1.00	2	79, 84	--	--	--
XDE-175-N-demethyl-L	0.0015 (LOD)	1	113	--	--	--
	0.005 (LOQ)	2	88, 91	--	--	--
	0.05	2	76, 77	--	--	--
	0.50	2	82, 88	--	--	--
	1.00	2	75, 79	--	--	--
	Loamy Sand Sediment					
XDE-175-J	0.0015 (LOD)	1	80	--	--	--
	0.005 (LOQ)	2	96, 101	--	--	--
	0.05	2	94, 102	--	--	--

Analyte	Fortification Level (µg/g)	Number of Tests	Recovery Range (%)	Mean Recovery (%)	Standard Deviation (%)	Relative Standard Deviation (%)
	0.50	2	95, 105	--	--	--
	1.00	2	92, 102	--	--	--
XDE-175-L	0.0015 (LOD)	1	127	--	--	--
	0.005 (LOQ)	2	95, 96	--	--	--
	0.05	2	95, 97	--	--	--
	0.50	2	97, 98	--	--	--
	1.00	2	87, 89	--	--	--
XDE-175-N-demethyl-J	0.0015 (LOD)	1	127	--	--	--
	0.005 (LOQ)	2	102, 107	--	--	--
	0.05	2	95, 101	--	--	--
	0.50	2	94, 99	--	--	--
	1.00	2	94, 98	--	--	--
XDE-175-N-demethyl-L	0.0015 (LOD)	1	107	--	--	--
	0.005 (LOQ)	2	102, 103	--	--	--
	0.05	2	89, 97	--	--	--
	0.50	2	95, 100	--	--	--
	1.00	2	94, 96	--	--	--
	Clay Loam Soil					
XDE-175-J	0.0015 (LOD)	1	93	--	--	--
	0.005 (LOQ)	2	90, 92	--	--	--
	0.05	2	89, 97	--	--	--
	0.50	2	98, 101	--	--	--
	1.00	2	93, 97	--	--	--
XDE-175-L	0.0015 (LOD)	1	113	--	--	--
	0.005 (LOQ)	2	88, 102	--	--	--
	0.05	2	92, 100	--	--	--
	0.50	2	90, 96	--	--	--
	1.00	2	99, 103	--	--	--
XDE-175-N-demethyl-J	0.0015 (LOD)	1	107	--	--	--
	0.005 (LOQ)	2	101, 106	--	--	--
	0.05	2	98	--	--	--
	0.50	2	93, 94	--	--	--
	1.00	2	94, 103	--	--	--
XDE-175-N-demethyl-L	0.0015 (LOD)	1	107	--	--	--
	0.005 (LOQ)	2	100, 101	--	--	--
	0.05	2	94, 104	--	--	--
	0.50	2	101, 103	--	--	--
	1.00	2	93, 94	--	--	--
	Silty Clay Loam Soil					

Analyte	Fortification Level (µg/g)	Number of Tests	Recovery Range (%)	Mean Recovery (%)	Standard Deviation (%)	Relative Standard Deviation (%)
XDE-175-J	0.0015 (LOD)	**1**	87	--	--	--
	0.005 (LOQ)	**2**	84, 86	--	--	--
	0.05	**2**	76, 89	--	--	--
	0.50	**2**	88, 94	--	--	--
	1.00	**2**	**66**, 76	--	--	--
XDE-175-L	0.0015 (LOD)	**1**	100	--	--	--
	0.005 (LOQ)	**2**	97, 99	--	--	--
	0.05	**2**	88, 94	--	--	--
	0.50	**2**	89, 95	--	--	--
	1.00	**2**	**63**, 70	--	--	--
XDE-175-N-demethyl-J	0.0015 (LOD)	**1**	120	--	--	--
	0.005 (LOQ)	**2**	100, 102	--	--	--
	0.05	**2**	94, 99	--	--	--
	0.50	**2**	88, 93	--	--	--
	1.00	**2**	76, 78	--	--	--
XDE-175-N-demethyl-L	0.0015 (LOD)	**1**	100	--	--	--
	0.005 (LOQ)	**2**	95, 101	--	--	--
	0.05	**2**	96, 98	--	--	--
	0.50	**2**	95, 96	--	--	--
	1.00	**2**	76, 77	--	--	--

Data (uncorrected recovery results; pp. Appendix A, pp. 37-39; Appendix A, Tables 2-5, pp. 45-52; Appendix B, pp. 82-85; Appendix B, Tables 2-5, pp. 92-99) were obtained from Appendix A, Tables 2-5, pp. 45-52; Appendix B, Tables 2-5, pp. 92-99 of MRID 46695143 and DER Attachment 2 (% recovery at LOD).

1 The soil matrices were partially characterized (p. 10). USDA classifications were reported, but particle percentages were not reported. Organic carbon percentages were 0.6%, 1.3%, 5.0%, 1.2%, 2.4% and 2.8% for loam soil, loamy sand soil, clay loam soil, silty clay loam soil, loamy sand sediment and sandy clay loam sediment, respectively.

2 One ion transition was monitored for each of the analytes (p. 14; Appendix A, pp. 33-35; Appendix B, pp. 78-80). Analytes were analyzed by liquid chromatography with positive-ion APCI tandem mass spectrometry for both quantitative and confirmatory HPLC; different LC columns were used for quantitation and confirmation. GRM 05.02 also used on-line SPE purification. Only results for the quantitative HPLC were reported (pp. 19-22).

Table 3. Independent Validation Method Recoveries for XDE-175 and Its Transformation Products in Loamy Soil and Clay Sediment[1,2]

Analyte	Fortification Level (µg/g)	Number of Tests	Recovery Range (%)	Mean Recovery (%)	Standard Deviation (%)	Relative Standard Deviation (%)
Method GRM 05.02 - Quantitative HPLC						
	Loamy Soil					
XDE-175-J	0.005 (LOQ)	5	108-116	112	3	3
	0.05	5	111-118	115	3	3
XDE-175-L	0.005 (LOQ)	5	86-110	101	10	10
	0.05	5	94-116	103	8	8
XDE-175-N-demethyl-J	0.005 (LOQ)	5	98-118	109	7	7
	0.05	5	102-112	107	4	4
XDE-175-N-demethyl-L	0.005 (LOQ)	5	102-113	108	6	5
	0.05	5	117-120	119	1	1
	Clay Sediment					
XDE-175-J	0.005 (LOQ)	5	95-119	105	10	9
	0.05	5	98-110	105	5	5
XDE-175-L	0.005 (LOQ)	5	107-111	110	2	1
	0.05	5	110-120	116	4	4
XDE-175-N-demethyl-J	0.005 (LOQ)	5	96-116	110	8	7
	0.05	5	95-119	110	10	9
XDE-175-N-demethyl-L	0.005 (LOQ)	5	89-97	92	3	4
	0.05	5	105-119	113	6	5
Method GRM 05.02 - Confirmatory HPLC						
	Loamy Soil					
XDE-175-J	0.005 (LOQ)	5	76-94	86	7	8
	0.05	5	97-102	99	2	2
XDE-175-L	0.005 (LOQ)	5	72-83	76	4	5
	0.05	5	89-100	95	4	5
XDE-175-N-demethyl-J	0.005 (LOQ)	5	87-99	90	5	6
	0.05	5	94-100	96	2	2
XDE-175-N-demethyl-L	0.005 (LOQ)	5	79-92	85	5	6
	0.05	5	93-102	99	4	4
	Clay Sediment					
XDE-175-J	0.005 (LOQ)	5	71-79	75	4	5
	0.05	5	72-80	76	3	4
XDE-175-L	0.005 (LOQ)	5	73-85	78	5	6
	0.05	5	71-80	75	4	5
XDE-175-N-demethyl-J	0.005 (LOQ)	5	71-86	82	6	8
	0.05	5	73-81	78	3	4
XDE-175-N-demethyl-L	0.005 (LOQ)	5	72-91	80	7	9
	0.05	5	74-82	78	3	4

Data (corrected results; pp. 22-24) were obtained from Tables 1-2, pp. 30-31 of MRID 46695144.

[1] "A loamy soil and a clay sediment" were used for validation (p. 14). Characterization for two unspecified soil matrices were provided (texture classification was not specified as USDA; soil origin Mogi Mirim Experimental Station; Appendix B, pp. 121-123).

[2] One ion transition was monitored for each of the analytes (pp. 16-20). Analytes were analyzed by liquid chromatography with positive-ion APCI tandem mass spectrometry for both quantitative and confirmatory HPLC; different LC columns were used for quantitation and confirmation.

III. Method Characteristics

In the ECM Methods and ILV, the LOQ value for XDE-175-J, XDE-175-L, XDE-175-N-demethyl-J and XDE-175-N-demethyl-L were established at 0.005 µg/g (pp. 13, 20-21, 24; Appendix A, Table 7, p. 53; Appendix B, Table 7, p. 100 of MRID 46695143; pp. 13, 25; Table 11, p. 40 of MRID 46695144). The LOD for all analytes in the ECM Methods was 0.0015 µg/g. The LOD was not reported in the ILV, but values for LOD were calculated. Following the method of Keith, L. H., *et al*. (see section **V. References** below), the LOD and LOQ for determination of XDE-175 and its transformation products in soil/sediment were calculated in the ECM Methods using the standard deviation from the 0.005 µg/g recovery results. The LOD was calculated as three times the standard deviation (3*s*), and the LOQ was calculated as ten times the standard deviation (10*s*) of the recovery results. The LOD values were also calculated in the ILV using the same methodology. The calculated values support the LOQ and LOD established for the study and are presented in **Table 4** below.

Table 4. Method Characteristics[1]

			XDE-175-J	XDE-175-L	XDE-175-N-demethyl-J	XDE-175-N-demethyl-L
Limit of Quantitation (LOQ)	Established		0.005 µg/g			
	Calculated	ECM	0.00387 µg/g (GRM 05.01) 0.00329 µg/g (GRM 05.02)	0.00365 µg/g (GRM 05.01) 0.00297 µg/g (GRM 05.02)	0.00230 µg/g (GRM 05.01) 0.00235 µg/g (GRM 05.02)	0.00270 µg/g (GRM 05.01) 0.00279 µg/g (GRM 05.02)
		ILV	0.00395 µg/g (GRM 05.02)	0.00403 µg/g (GRM 05.02)	0.00367 µg/g (GRM 05.02)	0.00478 µg/g (GRM 05.02)
Limit of Detection (LOD)	Established		0.0015 µg/g			
	Calculated	ECM	0.00116 µg/g (GRM 05.01) 0.00099 µg/g (GRM 05.02)	0.00109 µg/g (GRM 05.01) 0.00089 µg/g (GRM 05.02)	0.00069 µg/g (GRM 05.01) 0.00071 µg/g (GRM 05.02)	0.00081 µg/g (GRM 05.01) 0.00084 µg/g (GRM 05.02)
		ILV	0.00118 µg/g (GRM 05.02)	0.00121 µg/g (GRM 05.02)	0.00110 µg/g (GRM 05.02)	0.00143 µg/g (GRM 05.02)
Linearity (Least squares calibration curve r and concentration range)	ECM[2]		$r^2 = 0.9997$ (GRM 05.01) $r^2 = 0.9992$ (GRM 05.02)	$r^2 = 0.9996$ (GRM 05.01) $r^2 = 0.9995$ (GRM 05.02)	$r^2 = 0.9993$ (GRM 05.01) $r^2 = 0.9992$ (GRM 05.02)	$r^2 = 0.9995$ (GRM 05.01) $r^2 = 0.9981$ (GRM 05.02)
	ILV (GRM 05.02)[3]		$r^2 = 0.9998$ (Q) $r^2 = 0.9991$ (C)	$r^2 = 0.9987$ (Q) $r^2 = 0.9982$ (C)	$r^2 = 0.9997$ (Q) $r^2 = 0.9987$ (C)	$r^2 = 0.9981$ (Q) $r^2 = 0.9994$ (C)
	Concentration range		0.1-50 ng/mL			
Repeatable	ECM[4] (GRM 05.01 & GRM 05.02)		Yes at LOQ, 10×LOQ and 100×LOQ, but n = 2.			
			Yes at 200×LOQ, but n = 2, for all matrices/methods except one sample in silty clay loam soil in both methods.	Yes at 200×LOQ, but n = 2, for all matrices/methods except one sample in silty clay loam soil in GRM 05.02.	Yes at 200×LOQ, but n = 2.	
	ILV (GRM 05.02)[5]		Yes at LOQ and 10×LOQ (n = 5).			
Reproducible			Yes at the LOQ and 10×LOQ.			
Specific	ECM		Only chromatograms of loam soil (640) and loamy sand sediment (653) were provided. Only chromatograms of one matrix were provided for each HPLC method: the quantitative and confirmatory HPLC.			
		GRM 05.01	Matrix interferences were *ca*. 30-40% of the LOQ based on peak area estimation (confirmatory HPLC only).	Yes, no interferences were observed in the matrix control.		
		GRM 05.02	Yes, no interferences were observed in the matrix control.			

	ILV (GRM 05.02)	Yes, matrix interferences at the retention times of the analytes were <20% of the LOQ for the quantitative HPLC and <5% of the LOQ for the confirmatory HPLC, based on residue recovery and chromatograms.

Data were obtained from pp. 10, 13, 20-21; Appendix A, Tables 2-5, pp. 45-52; Appendix A, Table 7, p. 53; Appendix A, Figures 9-20, pp. 58-69; Appendix B, Tables 2-5, pp. 92-99; Appendix B, Table 7, p. 100; Appendix B, Figures 17-28, pp. 109-120 of MRID 46695143; pp. 13-14, 25; Tables 1-11, pp. 30-40; Figures 3-20, pp. 44-61; Appendix B, pp. 121-123; Appendix C, pp. 125-140 of MRID 46695144.

1 Methods GRM 05.01 and GRM 05.02 both used liquid chromatography with positive-ion APCI tandem mass spectrometry analysis; however, GRM 05.02 also used on-line SPE purification (pp. 13-14 of MRID 46695143).

2 Only quantitative HPLC results were provided in the ECM.

3 Only Method GRM 05.02 was performed by the ILV. Q = Quantitative HPLC analysis; C = Confirmatory HPLC analysis.

4 For ECM Methods GRM 05.01 and GRM 05.02, soil/sediment matrices were minimally characterized (p. 10 of MRID 46695143). USDA classifications were reported, but particle percentages were not reported. Organic carbon percentages were 0.6%, 1.3%, 5.0%, 1.2%, 2.4% and 2.8% for loam soil, loamy sand soil, clay loam soil, silty clay loam soil, loamy sand sediment and sandy clay loam sediment, respectively.

5 The matrices of the ILV were "a loamy soil and a clay sediment" (p. 14 of MRID 46695144). Characterization for two unspecified soil matrices were provided (texture classification was not specified as USDA; soil origin Mogi Mirim Experimental Station; Appendix B, pp. 121-123). Reported soil characterization data were 76.24% sand, 2.72% silt, 21.04% clay; 11.54 g/dm^3 organic matter for Sample 051329 Soil and 77.68% sand, 2.00% silt, 20.32% clay; 15.06 g/dm^3 organic matter for Sample 051330 Soil. It could not be determined if this data applied to the matrices of the validation.

IV. Method Deficiencies and Reviewer's Comments

1. In the ILV, no performance data was provided for the method GRM 05.01. Method GRM 05.01 was not attempted or validated by the ILV (p. 1 of MRID 46695144).

   The laboratory that conducted the validation (Mogi Mirim Regulatory Laboratory, Dow AgroSciences Ind. Ltda.) belonged to the same organization as the originating laboratory (Dow AgroSciences LLC); however, the study director of the ILV was not familiar with the method both in its development and subsequent use in field studies (pp. 1, 5 of MRID 46695143; pp. 1, 7, 14 of MRID 46695144). The equipment and supplies used in the ILV originated at the ILV. No personnel from the ECM laboratory visited the ILV laboratory during the ILV trial.

2. The soil matrices were insufficiently characterized in the ECM and ILV. In the ECM, USDA classifications were reported, but particle percentages were not reported (only % organic carbon was reported; sources not reported; p. 10 of MRID 46695143). The ILV validated the method with "a loamy soil and a clay sediment", but the texture classification was not specified as USDA (p. 14 of MRID 46695144). Characterization for two unspecified soil matrices was provided (Appendix B, pp. 121-123). The reported soil characterization data were 76.24% sand, 2.72% silt, 21.04% clay; 11.54 g/dm^3 organic matter for Sample 051329 Soil and 77.68% sand, 2.00% silt, 20.32% clay; 15.06 g/dm^3 organic matter for Sample 051330 Soil. Both of these correspond to <u>sandy clay loam</u> in the USDA soil texture triangle. Due to the lack of characterization data in the ECM and the disjointed characterization data in the ILV, it could not be determined if the ILV was provided with the most difficult soil type with which to validate the method.

3. In the ECM, the number of samples was insufficient for all analyses at the LOQ, 10×LOQ, 100×LOQ and 200×LOQ in both methods (n = 2; Appendix A, Tables 2-5, pp. 45-52; Appendix A, Figure 17, p. 66; Appendix B, Tables 2-5, pp. 92-99 of MRID 46695143). OSCPP guidelines recommend a minimum of five samples spiked at each fortification level.

   Three of the individual recoveries in the ECM were <70%: two recoveries of XDE-175-J in silty clay loam soil (GRM 05.01, 68%; GRM 05.02, 66%) and one recovery of XDE-175-L in silty clay loam soil (GRM 05.02, 63%; Appendix A, Tables 2-5, pp. 45-52; Appendix B, Tables 2-5, pp. 92-99 of MRID 46695143). OSCPP guidelines recommend that mean recoveries are within the range 70-120% and RSDs are ≤20%.

4. In both methods GRM 05.01 and GRM 05.02 of the ECM, representative chromatograms were only provided for the quantitative HPLC analysis for loamy sand sediment (653) and for the confirmatory HPLC analysis for loam soil (640; Appendix A, Figures 13-20, pp. 62-69; Appendix B, Figures 21-28, pp. 113-120 of MRID 46695143). OCSPP guidelines recommend that chromatograms are provided for all matrices and HPLC methods which were included in the validation. Also, a reagent blank was not included.

Based on data in the ECM recovery tables, matrix interferences were not found for all analytes/matrices (quantitative HPLC analysis results; Appendix A, Tables 2-5, pp. 45-52; Appendix B, Tables 2-5, pp. 92-99 of MRID 46695143). In the confirmatory HPLC analysis, the matrix was also free of interferences, except for the XDE-175-J analysis in loam soil. A peak at the retention time of the analyte was *ca*. 30-40% of the LOQ based on peak area estimation (peak areas were not reported; Appendix A, Figure 17, p. 66).

5. Recoveries were corrected in the ILV; the recovery data was corrected for residues found in the control samples (pp. 22-24; Tables 3-10, pp. 32-39). In the ECM, Both Methods GRM 05.01 and GRM 05.02 allowed for recovery data to be corrected for residues found in the control samples; however, residues were not quantified in any of the control samples (Appendix A, pp. 37-39; Appendix A, Tables 2-5, pp. 45-52; Appendix B, pp. 82-85; Appendix B, Tables 2-5, pp. 92-99).

6. The toxicological level of concern was not reported for the analytes in soil.

7. The following typographical error was noted in the ILV: the calibration curves were reportedly shown in “Appendix D”, when they were shown in Appendix C (p. 25 of MRID 46695144).

8. In the ILV, the communications between the ILV and the sponsor were documented (p. 26; Appendix D, pp. 141-145 of MRID 46695144).

9. In the ECM, it was determined that the spiking solutions for GRM 05.01 and GRM 05.02 and the calibration standards used in GRM 05.02 were stable for at least 276 days under refrigeration storage (p. 17-19 of MRID 46695143). The calibration standards used in GRM 05.01 were not stable for at least 276 days under refrigeration storage. Also, the extraction efficiency data from an aerobic degradation study was reported (Dow AgroSciences LLC Study ID 040068; 2005; pp. 15-16, 24).

10. It was reported for the ILV that the analytical procedure for one set of 23 samples required approximately three person hours for preparation (p. 26 of MRID 46695144). The on-line SPE and HPLC were conducted overnight unattended.

11. The reviewer noted that spinetoram is a mixture of XDE-175-J and XDE-175-L [3:1 ratio (J:L); p. 3; http://epa.gov/pesticides/chem_search/cleared_reviews/csr_PC-110009_12-Oct-10_a.pdf]. Additionally, the reviewer found that PC code 110008 is also associated with spinetoram (pp. 1, 38).

V. References

Keith, L. H.; Crummett, W.; Deegan, J., Jr.; Libby, R. A.; Taylor, J. K.; Wentler, G. *Anal. Chem.* 1983, *55*, 2210-2218 (Appendix A, p. 49; Appendix B, p. 107 of MRID 46695143).

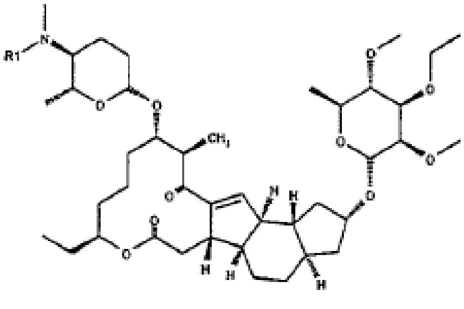
U.S. Environmental Protection Agency. 2012. Ecological Effects Test Guidelines, OCSPP 850.6100, Environmental Chemistry Methods and Associated Independent Laboratory Validation. Office of Chemical Safety and Pollution Prevention, Washington, DC. EPA 712-C-001.

40 CFR Part 136. Appendix B. Definition and Procedure for the Determination of the Method Detection Limit-Revision 1.11, pp. 317-319.

Attachment 1: Chemical Names and Structures

Spinetoram (XDE-175-J)

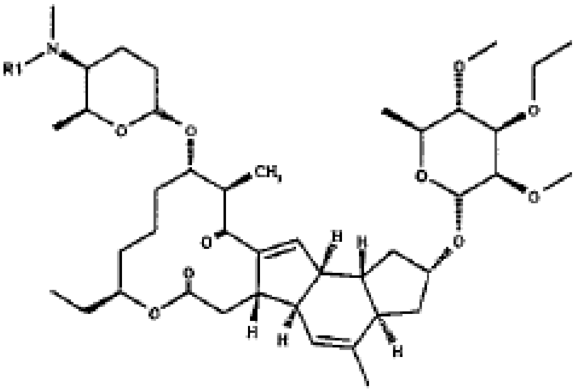
IUPAC Name:	(2R,3aR,5aR,5bS,9S,13S,14R,16aS,16bR)-13-{[(2R,5S,6R)-5-(dimethylamino)-6-methyltetrahydro-2H-pyran-2-yl]oxy}-9-ethyl-14-methyl-7,15-dioxo-2,3,3a,4,5,5a,5b,6,7,9,10,11,12,13,14,15,16a,16b-octadecahydro-1-H-as-indaceno[3,2-d]oxacyclododecin-2-yl 6-deoxy-3-O-ethyl-2,4-di-O-methyl-alpha-L-mannopyranoside
CAS Name:	1-H-as-indaceno[3,2-d]oxacyclododecin-7,15-dione, 2-[(6-deoxy-3-O-ethyl-2,4-di-O-methyl-a-L-mannopyranosyl)oxy]-13-[[(2R,5S,6R)-5-(dimethylamino)tetrahydro-6-methyl 2H-pyran-2-yl]oxy]-9-ethyl-2,3,3a,4,5,5a,5b,6,7,9,10,11,12,13,14,15,16a,16b-hexadecahydro 14-methyl-(2R,3aR,5aR,5bS,9S,13S,14R,16aS,16bR)
CAS Number:	187166-40-1
SMILES String:	



XDE-175-J, R1 = CH_3
XDE-175-*N*-Demethyl-J, R1 = H

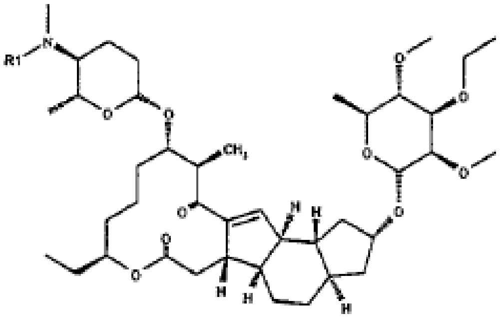
Spinetoram (XDE-175-L)

IUPAC Name:	(2S,3aR,5aS,5bS,9S,13S,14R,16aS,16bS)-13-{[(2R,5S,6R)-5-(dimethylamino)-6-methyltetrahydro-2H-pyran-2-yl]oxy}-9-ethyl-4,14-dimethyl-7,15-dioxo-2,3,3a,4,5,5a,5b,6,7,9,10,11,12,13,14,15,16a,16b-hexadecahydro-1-H-as-indaceno[3,2-d]oxacyclododecin-2-yl 6-deoxy-3-O-ethyl-2,4-di-O-methyl-alpha-L-mannopyranoside
CAS Name:	1-H-as-indaceno[3,2-d]oxacyclododecin-7,15-dione, 2-[(6-deoxy-3-O-ethyl-2,4-di-O-methyl-a-L-mannopyranosyl)oxy]-13-[[(2R,5S,6R)-5-(dimethylamino)tetrahydro-6-methyl-2H-pyran-2-yl]oxy]-9-ethyl-2,3,3a,4,5,5a,5b,6,7,9,10,11,12,13,14,15,16a,16b-tetradecahydro 4,14-dimethyl-(2S,3aR,5aS,5bS,9S,13S,14R,16aS,16bS)
CAS Number:	187166-15-0
SMILES String:	



XDE-175-L, R1 = CH_3
XDE-175-*N*-Demethyl-L, R1 = H

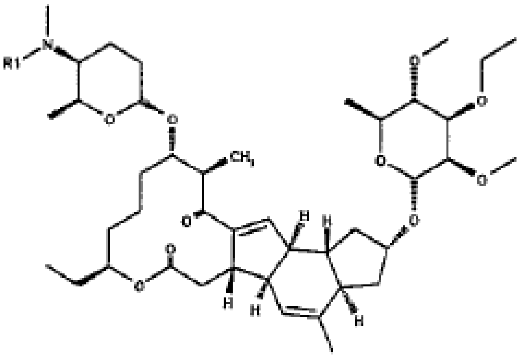
XDE-175-N-Demethyl-J
IUPAC Name: (2R,3aR,5aR,5bS,9S,13S,14R,16aS,16bR)- 9-ethyl-14-methyl-13-{[(2S,5S,6R)- 6-methyl-5-(methylamino)tetrahydro-2H-pyran-2-yl]oxy}-7,15-dioxo-2,3,3a,4,5,5a,5b,6,7,9,10,11,12,13,14,15,16a,16b-octadecahydro-1-H-as-indaceno[3,2-d]oxacyclododecin-2-yl 6-deoxy-3-O-ethyl-2,4-di-O-methyl-beta-L-mannopyranoside
CAS Name:
CAS Number:
SMILES String:



XDE-175-J, R1 = CH_3
XDE-175-*N*-Demethyl-J, R1 = H

XDE-175-N-Demethyl-L
IUPAC Name: (2S,3aR,5aS,5bS,9S,13S,14R,16aS,16bS)- 9-ethyl-4,14-dimethyl-13-{[(2S,5S,6R)- 6-methyl-5-(methylamino)tetrahydro-2H-pyran-2-yl]oxy}-7,15-dioxo-2,3,3a,4,5,5a,5b,6,7,9,10,11,12,13,14,15,16a,16b-hexadecahydro-1-H-as-indaceno[3,2-d]oxacyclododecin-2-yl 6-deoxy-3-O-ethyl-2,4-di-O-methyl-beta-L-mannopyranoside

CAS Name:
CAS Number:
SMILES String:



XDE-175-L, R1 = CH_3
XDE-175-*N*-Demethyl-L, R1 = H