

Analytical methods for pyraclostrobin (BAS 500 F) and its metabolites, BF 500-5 (Reg. No. 298327), BF 500-12 (500M59; Reg. No. 412053), BF 500-11 (500M60; Reg. No. 411847), BF 500-13 (500M62; Reg. No. 412785), BF 500-14 (500M76; Reg. No. 413038), and BF 500-15 (500M78; Reg. No. 377613), in water

Reports: ECM: EPA MRID No.: 50088704. Tilting, N. 2012. Validation of Analytical Method L0182/01: Determination of BAS 500 F (Pyraclostrobin) and Its Metabolites Reg.No 412053 (500M59), Reg No. 411847 (500M60), Reg No. 412785 (500M62), Reg No. 413038 (500M76), and Reg No. 377613 (500M78) in ground- surface- and tapwater using LC-MS/MS. BASF Study Code: 370958. BASF Registration Document No.: 2012/1009641. Report prepared by BASF SE, Crop Protection, Limburgerhof, Germany; and sponsored and submitted by BASF Corporation, Research Triangle Park, North Carolina; 117 pages. Final report issued February 10, 2012.

ILV: EPA MRID No. 50088705. Bianca, C.M. 2015. Independent Laboratory Validation of BASF Method L0182/02: "Determination of BAS 500 F (Pyraclostrobin) and Its Metabolites BF 500-5 (Reg. No. 298327), BF 500-12 (Reg. No. 412053), BF 500-11 (Reg. No. 411847), BF 500-13 (Reg. No. 412785), BF 500-14 (Reg. No. 413038) and BF 500-15 (Reg. No. 377613) in ground- and surface- water by LC/MS/MS. BASF Study No.: 714839. BASF Registration Document No.: 2015/7001873. Report prepared by JRF America, Audubon, Pennsylvania, sponsored and submitted by BASF Crop Protection, Research Triangle Park, North Carolina; 313 pages. Final report issued May 1, 2014; amended report issued August 26, 2015.

Document No.: MRIDs 50088704 & 50088705

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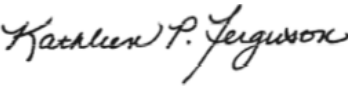
Statements: ECM: The study was conducted in accordance with OECD and German Good Laboratory Practice (GLP) standards, which are fairly compatible with USEPA FIFRA (40 CFR Part 160) and TSCA GLP standards (p. 3 of MRID 50088704). Signed and dated No Data Confidentiality, GLP and Quality Assurance statements were provided (pp. 2-4). An authenticity statement was included with the GLP statement.

ILV: The study was conducted in accordance with USEPA FIFRA GLP standards (p. 3 of MRID 50088705). 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**. Two sets of performance data were not included for the determination of residues of pyraclostrobin metabolite BF 500-5. The specificity of the method for BF 500-14, BF 500-15 and BF 500-5 was not supported by the ILV representative chromatograms. In the ECM, no 10×LOQ chromatograms were provided and the specificity of the method was not fully supported for BF 500-15 by the ECM representative chromatograms.

PC Code: 099100

EFED Final Reviewer: Lewis Ross Brown, III Signature: Lewis Ross Brown, III
Environmental Biologist Date: April 25, 2018

CDM/CSS- Dynamac JV Reviewers:	Lisa Muto,	Signature:	
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This Data Evaluation Record may have been altered by the Environmental Fate and Effects Division subsequent to signing by CDM/CSS-Dynamac JV personnel.

Executive Summary

The analytical methods, BASF Method L0182/01 and BASF Method L0182/02, are designed for the quantitative determination of pyraclostrobin (BAS 500 F) in water at the LOQ of 0.003 µg/L using LC/MS/MS and of pyraclostrobin metabolites BF 500-12 (500M59; Reg. No. 412053), BF 500-11 (500M60; Reg. No. 411847), BF 500-13 (500M62; Reg. No. 412785), BF 500-14 (500M76; Reg. No. 413038), and BF 500-15 (500M78; Reg. No. 377613) in water at the LOQ of 0.03 µg/L using LC/MS/MS. The LOQ is less than the lowest toxicological level of concern of pyraclostrobin (estuarine/marine invertebrate chronic toxicity NOEAC of 0.5 µg/L; DP 417743), and BF 500-12, BF 500-11, BF 500-13, BF 500-14, and BF 500-15 (which are not of concern for aquatic organisms; DP 417743). The analytical method, BASF Method L0182/02, is also designed for the quantitative determination of pyraclostrobin metabolite BF 500-5 (Reg. No. 298327) in water at the LOQ of 0.03 µg/L using LC/MS/MS. The LOQ is less than the lowest toxicological level of concern of BF 500-5 (which is not of concern for aquatic organisms; DP 417743). The ILV was based on BASF Method L0182/02, not BASF Method L0182/01; however, BASF Method L0182/01 and BASF Method L0182/02 appeared to be identical, except for the BF 500-5 analyte. Therefore, the ILV was considered to be an independent validation of BASF Method L0182/01, as well as BASF Method L0182/02. However, two sets of performance data were not included for the portion of BASF Method L0182/02 which included BF 500-5 as an analyte; only the ILV data was submitted. The ECM performed BASF Method L0182/01 using uncharacterized drinking, surface and ground water; no 10×LOQ chromatograms were provided in the ECM. The ILV performed BASF Method L0182/02 using characterized surface and drinking (well) water. The ILV validated the method for pyraclostrobin and its metabolites BF 500-5, BF 500-12, BF 500-11, BF 500-13, BF 500-14, and BF 500-15 in the first trial for surface water and in the second trial for drinking water with the addition of a pyraclostrobin metabolite BF 500-5 and insignificant modifications to the analytical instrumentation. All ILV and ECM data regarding repeatability, accuracy, and precision were satisfactory for all analytes in all matrices. ILV linearity was not satisfactory for BF 500-5, BF 500-12, BF 500-14, and BF 500-15. Significant baseline noise affected the analyte peak integration of BF 500-14 and/or BF 500-15 in the surface water chromatograms of the ILV and ECM. The BF 500-5 analyte peak was small compared to baseline noise in both ILV matrices; the BF 500-14 analyte peak was small compared to baseline noise in the drinking water matrix. In the ECM representative chromatograms, major baseline noise interfered with peak integration of BF 500-15 in the surface water matrix.

Table 1. Analytical Method Summary

Analyte(s) by Pesticide ¹	MRID		EPA Review	Matrix	Method Date (dd/mm/yyyy)	Registrant	Analysis	Limit of Quantitation (LOQ)
	Environmental Chemistry Method	Independent Laboratory Validation						
Pyraclostrobin (BAS 500 F) BF 500-12 (500M59; Reg. No. 412053) BF 500-11 (500M60; Reg. No. 411847) BF 500-13 (500M62; Reg. No. 412785) BF 500-14 (500M76; Reg. No. 413038) BF 500-15 (500M78; Reg. No. 377613)	50088704 ²	50088705		Water ^{4,5}	10/02/2012	BASF Corporation	LC/MS/MS	0.03 µg/L
BF 500-5 (Reg. No. 298327)	None submitted ³				03/02/2014 ⁶			

1 Pyraclostrobin (BAS 500 F) = Methyl 2-[1-(4-chlorophenyl)pyrazol-3-yloxymethyl]-N-methoxycarbanilate; BF 500-5 (Reg. No. 298327) = 1-(4-Chlorophenyl)-1H-pyrazol-3-ol; BF 500-12 (500M59; Reg. No. 412053) = Methyl N-(2-{[1-(4-hydroxyphenyl)-1H-pyrazol-3-yl]oxymethyl}phenyl) N-methoxy carbamate; BF 500-11 (500M60; Reg. No. 411847) = Methyl N-methoxy N-{2-[(1H-pyrazol-3-yl)oxymethyl]phenyl}carbamate; BF 500-13 (500M62; Reg. No. 412785) = Methyl N-[2-(1H-pyrazol-3-yloxymethyl)phenyl]carbamate; BF 500-14 (500M76; Reg. No. 413038) = Methyl N-(2-[2-(4-chlorophenyl)-5-oxo-2,5-dihydro-pyrazol-1-ylmethyl]-phenyl) N-methoxy carbamate; and BF 500-15 (500M78; Reg. No. 377613) = 1-(4-Hydroxyphenyl)-1H-pyrazol-3-ol.

2 BASF Method L0182/01.

3 BF 500-5 was not included as an analyte in ECM MRID 50088704 which was a validation of BASF Method L0182/01. BF 500-5 was included as an analyte in the ILV MRID 50099705 which was an ILV of BASF Method L0182/02. BASF Method L0182/01 and BASF Method L0182/02 appeared to be identical, but BF 500-5 was not included as an analyte in BASF Method L0182/01.

4 In the ECM, ground water was collected from the Schifferstadt Water Supply, surface water was collected from Kelmetschweiher, and tap water was collected from Limburgerhof (p. 14 of MRID 50088704). The water matrices were not characterized; the water sources were not further specified.

5 In the ILV, surface (lake) water (pH 6.9; 33 mg equiv. CaCO₃/L; 7.9 ppm total organic carbon) was collected from Jordan Lake, Cary, North Carolina, and drinking (well) water (pH 6.8; <1 mg equiv. CaCO₃/L; 0.9 ppm total organic carbon) was collected from BASF Research Triangle Park, North Carolina (p. 18; Appendix B, pp. 235-237 of MRID 50088705). Water samples were characterized by Agvise Laboratories, Northwood, North Dakota.

6 BASF Method L0182/02 report date from p. 38 of ILV MRID 50088705.

I. Principle of the Method

Water samples (50 mL) were fortified with mixed fortification solutions in 150 mL glass flasks and acidified with 500 µL of formic acid (Appendix III, pp. 107-112, 115 of MRID 50088704). A C₁₈-

solid phase extraction (SPE) cartridge (6 mL/500 mg) was pre-conditioned with 5 mL each of ethyl acetate and 0.1% formic acid in water. The cartridge was not allowed to dry. The water sample was applied to the cartridge with vacuum. The cartridge was washed with 5 mL each of 0.1% formic acid in water. Full vacuum was applied to the column for 30 seconds. The analytes were eluted with 2 x 5 mL of ethyl acetate. The solvent of the eluate was completely evaporated via N-Elvap at 40°C (or similar conditions). The residue was reconstituted in acetonitrile:Millipore water (2:8, v:v) prior to analysis by LC/MS/MS (higher dilutions were used for higher fortifications, as necessary).

Water samples were analyzed for pyraclostrobin (BAS 500 F) and its metabolites, BF 500-12 (500M59; Reg. No. 412053), BF 500-11 (500M60; Reg. No. 411847), BF 500-13 (500M62; Reg. No. 412785), BF 500-14 (500M76; Reg. No. 413038), and BF 500-15 (500M78; Reg. No. 377613), using Agilent HP 1100 LC (Atlantis T3 column, 3 mm x 150 mm, 3 µm column; column temperature 40°C) using a gradient mobile phase of (A) water:formic acid (1000:1, v:v) and (B) acetonitrile:formic acid (1000:1, v:v) [time ratio A:B; 0.0-1 min. 85:15, 7-13 min. 10:90, 13.10-17 min. 85:15] coupled with a PE Sciex API 4000 mass spectrometer using positive electrospray ionization in Multiple Reaction Monitoring (MRM) mode (Appendix III, p. 113 of MRID 50088704). Injection volume was 50 µL. Two ion transitions were monitored for each analyte (quantitation and confirmatory, respectively) as follows: m/z 388→194 and m/z 388→163 for pyraclostrobin (BAS 500 F); m/z 370→194 and m/z 370→278 for BF 500-12 (500M59); m/z 278→194 and m/z 278→149 for BF 500-11 (500M60); m/z 248→132 and m/z 248→216 for BF 500-13 (500M62); m/z 388→241 and m/z 388→300 for BF 500-14 (500M76); and m/z 177→135 and m/z 177→132 for BF 500-15 (500M78), respectively. Expected retention times were *ca.* 9.21, 7.61, 6.63, 6.58, 7.06 and 5.12 minutes for pyraclostrobin (BAS 500 F), BF 500-12 (500M59), BF 500-11 (500M60), BF 500-13 (500M62), BF 500-14 (500M76), and BF 500-15 (500M78), respectively.

ILV MRID 50099705 was an ILV of BASF Method L0182/02, not BASF Method L0182/01; however, BASF Method L0182/01 appeared to be the same as BASF Method L0182/02, except for the addition of BF 500-5 as an analyte (pp. 7, 18, 38 of MRID 50088705). In the ILV, the ECM was performed as written, except for the addition of a pyraclostrobin metabolite BF 500-5 (Reg. No. 298327) and use of a different LC/MS/MS system (pp. 21-27; Table 10.40, pp. 80-81). Waters Acquity UPLC coupled to an AB Sciex 4000 Qtrap® with Turbo Ion Spray was used. All LC/MS conditions were the same. Two ion transitions were monitored for each analyte (quantitation and confirmatory, respectively) as follows: m/z 389→195 and m/z 389→164 for pyraclostrobin (BAS 500 F); m/z 196→154 and m/z 196→118 for BF 500-5 (Reg. No. 298327); m/z 370→194 and m/z 370→278 for BF 500-12 (500M59; Reg. No. 412053); m/z 278→194 and m/z 278→149 for BF 500-11 (500M60; Reg. No. 411847); m/z 248→132 and m/z 248→164 for BF 500-13 (500M62; Reg. No. 412785); m/z 389→242 and m/z 389→301 for BF 500-14 (500M76; Reg. No. 413038); and m/z 177→135 and m/z 177→132 for BF 500-15 (500M78; Reg. No. 377613), respectively. Expected retention times were *ca.* 6.62, 6.62, 7.25, 6.13, 6.10, 6.60 and 4.55 minutes for pyraclostrobin (BAS 500 F), BF 500-12 (500M59), BF 500-11 (500M60), BF 500-13 (500M62), BF 500-14 (500M76), and BF 500-15 (500M78), respectively. No other modifications to the ECM were reported.

The Limit of Quantification (LOQ) and Limit of Detection (LOD) for pyraclostrobin (BAS 500 F) in water were 0.003 µg/L and 0.0006 µg/L, respectively, in the ECM and ILV (p. 5; Appendix III, p. 102 of MRID 50088704; p. 7 of MRID 50088705). The LOQ and LOD for pyraclostrobin metabolites BF 500-12 (500M59; Reg. No. 412053), BF 500-11 (500M60; Reg. No. 411847), BF

500-13 (500M62; Reg. No. 412785), BF 500-14 (500M76; Reg. No. 413038), and BF 500-15 (500M78; Reg. No. 377613) in water were 0.03 µg/L and 0.006 µg/L, respectively, in the ECM and ILV. The LOQ and LOD for pyraclostrobin metabolite BF 500-5 (Reg. No. 298327) in water were 0.03 µg/L and 0.006 µg/L, respectively, in the ILV; BF 500-5 was not included as analyte in the ECM.

II. Recovery Findings

ECM (MRID 50088704): Mean recoveries and relative standard deviations (RSDs) were within guideline requirements (mean 70-120%; RSD ≤20%) for analysis of pyraclostrobin in three water matrices at fortification levels of 0.003 µg/L (LOQ) and 0.03 µg/L (10×LOQ) and for analysis of pyraclostrobin metabolites BF 500-12 (500M59; Reg. No. 412053), BF 500-11 (500M60; Reg. No. 411847), BF 500-13 (500M62; Reg. No. 412785), BF 500-14 (500M76; Reg. No. 413038), and BF 500-15 (500M78; Reg. No. 377613) in three water matrices at fortification levels of 0.03 µg/L (LOQ) and 0.3 µg/L (10×LOQ; Tables 1-6, pp. 22-24). Pyraclostrobin and its metabolites were identified using two ion transitions; performance data (recovery results) from primary and confirmatory analyses were comparable. The ground water was collected from the Schifferstadt Water Supply, surface water was collected from Kelmetschweiher, and tap water was collected from Limburgerhof (p. 14). The water matrices were not characterized; the water sources were not further specified.

ILV (MRID 50088705): Mean recoveries and RSDs were within guideline requirements for analysis of pyraclostrobin in two water matrices at fortification levels of 0.003 µg/L (LOQ) and 0.03 µg/L (10×LOQ) and for analysis of pyraclostrobin metabolites BF 500-5 (Reg. No. 298327), BF 500-12 (500M59; Reg. No. 412053), BF 500-11 (500M60; Reg. No. 411847), BF 500-13 (500M62; Reg. No. 412785), BF 500-14 (500M76; Reg. No. 413038), and BF 500-15 (500M78; Reg. No. 377613) in two water matrices at fortification levels of 0.03 µg/L (LOQ) and 0.3 µg/L (10×LOQ; Tables 1-2, pp. 31-34). BF 500-15 not included in drinking water validation, only the surface water validation. Pyraclostrobin and its metabolites were identified using two ion transitions; performance data (recovery results) from primary and confirmatory analyses were comparable, except for the drinking water validations of BF 500-5 and BF 500-14 at the LOQ which were fairly comparable. The surface (lake) water (pH 6.9; 33 mg equiv. CaCO₃/L; 7.9 ppm total organic carbon) was collected from Jordan Lake, Cary, North Carolina, and drinking (well) water (pH 6.8; <1 mg equiv. CaCO₃/L; 0.9 ppm total organic carbon) was collected from BASF Research Triangle Park, North Carolina (p. 18; Appendix B, pp. 235-237). Water samples were characterized by Agvise Laboratories, Northwood, North Dakota. The method for pyraclostrobin and its metabolites BF 500-5, BF 500-12, BF 500-11, BF 500-13, BF 500-14, and BF 500-15 was validated in the first trial in surface water with the addition of a pyraclostrobin metabolite BF 500-5 (Reg. No. 298327) and insignificant modifications to the analytical instrumentation (pp. 11, 30). The method for pyraclostrobin and its metabolites BF 500-5, BF 500-12, BF 500-11, BF 500-13, and BF 500-14 was validated in the second trial in drinking water with the addition of a pyraclostrobin metabolite BF 500-5 (Reg. No. 298327) and insignificant modifications to the analytical instrumentation. BF 500-5 was not included as analyte in the ECM; therefore, two sets of validation data were not included for BF 500-5.

Table 2. Initial Validation Method Recoveries for Pyraclostrobin (BAS 500 F) and Its Metabolites BF 500-12 (500M59), BF 500-11 (500M60), BF 500-13 (500M62), BF 500-14 (500M76), and BF 500-15 (500M78) in Water

Analyte ¹	Fortification Level (µg/L)	Number of Tests	Recovery Range (%)	Mean Recovery (%)	Standard Deviation (%)	Relative Standard Deviation (%)
Ground Water^{2,3}						
Quantitation ion						
Pyraclostrobin (BAS 500 F)	0.003 (LOQ)	5	80.8-100.0	87.9	6.5	7.4
	0.03	5	90.8-97.6	94.2	2.4	2.6
BF 500-12 (500M59)	0.03 (LOQ)	5	94.4-98.8	95.9	1.7	1.7
	0.3	5	95.2-102.8	97.8	2.7	2.8
BF 500-11 (500M60)	0.03 (LOQ)	5	92.8-102.4	95.7	3.6	3.8
	0.3	5	96.4-103.6	98.8	2.6	2.7
BF 500-13 (500M62)	0.03 (LOQ)	5	95.6-99.2	97.0	1.4	1.4
	0.3	5	90.4-101.2	95.4	3.7	3.9
BF 500-14 (500M76)	0.03 (LOQ)	5	90.0-104.4	97.4	5.5	5.7
	0.3	5	99.2-106.8	102.8	3.2	3.1
BF 500-15 (500M78)	0.03 (LOQ)	5	90.8-93.6	92.2	1.0	1.0
	0.3	5	99.2-109.2	103.1	3.5	3.4
Confirmatory ion						
Pyraclostrobin (BAS 500 F)	0.003 (LOQ)	5	85.2-103.2	92.6	6.8	7.4
	0.03	5	92.8-105.6	99.0	5.3	5.3
BF 500-12 (500M59)	0.03 (LOQ)	5	96.4-102.4	98.9	2.6	2.6
	0.3	5	91.2-102.0	95.8	3.7	3.9
BF 500-11 (500M60)	0.03 (LOQ)	5	92.8-99.6	95.2	2.3	2.4
	0.3	5	89.6-99.2	95.6	3.3	3.4
BF 500-13 (500M62)	0.03 (LOQ)	5	96.8-105.6	99.8	3.1	3.1
	0.3	5	93.2-104.8	97.7	4.0	4.0
BF 500-14 (500M76)	0.03 (LOQ)	5	98.4-107.2	103.4	3.4	3.3
	0.3	5	94.0-108.8	99.3	5.2	5.3
BF 500-15 (500M78)	0.03 (LOQ)	5	89.2-98.0	93.5	2.8	3.0
	0.3	5	94.8-104.8	99.8	4.2	4.2
Surface Water^{2,3}						
Quantitation ion						
Pyraclostrobin (BAS 500 F)	0.003 (LOQ)	5	82.8-94.0	89.3	3.7	4.1
	0.03	5	93.2-105.2	97.5	4.5	4.6
BF 500-12 (500M59)	0.03 (LOQ)	5	94.4-99.2	97.1	1.8	1.8
	0.3	5	94.4-98.8	96.9	1.7	1.7
BF 500-11 (500M60)	0.03 (LOQ)	5	85.2-89.6	86.7	1.5	1.8
	0.3	5	81.2-86.8	85.0	2.1	2.4
BF 500-13 (500M62)	0.03 (LOQ)	5	89.6-94.4	91.2	1.7	1.9
	0.3	5	82.4-86.0	83.8	1.3	1.6
BF 500-14 (500M76)	0.03 (LOQ)	5	92.8-102.8	97.6	3.6	3.7
	0.3	5	86.4-104.4	97.7	6.0	6.2
BF 500-15 (500M78)	0.03 (LOQ)	5	91.6-101.2	95.0	3.5	3.7
	0.3	5	91.6-105.6	97.2	5.0	5.2
Confirmatory ion						
Pyraclostrobin (BAS 500 F)	0.003 (LOQ)	5	86.4-102.0	93.8	5.7	6.1
	0.03	5	96.0-103.6	100.4	2.6	2.6
	0.03 (LOQ)	5	94.8-102.4	98.6	2.9	3.0

Analyte ¹	Fortification Level (µg/L)	Number of Tests	Recovery Range (%)	Mean Recovery (%)	Standard Deviation (%)	Relative Standard Deviation (%)
BF 500-12 (500M59)	0.3	5	94.8-100.8	97.8	2.3	2.3
BF 500-11 (500M60)	0.03 (LOQ)	5	84.8-93.2	87.6	3.0	3.4
	0.3	5	85.2-89.6	88.4	1.7	1.9
BF 500-13 (500M62)	0.03 (LOQ)	5	86.8-99.2	91.1	4.3	4.7
	0.3	5	80.0-85.2	82.0	1.9	2.4
BF 500-14 (500M76)	0.03 (LOQ)	5	94.4-108.0	104.0	4.9	4.7
	0.3	5	96.0-106.8	102.1	3.5	3.4
BF 500-15 (500M78)	0.03 (LOQ)	5	96.4-102.0	100.3	2.0	2.0
	0.3	5	95.2-110.4	103.3	5.0	4.8
Tap Water^{2,3}						
Quantitation ion						
Pyraclostrobin (BAS 500 F)	0.003 (LOQ)	5	82.0-95.2	89.6	4.4	4.9
	0.03	5	92.8-108.4	103.0	5.3	5.2
BF 500-12 (500M59)	0.03 (LOQ)	5	96.8-100.4	98.9	1.3	1.3
	0.3	5	98.0-105.6	101.3	2.9	2.9
BF 500-11 (500M60)	0.03 (LOQ)	5	94.4-99.2	97.3	2.0	2.1
	0.3	5	90.0-99.6	96.4	3.3	3.4
BF 500-13 (500M62)	0.03 (LOQ)	5	96.4-101.2	99.3	1.6	1.6
	0.3	5	86.0-100.8	93.5	5.7	6.1
BF 500-14 (500M76)	0.03 (LOQ)	5	81.6-108.0	100.6	9.8	9.7
	0.3	5	94.0-108.8	101.0	4.7	4.7
BF 500-15 (500M78)	0.03 (LOQ)	5	93.6-98.8	96.7	2.2	2.3
	0.3	5	96.0-101.2	99.2	1.7	1.7
Confirmatory ion						
Pyraclostrobin (BAS 500 F)	0.003 (LOQ)	5	88.8-95.6	92.1	2.9	3.1
	0.03	5	90.4-119.2	104.4	9.8	9.4
BF 500-12 (500M59)	0.03 (LOQ)	5	95.2-103.2	98.8	2.6	2.6
	0.3	5	98.0-107.2	101.8	3.3	3.2
BF 500-11 (500M60)	0.03 (LOQ)	5	92.8-100.8	95.7	2.8	2.9
	0.3	5	93.6-101.6	95.9	3.0	3.1
BF 500-13 (500M62)	0.03 (LOQ)	5	92.0-107.6	98.0	5.4	5.5
	0.3	5	84.8-106.8	94.8	7.2	7.6
BF 500-14 (500M76)	0.03 (LOQ)	5	92.8-109.6	100.6	6.2	6.1
	0.3	5	96.4-114.0	105.0	5.9	5.6
BF 500-15 (500M78)	0.03 (LOQ)	5	90.8-98.4	95.8	2.7	2.8
	0.3	5	93.6-102.0	99.4	3.1	3.1

Data (uncorrected recovery results, Tables 1-4, pp. 15-18) were obtained from Tables 1-6, pp. 22-24 of MRID 50088704. The metabolite, MF 500-5, was not included in the ECM.

1 Pyraclostrobin (BAS 500 F) = Methyl 2-[1-(4-chlorophenyl)pyrazol-3-yloxymethyl]-N-methoxycarbamate; BF 500-12 (500M59; Reg. No. 412053) = Methyl N-(2-([1-(4-hydroxyphenyl)-1H-pyrazol-3-yl]oxymethyl)phenyl) N-methoxy carbamate; BF 500-11 (500M60; Reg. No. 411847) = Methyl N-methoxy N-{2-[(1H-pyrazol-3-yl)oxymethyl]phenyl} carbamate; BF 500-13 (500M62; Reg. No. 412785) = Methyl N-[2-(1H-pyrazol-3-yloxymethyl)phenyl] carbamate; BF 500-14 (500M76; Reg. No. 413038) = Methyl N-(2-[2-(4-chlorophenyl)-5-oxo-2,5-dihydro-pyrazol-1-ylmethyl]-phenyl) N-methoxy carbamate; and BF 500-15 (500M78; Reg. No. 377613) = 1-(4-Hydroxyphenyl)-1H-pyrazol-3-ol.

2 The ground water was collected from the Schifferstadt Water Supply, surface water was collected from Kelmetschweiher, and tap water was collected from Limburgerhof (p. 14). The water matrices were not characterized; the water sources were not further specified.

3 Two ion transitions were monitored for each analyte (quantitation and confirmatory, respectively) as follows: *m/z*

388→194 and m/z 388→163 for pyraclostrobin (BAS 500 F); m/z 370→194 and m/z 370→278 for BF 500-12 (500M59); m/z 278→194 and m/z 278→149 for BF 500-11 (500M60); m/z 248→132 and m/z 248→216 for BF 500-13 (500M62); m/z 388→241 and m/z 388→300 for BF 500-14 (500M76); and m/z 177→135 and m/z 177→132 for BF 500-15 (500M78), respectively.

Table 3. Independent Validation Method Recoveries for Pyraclostrobin (BAS 500 F) and Its Metabolites BF 500-5, BF 500-12 (500M59), BF 500-11 (500M60), BF 500-13 (500M62), BF 500-14 (500M76), and BF 500-15 (500M78) in Water

Analyte ¹	Fortification Level (µg/L)	Number of Tests	Recovery Range (%)	Mean Recovery (%)	Standard Deviation (%)	Relative Standard Deviation (%)
Surface (Lake) Water^{2,3}						
Quantitation ion						
Pyraclostrobin (BAS 500 F)	0.003 (LOQ)	5	96-113	103	6.82	6.61
	0.03	5	98-107	104	3.41	3.27
BF 500-5 ⁴	0.03 (LOQ)	5	62-90	78.2	10.6	13.6
	0.3	5	77-106	90.8	11.0	12.1
BF 500-12 (500M59)	0.03 (LOQ)	5	83-92	86.7	3.40	3.92
	0.3	5	89-98	93.1	3.12	3.35
BF 500-11 (500M60)	0.03 (LOQ)	5	98-105	101	2.89	2.85
	0.3	5	99-106	103	3.04	2.95
BF 500-13 (500M62)	0.03 (LOQ)	5	93-99	95.6	2.93	3.06
	0.3	5	96-104	99.8	3.17	3.18
BF 500-14 (500M76)	0.03 (LOQ)	5	88-110	99.5	7.68	7.72
	0.3	5	84-116	104	12.36	11.9
BF 500-15 (500M78)	0.03 (LOQ)	5	86-104	91.2	7.66	8.40
	0.3	5	111-120	116	3.34	2.89
Confirmatory ion						
Pyraclostrobin (BAS 500 F)	0.003 (LOQ)	5	101-116	107	5.58	5.19
	0.03	5	101-111	106	3.47	3.27
BF 500-5 ⁴	0.03 (LOQ)	5	74-118	95.2	15.8	16.6
	0.3	5	80-105	95.0	10.4	11.0
BF 500-12 (500M59)	0.03 (LOQ)	5	83-89	85.4	2.60	3.05
	0.3	5	87-90	88.1	0.91	1.04
BF 500-11 (500M60)	0.03 (LOQ)	5	96-107	101	4.57	4.53
	0.3	5	98-103	101	2.22	2.19
BF 500-13 (500M62)	0.03 (LOQ)	5	94-100	97.0	3.00	3.10
	0.3	5	96-103	99.9	2.64	2.65
BF 500-14 (500M76)	0.03 (LOQ)	5	97-116	108	7.65	7.08
	0.3	5	97-112	104	6.40	6.18
BF 500-15 (500M78)	0.03 (LOQ)	5	88-106	94.2	7.09	7.53
	0.3	5	108-121	114	6.45	5.68
Drinking (Well) Water^{2,3}						
Quantitation ion						
Pyraclostrobin (BAS 500 F)	0.003 (LOQ)	5	72-97	88.9	9.61	10.8
	0.03	5	97-110	102	5.51	5.41
BF 500-5 ⁴	0.03 (LOQ)	5	70-113	87.0	17.22	19.8
	0.3	5	92-105	98.2	5.27	5.37
BF 500-12 (500M59)	0.03 (LOQ)	5	74-93	86.6	7.94	9.17
	0.3	5	95-102	98.3	2.82	2.87

BF 500-11 (500M60)	0.03 (LOQ)	5	91-99	94.7	2.83	2.99
	0.3	5	96-107	101	4.22	4.16
BF 500-13 (500M62)	0.03 (LOQ)	5	91-100	95.4	3.63	3.80
	0.3	5	92-105	97.4	5.11	5.25
BF 500-14 (500M76)	0.03 (LOQ)	5	70-113	86.3	16.76	19.4
	0.3	5	85-103	91.0	7.15	7.86
BF 500-15 (500M78)	0.03 (LOQ)	5	Not included as an analyte			
	0.3	5				
Confirmatory ion						
Pyraclostrobin (BAS 500 F)	0.003 (LOQ)	5	74-97	89.8	9.41	10.5
	0.03	5	92-111	105	8.19	7.83
BF 500-5 ⁴	0.03 (LOQ)	5	90-109	100	9.31	9.30
	0.3	5	89-110	101	8.87	8.80
BF 500-12 (500M59)	0.03 (LOQ)	5	76-99	83.8	8.88	10.6
	0.3	5	86-108	94.6	8.38	8.86
BF 500-11 (500M60)	0.03 (LOQ)	5	84-96	90.5	4.49	4.97
	0.3	5	93-99	96.5	2.57	2.66
BF 500-13 (500M62)	0.03 (LOQ)	5	91-99	95.9	2.90	3.03
	0.3	5	97-101	98.9	2.03	2.05
BF 500-14 (500M76)	0.03 (LOQ)	5	73-92	83.6	7.36	8.80
	0.3	5	92-117	103	9.21	8.93
BF 500-15 (500M78)	0.03 (LOQ)	5	Not included as an analyte			
	0.3	5				

Data (uncorrected recovery results, pp. 36-38) were obtained from Tables 1-2, pp. 31-34 of MRID 50088705.

1 Pyraclostrobin (BAS 500 F) = Methyl 2-[1-(4-chlorophenyl)pyrazol-3-yloxy]methyl-N-methoxycarbamate; BF 500-5 (Reg. No. 298327) = 1-(4-Chlorophenyl)-1H-pyrazol-3-ol; BF 500-12 (500M59; Reg. No. 412053) = Methyl N-(2-[[1-(4-hydroxyphenyl)-1H-pyrazol-3-yl]oxymethyl]phenyl) N-methoxy carbamate; BF 500-11 (500M60; Reg. No. 411847) = Methyl N-methoxy N-{2-[(1H-pyrazol-3-yl)oxymethyl]phenyl}carbamate; BF 500-13 (500M62; Reg. No. 412785) = Methyl N-[2-(1H-pyrazol-3-yloxy)methyl]phenylcarbamate; BF 500-14 (500M76; Reg. No. 413038) = Methyl N-(2-[2-(4-chlorophenyl)-5-oxo-2,5-dihydro-pyrazol-1-ylmethyl]-phenyl) N-methoxy carbamate; and BF 500-15 (500M78; Reg. No. 377613) = 1-(4-Hydroxyphenyl)-1H-pyrazol-3-ol.

2 The surface (lake) water (pH 6.9; 33 mg equiv. CaCO₃/L; 7.9 ppm total organic carbon) was collected from Jordan Lake, Cary, North Carolina, and drinking (well) water (pH 6.8; <1 mg equiv. CaCO₃/L; 0.9 ppm total organic carbon) was collected from BASF Research Triangle Park, North Carolina (p. 18; Appendix B, pp. 235-237). Water samples were characterized by Agvise Laboratories, Northwood, North Dakota.

3 Two ion transitions were monitored for each analyte (quantitation and confirmatory, respectively) as follows: *m/z* 389→195 and *m/z* 389→164 for pyraclostrobin (BAS 500 F); *m/z* 196→154 and *m/z* 196→118 for BF 500-5 (Reg. No. 298327); *m/z* 370→194 and *m/z* 370→278 for BF 500-12 (500M59; Reg. No. 412053); *m/z* 278→194 and *m/z* 278→149 for BF 500-11 (500M60; Reg. No. 411847); *m/z* 248→132 and *m/z* 248→164 for BF 500-13 (500M62; Reg. No. 412785); *m/z* 389→242 and *m/z* 389→301 for BF 500-14 (500M76; Reg. No. 413038); and *m/z* 177→135 and *m/z* 177→132 for BF 500-15 (500M78; Reg. No. 377613), respectively.

4 BF 500-5 was not included as an analyte in the ECM MRID 50088704; however, BF 500-5 was included as an analyte in BASF Method L0182/02 which was the ECM cited by the ILV. BASF Method L0182/01 and BASF Method L0182/02 appeared to be the same, except for the inclusion or exclusion of BF 500-5.

III. Method Characteristics

The LOQ and LOD for pyraclostrobin (BAS 500 F) in water were 0.003 µg/L and 0.0006 µg/L, respectively, in the ECM and ILV (p. 5; Appendix III, pp. 102, 116 of MRID 50088704; p. 7 of MRID 50088705). The LOQ and LOD for pyraclostrobin metabolites BF 500-12 (500M59; Reg. No. 412053), BF 500-11 (500M60; Reg. No. 411847), BF 500-13 (500M62; Reg. No. 412785), BF

500-14 (500M76; Reg. No. 413038), and BF 500-15 (500M78; Reg. No. 377613) in water were 0.03 µg/L and 0.006 µg/L, respectively, in the ECM and ILV. The LOQ and LOD for pyraclostrobin metabolite BF 500-5 (Reg. No. 298327) in water were 0.03 µg/L and 0.006 µg/L, respectively, in the ILV; BF 500-5 was not included as analyte in the ECM. In the ECM and ILV, the LOQ was defined as the lowest fortification level successfully tested, and the LOD was set to 20% of the LOQ. No calculations or comparisons to background levels were reported to justify the LOQ and LOD for the method in the ECM and ILV.

Table 4. Method Characteristics

Analyte ¹		Pyraclostrobin (BAS 500 F)	BF 500-12 (500M59; Reg. No. 412053)	BF 500-11 (500M60; Reg. No. 411847)	BF 500-13 (500M62; Reg. No. 412785)	BF 500-14 (500M76; Reg. No. 413038)	BF 500-15 (500M78; Reg. No. 377613)	BF 500-5 (Reg. No. 298327)		
Limit of Quantitation (LOQ)	ECM	0.003 µg/L	0.03 µg/L					Not performed		
	ILV		0.03 µg/L							
Limit of Detection (LOD)	ECM	0.0006 µg/L	0.006 µg/L					Not performed		
	ILV		0.006 µg/L							
Linearity (calibration curve r ² and concentration range)	ECM	Ground Water	r ² = 0.9987 (Q) r ² = 0.9995 (C)	r ² = 0.9995 (Q) r ² = 0.9991 (C)	r ² = 0.9995 (Q) r ² = 0.9996 (C)	r ² = 0.9998 (Q) r ² = 0.9989 (C)	r ² = 0.9994 (Q) r ² = 0.9988 (C)	r ² = 0.9995 (Q) r ² = 0.9990 (C)	Not performed	
		Surface Water	r ² = 0.9997 (Q) r ² = 0.9990 (C)	r ² = 0.9998 (Q) r ² = 0.9997 (C)	r ² = 0.9998 (Q) r ² = 0.9996 (C)	r ² = 0.9995 (Q) r ² = 0.9981 (C)	r ² = 0.9979 (Q) & C)	r ² = 0.9996 (Q) r ² = 0.9990 (C)		
		Drinking Water	r ² = 0.9995 (Q) r ² = 0.9989 (C)	r ² = 0.9997 (Q) r ² = 0.9996 (C)	r ² = 0.9996 (Q) r ² = 0.9993 (C)	r ² = 0.9990 (Q) r ² = 0.9987 (C)	r ² = 0.9986 (Q) r ² = 0.9976 (C)	r ² = 0.9997 (Q) r ² = 0.9999 (C)		
	Concentration		0.0005-0.005 ng	0.005-0.05 ng						
	ILV ²	Surface Water	r ² = 0.9954 (Q) r ² = 0.9966 (C)	r ² = 0.9946 (Q) r ² = 0.9928 (C)	r ² = 0.9976 (Q) r ² = 0.9990 (C)	r ² = 0.9984 (Q) r ² = 0.9966 (C)	r ² = 0.9928 (Q) r ² = 0.9932 (C)	r ² = 0.9825 (Q) r ² = 0.9853 (C)		r ² = 0.9948 (Q) r ² = 0.9888 (C)
		Drinking Water	r ² = 0.9970 (Q) r ² = 0.9958 (C)	r ² = 0.9942 (Q) r ² = 0.9938 (C)	r ² = 0.9960 (Q) r ² = 0.9956 (C)	r ² = 0.9980 (Q) r ² = 0.9964 (C)	r ² = 0.9868 (Q) r ² = 0.9870 (C)	Not performed		r ² = 0.9924 (Q) r ² = 0.9964 (C)
Concentration		0.005-0.1 ng/mL	0.05-1.0 ng/mL							
Repeatable	ECM ³	Yes at LOQ and 10×LOQ (uncharacterized ground, surface and drinking water)						Not performed		
	ILV ^{4,5}	Yes at LOQ and 10×LOQ (characterized surface and drinking water)					Yes at LOQ and 10×LOQ (characterized surface water) ⁶		Yes at LOQ and 10×LOQ (characterized surface and drinking water)	
Reproducible		Yes at LOQ and 10×LOQ						Could not be determined		

Analyte ¹		Pyraclostrobin (BAS 500 F)	BF 500-12 (500M59; Reg. No. 412053)	BF 500-11 (500M60; Reg. No. 411847)	BF 500-13 (500M62; Reg. No. 412785)	BF 500-14 (500M76; Reg. No. 413038)	BF 500-15 (500M78; Reg. No. 377613)	BF 500-5 (Reg. No. 298327)	
Specific	ECM	No 10×LOQ chromatograms were provided.						Yes, no matrix interferences were observed in the ground or drinking water matrices; minor baseline noise was noted. Major baseline noise which interfered with peak integration was observed in the surface water matrix. ⁷	Not performed
		Yes, no matrix interferences were observed. Minor baseline interference around the analyte peak was observed in the majority of C chromatograms, especially in surface water.							
	ILV ⁸	Yes, no matrix interferences were observed in the surface and drinking water matrices; minor baseline noise surrounded the analyte peak in Q chromatograms.	Yes, matrix interferences were <2% of the LOQ in surface water and <7% of the LOQ in drinking water; minor baseline noise surrounded the analyte peak in C chromatograms.	Yes, matrix interferences were <2% of the LOQ in surface and drinking water matrices. Baseline noise surrounded the analyte peak in C chromatograms.	Yes, no matrix interferences were observed in the surface water matrix, and matrix interferences were <1% of the LOQ in the drinking water matrix.	No matrix interferences were observed in the surface water matrix; however, major baseline noise which interfered with peak integration was observed. ⁹ Matrix interferences were <13% in the drinking water matrix; analyte peak was small compared to	Yes, no matrix interferences were observed in the surface water matrix; however, major baseline noise which interfered with peak integration was observed. ¹¹ No fortification in drinking water was performed.	No matrix interferences were observed in the surface and drinking water matrices; however, analyte peak was small compared to baseline noise. ¹²	

Analyte ¹	Pyraclostrobin (BAS 500 F)	BF 500-12 (500M59; Reg. No. 412053)	BF 500-11 (500M60; Reg. No. 411847)	BF 500-13 (500M62; Reg. No. 412785)	BF 500-14 (500M76; Reg. No. 413038)	BF 500-15 (500M78; Reg. No. 377613)	BF 500-5 (Reg. No. 298327)
					baseline noise. ¹⁰		

Data were obtained from p. 5; Appendix III, pp. 102, 116; Tables 1-6, pp. 22-24 (recovery data); Appendix I, pp. 26-37 (calibration curves & standards); Appendix I, pp. 38-73 (chromatograms); Appendix II, pp. 74-91 (correlation coefficients) of MRID 50088704; pp. 7, 11, 30, 35-36; Tables 1-2, pp. 31-34 (recovery data); Figures 11.1-11.13, pp. 83-95 (calibration curves); Figures 11.14-11.143, pp. 96-225 (chromatograms) of MRID 50088705. Q = Quantitation ion transition; C = Confirmatory ion transition.

- 1 Pyraclostrobin (BAS 500 F) = Methyl 2-[1-(4-chlorophenyl)pyrazol-3-yloxymethyl]-N-methoxycarbamate; BF 500-5 (Reg. No. 298327) = 1-(4-Chlorophenyl)-1H-pyrazol-3-ol; BF 500-12 (500M59; Reg. No. 412053) = Methyl N-(2-[[1-(4-hydroxyphenyl)-1H-pyrazol-3-yl]oxymethyl]phenyl) N-methoxy carbamate; BF 500-11 (500M60; Reg. No. 411847) = Methyl N-methoxy N-{2-[(1H-pyrazol-3-yl)oxymethyl]phenyl} carbamate; BF 500-13 (500M62; Reg. No. 412785) = Methyl N-[2-(1H-pyrazol-3-yloxymethyl)phenyl]carbamate; BF 500-14 (500M76; Reg. No. 413038) = Methyl N-(2-[2-(4-chlorophenyl)-5-oxo-2,5-dihydro-pyrazol-1-ylmethyl]-phenyl) N-methoxy carbamate; and BF 500-15 (500M78; Reg. No. 377613) = 1-(4-Hydroxyphenyl)-1H-pyrazol-3-ol.
 - 2 Correlation coefficients (r²) values were reviewer-calculated from r values provided in the study report (Figures 11.1-11.13, pp. 83-95 of MRID 50088705; DER Attachment 2). Calibration curves were matrix-based.
 - 3 In the ECM, ground water was collected from the Schifferstadt Water Supply, surface water was collected from Kelmetschweiher, and tap water was collected from Limburgerhof (p. 14 of MRID 50088704). The water matrices were not characterized; the water sources were not further specified.
 - 4 In the ILV, surface (lake) water (pH 6.9; 33 mg equiv. CaCO₃/L; 7.9 ppm total organic carbon) was collected from Jordan Lake, Cary, North Carolina, and drinking (well) water (pH 6.8; <1 mg equiv. CaCO₃/L; 0.9 ppm total organic carbon) was collected from BASF Research Triangle Park, North Carolina (p. 18; Appendix B, pp. 235-237 of MRID 50088705). Water samples were characterized by Agvise Laboratories, Northwood, North Dakota.
 - 5 In the ILV, the method for pyraclostrobin and its metabolites BF 500-5, BF 500-12, BF 500-11, BF 500-13, BF 500-14, and BF 500-15 was validated in the first trial in surface water with the addition of a pyraclostrobin metabolite BF 500-5 (Reg. No. 298327) and insignificant modifications to the analytical instrumentation (pp. 11, 30 of MRID 50088705). The method for pyraclostrobin and its metabolites BF 500-5, BF 500-12, BF 500-11, BF 500-13, and BF 500-14 was validated in the second trial in drinking water with the addition of a pyraclostrobin metabolite BF 500-5 (Reg. No. 298327) and insignificant modifications to the analytical instrumentation. BF 500-5 was not included as analyte in the ECM; therefore, two sets of validation data were not included for BF 500-5.
 - 6 BF 500-15 not included in drinking water validation, only the surface water validation.
 - 7 Appendix I, pp. 70-71 of MRID 50088704.
 - 8 Matrix interference percentages were based on peak areas in provided chromatograms.
 - 9 Figures 11.72-11.73, pp. 154-155 of MRID 50088705.
 - 10 Figures 11.142-11.143, pp. 224-225 of MRID 50088705.
 - 11 Figures 11.82-11.83, pp. 164-165 of MRID 50088705.
 - 12 Figures 11.32-11.33, pp. 114-115 and Figures 11.102-11.103, pp. 184-185 of MRID 50088705.
- Linearity is satisfactory when r² ≥ 0.995.

IV. Method Deficiencies and Reviewer's Comments

1. The ILV MRID 50099705 was an ILV of BASF Method L0182/02, not BASF Method L0182/01; however, BASF Method L0182/01 appeared to be the same as BASF Method L0182/02, except for the addition of BF 500-5 as an analyte (pp. 7, 18, 38; Appendix D, pp. 280-281 of MRID 50088705). Therefore, the reviewer considered the ILV MRID 50099705 to be an independent validation of BASF Method L0182/01, as well as BASF Method L0182/02.

The reviewer noted that the ILV reported that mobile phase B was 0.1% formic acid prepared in acetonitrile versus methanol, but the ILV did not specify to which method it was referring (Appendix D, p. 281 of MRID 50088705). If the method was BASF Method L0182/01, the reviewer believed that the method modification of a mobile phase change did not actually occur since mobile phase B of was 0.1% formic acid prepared in acetonitrile (see Reviewer's Comment #9).

2. BF 500-5 was not included as an analyte in ECM MRID 50088704 which was a validation of BASF Method L0182/01. BF 500-5 was included as an analyte in the ILV MRID 50099705 which was an ILV of BASF Method L0182/02. Two sets of performance data were not included for the portion of BASF Method L0182/02 which included BF 500-5 as an analyte; only the ILV data was submitted. OCSPP guidelines state that two sets of performance data are submitted, one for the initial or other internal validation and one for the ILV.
3. In the ILV, linearity was not satisfactory for BF 500-5 [surface water $r^2 = 0.9948$ (Q) and 0.9888 (C), drinking water $r^2 = 0.9924$ (Q)], BF 500-12 [surface water $r^2 = 0.9946$ (Q) and 0.9928 (C), drinking water $r^2 = 0.9942$ (Q) and 0.9938 (C)] BF 500-14 [surface water $r^2 = 0.9928$ (Q) and 0.9932 (C), drinking water $r^2 = 0.9868$ (Q) and 0.9870 (C)] and BF 500-15 [surface water $r^2 = 0.9825$ (Q) and 0.9853 (C); p. 36; Figures 11.1-11.13, pp. 83-95 of MRID 50088705; DER Attachment 2]. Linearity is satisfactory when $r^2 \geq 0.995$.
4. The specificity of the method for BF 500-14, BF 500-15 and BF 500-5 was not supported by the ILV representative chromatograms. Major baseline noise interfered with peak integration of BF 500-14 and BF 500-15 in the surface water matrix (Figures 11.72-11.73, pp. 154-155; Figures 11.82-11.83, pp. 164-165 of MRID 50088705). In the drinking water matrix, the analyte peak of BF 500-14 was small compared to baseline noise (Figures 11.142-11.143, pp. 224-225). The analyte peak of BF 500-5 was small compared to baseline noise in both water matrices (Figures 11.32-11.33, pp. 114-115; Figures 11.102-11.103, pp. 184-185).
5. No $10 \times \text{LOQ}$ chromatograms were provided in the ECM. OCSPP guidelines state that representative chromatograms should be provided for reagent blanks, matrix blanks, standard curves, and spiked samples at the LOQ and $10 \times \text{LOQ}$ for all analytes in each matrix.

6. The specificity of the method for BF 500-15 in the surface water matrix was not supported by the ECM representative chromatograms since major baseline noise interfered with peak integration of BF 500-15 (Appendix I, pp. 70-71 of MRID 50088704).
7. Water matrices were not characterized in the ECM (p. 14 of MRID 50088704).
8. The estimations of LOQ and LOD in ECM and ILV were not based on scientifically acceptable procedures as defined in 40 CFR Part 136 (p. 5; Appendix III, pp. 102, 116 of MRID 50088704; p. 7 of MRID 50088705). In the ECM and ILV, the LOQ was defined as the lowest fortification level successfully tested, and the LOD was set to 20% of the LOQ. No calculations or comparisons to background levels were reported to justify the LOQ and LOD for the method in the ECM and ILV.
9. In the ECM, BASF Method L0182/01 (non-GLP) was provided (Appendix III, pp. 101-). The reviewer noted the following apparent typographical error: the composition of HPLC mobile phase B was titled as 0.1% Formic acid in methanol instead of 0.1% Formic acid in acetonitrile (Appendix III, p. 108). The reviewer assumed that this was a typographical error since the preparation details list acetonitrile instead of methanol and the mobile phase in the Instrumentation and Conditions table listed ACN/formic acid 1000/1, v/v, as Mobile Phase B (p. 19; Appendix III, pp. 108, 113). Additionally, the reviewer believed that the ILV was misled by this uncorrected typographical error if the ILV was referring to BASF Method L0182/01 when reporting that mobile phase B was changed from 0.1% formic acid prepared in methanol to 0.1% formic acid prepared in acetonitrile (see Reviewer's Comment #1; Appendix D, p. 281)
10. The Protocol Change Forms were provided in Appendix D of the ILV (Appendix D, pp. 279-288 of MRID 50088705). The changes involved the addition of BF 500-5 as an analyte, the removal of tap water as a test matrix, the removal of the ground water fortification with BF 500-15, change of analyte naming and correction of errors.
11. In the ECM, the analytical standards in acetonitrile:water (20:80, v:v) were found to be stable for at least two weeks when stored refrigerated in the dark (temperature not specified; p. 20; Tables 43-48, pp. 92-94 of MRID 50088704). The extracts of the water samples were found to be stable (>70% recovery) after one week under refrigeration (temperature not specified; p. 20; Tables 49-54, pp. 95-100).
12. The ILV study author documented all communications between the ILV and Study Monitor (Appendix F, pp. 303-305 of MRID 50088705). Communications consisted of protocol questions and changes, trial problems, sponsor suggestions, and trial completions. No communications between the ILV and ECM staff occurred.
13. It was reported for the ILV that analysis of 13 samples required *ca.* 8 hours of work including the calculation of the results and reporting of all raw data under GLP (p. 35 of MRID 50088705).

V. References

- 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.

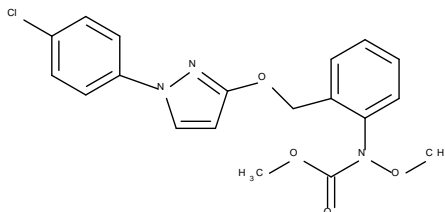
Attachment 1: Chemical Names and Structures**Pyraclostrobin (BAS 500 F)**

IUPAC Name: Methyl 2-[1-(4-chlorophenyl)pyrazol-3-yloxymethyl]-N-methoxycarbanilate

CAS Name: Methyl N-[2-[[[1-(4-chlorophenyl)-1H-pyrazol-3-yl]oxy]methyl]phenyl]-N-methoxycarbamate

CAS Number: 175013-18-0

SMILES String: COC(=O)N(c1ccccc1COc2ccn(n2)c3ccc(cc3)Cl)OC

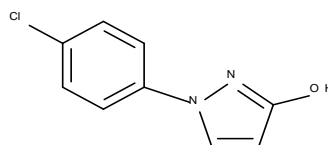
**BF 500-5 (Reg. No. 298327)**

IUPAC Name: Not reported

CAS Name: 1-(4-Chlorophenyl)-1H-pyrazol-3-ol

CAS Number: Not reported

SMILES String: c1cc(ccc1n2ccc(n2)O)Cl

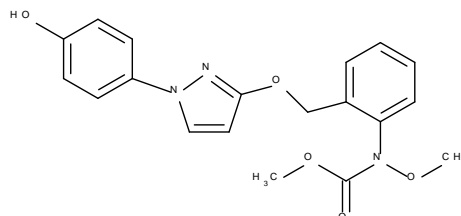
**BF 500-12 (500M59; Reg. No. 412053)**

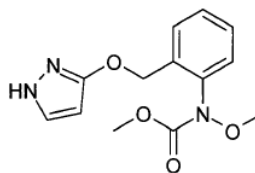
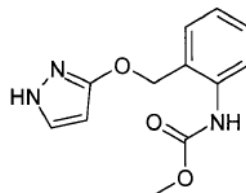
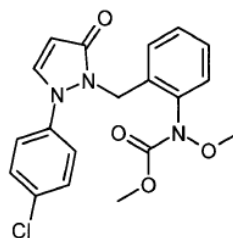
IUPAC Name: Methyl N-(2-{[1-(4-hydroxyphenyl)-1H-pyrazol-3-yl]oxymethyl}phenyl)-N-methoxy carbamate

CAS Name: Methyl N-[[[1-(4-hydroxyphenyl)pyrazol-3-yl]oxy]-tolyl]-methoxycarbamate

CAS Number: Not reported

SMILES String: COC(=O)N(c1ccccc1COc2ccn(n2)c3ccc(O)c3)OC



BF 500-11 (500M60; Reg. No. 411847)**IUPAC Name:** Methyl N-methoxy N-{2-[(1H-pyrazol-3-yl)oxymethyl]phenyl} carbamate**CAS Name:** Not reported**CAS Number:** 175013-17-9**SMILES String:** Not found**BF 500-13 (500M62; Reg. No. 412785)****IUPAC Name:** Methyl N-[2-(1H-pyrazol-3-yl)oxymethyl]phenyl]carbamate**CAS Name:** Not reported**CAS Number:** Not reported**SMILES String:** Not found**BF 500-14 (500M76; Reg. No. 413038)****IUPAC Name:** Methyl N-(2-[2-(4-chlorophenyl)-5-oxo-2,5-dihydro-pyrazol-1-ylmethyl]-phenyl) N-methoxy carbamate**CAS Name:** Not reported**CAS Number:** Not reported**SMILES String:** Not found

BF 500-15 (500M78; Reg. No. 377613)**IUPAC Name:** 1-(4-Hydroxyphenyl)-1H-pyrazol-3-ol**CAS Name:** Not reported**CAS Number:** Not reported**SMILES String:** Not found