

Analytical method for dimethenamid-P (BAS 656 H) and its metabolites M23, M27, and M31 in water

Reports: ECM: EPA MRID No.: 50362702. Class, T., and S. Jooss. 2012. Determination of Dimethenamid-P and its Metabolites M23, M27 and M31 in Water. BASF Registration Document No. 2012/1278546. PTRL Europe ID: P 2711 G. BASF IPMS: 353459. Report prepared by PTRL Europe, Ulm, Germany, and sponsored and submitted by BASF Corporation, Research Triangle Park, North Carolina; 77 pages. Final report issued October 29, 2012.

ILV: EPA MRID No. 50362703. Lui, W., and Y. Shi. 2017. Independent Laboratory Validation of BASF Analytical Method: "Determination of Dimethenamid-p and its Metabolites M23, M27 and M31 in Water" for Dimethenamid-p only. BASF Study No.: 732240. Alliance Pharma Study No.: 140232. BASF Registration Document No. 2016/7011586. Report prepared by Alliance Pharma, Malvern, Pennsylvania, and sponsored and submitted by BASF Crop Protection, Research Triangle Park, North Carolina; 46 pages. Final report issued March 18, 2014; amended report issued June 2, 2017.

Document No.: MRIDs 50362702 & 50362703

Guideline: 850.6100

Statements: ECM: The study was conducted in accordance with German Good Laboratory Practice (GLP) standards, which are based on OECD, EPA FIFRA and FDA, and Japanese GLP, and EC guidance document on residue analytical methods (pp. 3, 6, 8; Appendix 2, p. 77 of MRID 50362702). Signed and dated GLP and Quality Assurance statements were provided (pp. 2-3, 6-8; Appendix 2, p. 77). An Authenticity statement was included with the Quality Assurance statement.

ILV: The study was conducted in accordance with USEPA FIFRA GLP (40 CFR Part 160) standards (p. 3 of MRID 50362703). Signed and dated No Data Confidentiality, GLP, Quality Assurance, Authenticity statements were provided (pp. 2-5).


Classification: This analytical method is classified as acceptable for the parent compound Dimethenamid-P and supplemental for metabolites M23, M27, and M31. There are not enough data to assess the validity of the metabolites because they were not addressed in the ILV report. However, the metabolites are not residues of concern.

PC Code: 129051

EFED Final Reviewer: Sheng Lin, Ph.D.,
Physical Scientist

Signature:
Date: 1/5/2021

CDM/CSS-Dynamac JV Reviewers: Lisa Muto,
Environmental Scientist

Signature: 
Date: 01/10/2019

Mary Samuel, M.S.,
Environmental Scientist

Signature: 

Date: 01/10/2019

This Data Evaluation Record may have been altered by the Environmental Fate and Effects Division subsequent to signing by CDM/CSS-Dynamac JV personnel. The CDM/CSS-Dynamac Joint Venture role does not include establishing Agency policies.

Executive Summary

The analytical method, BASF Registration Document No. 2012/1278546, is designed for the quantitative determination of dimethenamid-P (BAS 656 H) and its metabolites M23, M27, and M31 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 in water for all analytes. The ECM validated the method using two characterized water matrices, drinking and surface water. The ILV validated the method using one characterized water matrix, drinking water. The number of trials was not reported in the ILV; however, the reviewer believed that the ILV successfully validated the ECM in the first trial since the method was performed as written with only minor LC/MS instrument and parameter modifications. However, only the parent dimethenamid-P was included as an analyte in the ILV; M23, M27, and M31 were not included in the ILV trial. **An independent laboratory validation of BASF Registration Document No. 2012/1278546 including dimethenamid-P metabolites M23, M27, and M31 should be submitted to determine the reproducibility of the method for these analytes.** All submitted ECM and ILV data pertaining to precision, repeatability, reproducibility, linearity, and specificity was acceptable, except for specificity of M23 since significant baseline noise around the analyte peak which interfered with analyte integration and attenuation at LOQ was observed in ECM representative chromatograms of both waters.

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						
Dimethenamid-P (BAS 656 H)	50362702 ¹	50362703 ²		Water	29/10/2012	BASF Corporation	LC/MS/MS	0.03 µg/L
M23		None submitted ³						
M27								
M31								

¹ In ECM, the drinking water (pH 7.72, total hardness 2.33 mmol/L (Deutsche Härtegrade, 13.0°d), total organic carbon not reported) obtained from a PTRL Europe laboratory tap in Ulm, Germany, and the surface (river) water (pH 8.09, total hardness 1.26 mmol/L (Deutsche Härtegrade, 7.1°d), total organic carbon 1.30 mg/L) obtained from Brenz River in Herbrechtingen, Germany, were used in the study and characterized by Institute Alpha, Ulm, Germany (p. 14 of MRID 50362702).

³ In the ILV, the water matrix used in the study were provided by the Sponsor, BASF (p. 12; Appendix 2, p. 36 of MRID 50362703). The drinking water (pH 7.2, hardness 25 mg equivalent CaCO₃/L, total organic carbon 3.1 ppm) was characterized by Agvise Laboratories, Northwood, North Dakota. The source of the water was not further described.

³ M23, M27, and M31 were not included in the ILV trial.

I. Principle of the Method

Water samples (1 mL) containing 0.1% formic acid in an autosampler vial were fortified (0.020 mL of 1.5 ng/mL or with 0.020 mL of 15 ng/mL fortification solution), as necessary (p. 17 of MRID 50362702). The samples were vortexed and analyzed by LC/MS/MS.

Analytes were identified and quantified by LC/MS/MS using an Agilent 1200 SL HPLC coupled with an Applied Biosystems MDS Sciex API 5500 MS (pp. 17-18 of MRID 50362702). The following conditions were employed: Agilent Zorbax SB C18 analytical column (150 mm x 4.6 mm, 5 µm particle size; column temperature 40°C) eluted with a gradient mobile phase of (A) 0.1% formic acid in water and (B) 0.1% formic acid in methanol [time, percent A:B; time 0.00 min. 80:20, 5.0-10.0 min. 20:80, 10.1-15.0 min. 80:20] using an injection volume of 100 µL and Turbo spray ESI ionization MRM scan mode (positive for parent and negative for metabolites). Analytes were identified using two ion transitions (quantitation and confirmation, respectively): m/z 276→244 and m/z 276→168 for dimethenamid-P (BAS 656 H), m/z 270→198 and m/z 270→166 for M23, m/z 320.1→121 and m/z 320.1→80 for M27, and m/z 346→240 and m/z 346→198 for M31. Expected retention times were *ca.* 6.5, 5.4, 5.1, and 5.2 minutes for dimethenamid-P, M23, M27, and M31, respectively.

In the ILV, the ECM was performed as written, except for minor LC/MS instrument and parameter modifications; however, only the parent dimethenamid-P was included as an analyte (pp. 7, 12, 14; Table 4, p. 20 of MRID 50362703). Analytes were identified and quantified by LC/MS/MS using a Shimadzu LC-30AD Series HPLC coupled with a Sciex API 5500 MS. The following conditions were employed: Zorbax Eclipse XDB C18 analytical column (150 mm x 3 mm, 3.5 µm particle size; column temperature not reported) eluted with a gradient mobile phase

of (A) 0.1% formic acid in water and (B) 0.1% formic acid in methanol [time, percent A:B; time 0.00 min. 80:20, 5.0-8.6 min. 20:80, 9.1-11.0 min. 0:100, 11.1-15.0 min. 80:20] using an injection volume of 50 μ L and positive electrospray ionization MRM scan mode. The LC/MS conditions were the same as the ECM; however, the MS temperature was reported as 500°C. Dimethenamid-P was identified using two ion transitions (quantitation and confirmation, respectively): m/z 276.0 \rightarrow 244.1 and m/z 276.0 \rightarrow 168.1; the MS transitions were similar to those of ECM. Expected retention time was *ca.* 8.04 minutes for dimethenamid-P.

The Limit of Quantification (LOQ) for dimethenamid-P and its metabolites [M23, M27, and M31] in water was 0.03 μ g/L in the ECM and ILV (p. 12 of MRID 50362702; p. 7 of MRID 50362703). The Limit of Determination (LOD) for all analytes in water and water was reported as 0.009 μ g/L (30% of the LOQ) in the ECM and ILV.

II. Recovery Findings

ECM (MRID 50362702): Mean recoveries and RSDs were within guideline requirements [means between 70% and 120% and relative standard deviations (RSD) \leq 20%] for analysis of dimethenamid-P (BAS 656 H), M23, M27, and M31 at fortification levels of 0.03 μ g/L (LOQ) and 0.3 μ g/L (10 \times LOQ) in two water matrices (Tables 1-2, pp. 23-26). All analytes were identified using two ion transitions; performance data (recovery results) for the quantitation and confirmation ion analyses were comparable, but the reviewer noted that the RSD for M31 in drinking water differed between quantitation and confirmation analyses. The drinking water (pH 7.72, total hardness 2.33 mmol/L (Deutsche Härtegrade, 13.0°d), total organic carbon not reported) obtained from a PTRL Europe laboratory tap in Ulm, Germany, and the surface (river) water (pH 8.09, total hardness 1.26 mmol/L (Deutsche Härtegrade, 7.1°d), total organic carbon 1.30 mg/L) obtained from Brenz River in Herbrechtingen, Germany, were used in the study and characterized by Institute Alpha, Ulm, Germany (p. 14).

ILV (MRID 50362703): Mean recoveries and RSDs were within guideline requirements for analysis of dimethenamid-P (BAS 656 H) at fortification levels of 0.03 μ g/L (LOQ) and 0.3 μ g/L (10 \times LOQ) in one water matrix (p. 7). Dimethenamid-P was identified using two ion transitions; performance data (recovery results) for the quantitation and confirmation ion analyses were comparable. The water matrix used in the study were provided by the Sponsor, BASF (p. 12; Appendix 2, p. 36 of MRID 50362703). The drinking water (pH 7.2, hardness 25 mg equivalent CaCO_3 /L, total organic carbon 3.1 ppm) was characterized by Agvise Laboratories, Northwood, North Dakota. The source of the water was not further described. The number of trials was not reported in the ILV; however, the reviewer believed that the ILV successfully validated the ECM in the first trial since the method was performed as written with only minor LC/MS instrument and parameter modifications (pp. 7, 12, 14-15). However, only the parent dimethenamid-P was included as an analyte; M23, M27, and M31 were not included in the ILV trial.

Table 2. Initial Validation Method Recoveries for Dimethenamid-P (BAS 656 H) and its Metabolites M23, M27, and M31 in Water^{1,2}

Analyte	Fortification Level (µg/L)	Number of Tests	Recovery Range (%)	Mean Recovery (%)	Standard Deviation (%) ³	Relative Standard Deviation (%)
Drinking Water						
Quantitation Ion Transition						
Dimethenamid-P (BAS 656 H)	0.03 (LOQ)	5	102-118	109	6	5
	0.3	5	105-114	110	5	4
M23	0.03 (LOQ)	5	104-111	107	3	3
	0.3	5	91-111	98	6	6
M27	0.03 (LOQ)	5	98-114	106	8	7
	0.3	5	89-103	96	6	6
M31	0.03 (LOQ)	5	98-114	107	7	6
	0.3	5	91-104	97	5	5
Confirmation Ion Transition						
Dimethenamid-P (BAS 656 H)	0.03 (LOQ)	5	101-116	108	6	6
	0.3	5	104-115	110	5	5
M23	0.03 (LOQ)	5	92-116	103	9	8
	0.3	5	92-104	98	6	6
M27	0.03 (LOQ)	5	101-116	110	6	6
	0.3	5	89-104	97	7	7
M31	0.03 (LOQ)	5	85-115	97	11	12
	0.3	5	84-98	91	6	6
Surface Water						
Quantitation Ion Transition						
Dimethenamid-P (BAS 656 H)	0.03 (LOQ)	5	99-116	109	7	7
	0.3	5	105-116	109	5	4
M23	0.03 (LOQ)	5	99-116	108	8	7
	0.3	5	96-109	104	6	6
M27	0.03 (LOQ)	5	106-113	110	3	3
	0.3	5	99-111	105	6	5
M31	0.03 (LOQ)	5	101-119	110	8	8
	0.3	5	95-110	103	6	6
Confirmation Ion Transition						
Dimethenamid-P (BAS 656 H)	0.03 (LOQ)	5	104-117	110	6	5
	0.3	5	104-116	110	5	5
M23	0.03 (LOQ)	5	93-112	102	8	8
	0.3	5	97-110	103	6	6
M27	0.03 (LOQ)	5	98-117	110	7	7
	0.3	5	99-113	106	6	6
M31	0.03 (LOQ)	5	92-110	99	7	7
	0.3	5	96-109	103	5	5

Data (uncorrected recovery results, p. 19) were obtained from Tables 1-2, pp. 23-26 of MRID 50362702 and DER Attachment 2.

¹ The drinking water (pH 7.72, total hardness 2.33 mmol/L (Deutsche Härtegrade, 13.0°d), total organic carbon not reported) obtained from a PTRL Europe laboratory tap in Ulm, Germany, and the surface (river) water (pH 8.09, total hardness 1.26 mmol/L (Deutsche Härtegrade, 7.1°d), total organic carbon 1.30 mg/L) obtained from Brenz

- River in Herbrechtingen, Germany, were used in the study and characterized by Institute Alpha, Ulm, Germany (p. 14).
- 2 Analytes were identified using two ion transitions (quantitation and confirmation, respectively): m/z 276→244 and m/z 276→168 for dimethenamid-P (BAS 656 H), m/z 270→198 and m/z 270→166 for M23, m/z 320.1→121 and m/z 320.1→80 for M27, and m/z 346→240 and m/z 346→198 for M31.
- 3 Standard deviations were reviewer-calculated since these values were not reported in the study report. The rules of significant figures were followed.

Table 3. Independent Validation Method Recoveries for Dimethenamid-P (BAS 656 H) in Water^{1,2,3}

Analyte	Fortification Level (µg/L)	Number of Tests	Recovery Range (%)	Mean Recovery (%)	Standard Deviation (%) ¹	Relative Standard Deviation (%)
Drinking Water						
Quantitation Ion Transition						
Dimethenamid-P (BAS 656 H)	0.03 (LOQ)	5	90-103	97	6.0	6.1
	0.3	5	95-103	99	2.8	2.8
Confirmation Ion Transition						
Dimethenamid-P (BAS 656 H)	0.03 (LOQ)	5	93-100	97	2.8	2.9
	0.3	5	98-103	100	1.9	1.9

Data (recovery results were corrected when residues were quantified in the controls, pp. 12-13) were obtained from p. 7 of MRID 50362703.

- 1 The water matrix used in the study were provided by the Sponsor, BASF (p. 12; Appendix 2, p. 36). The drinking water (pH 7.2, hardness 25 mg equivalent CaCO₃/L, total organic carbon 3.1 ppm) was characterized by Agvise Laboratories, Northwood, North Dakota. The source of the water was not further described.
- 2 Dimethenamid-P was identified using two ion transitions (quantitation and confirmation, respectively): m/z 276.0→244.1 and m/z 276.0→168.1; the MS transitions were similar to those of ECM.
- 3 Dimethenamid-P metabolites M23, M27, and M31 were not included in the ILV.

III. Method Characteristics

The LOQ for dimethenamid-P and its metabolites [M23, M27, and M31] in water and water was 0.03 µg/L in the ECM and ILV (p. 12 of MRID 50362702; p. 7 of MRID 50362703). The LOD for all analytes in water and water was reported as 0.009 µg/L (30% of the LOQ). No calculations for the LOQ and LOD were provided in the ECM and ILV. The LOQ and LOD were reported in the ILV from the ECM without justification.

Table 4. Method Characteristics

Analyte		Dimethenamid-P (BAS 656 H)	M23	M27	M31
Limit of Quantitation (LOQ)		0.03 µg/L			
Limit of Detection (LOD)		0.009 µg/L (30% of the LOQ)			
Linearity (calibration curve r ² and concentration range) ¹	ECM	r ² = 0.9982 (Q) r ² = 0.9966 (C)	r ² = 0.9970 (Q & C)	r ² = 0.9974 (Q) r ² = 0.9964 (C)	r ² = 0.9976 (Q) r ² = 0.9982 (C)
	ILV	r ² = 0.9986 (Q) r ² = 0.9990 (C)	Not performed		
	Concentration Range	0.009-0.4 ng/mL			
Repeatable	ECM ²	Yes at LOQ and 10×LOQ (two characterized waters).			
	ILV ^{3,4}	Yes at LOQ and 10×LOQ (one characterized water)	Not performed		
Reproducible		Yes at LOQ and 10×LOQ.	Could not be determined; only one set of performance data was submitted.		
Specificity	ECM	Yes, matrix interferences were <17% of the LOQ (based on peak area). Minor baseline noise was noted around the analyte peak which interfered with analyte integration and attenuation at LOQ.	No , matrix interferences were <7% of the LOQ (based on peak area); however, significant baseline noise was noted around the analyte peak which interfered with analyte integration and attenuation at LOQ. ⁵	Yes, matrix interferences were <3% of the LOQ (based on peak area).	Yes, matrix interferences were <4% (Q) and <23% (C) of the LOQ (based on peak area).
	ILV	Yes, matrix interferences were <5% of the LOQ (based on peak area).	Not performed		

Data were obtained from p. 12 (LOQ/LOD); Tables 1-2, pp. 23-26 (recovery data); Figures 1-7, pp. 29-35 (calibration curve); Figures 17-40, pp. 45-68 (chromatograms) of MRID 50362702; p. 7 (LOQ/LOD & recovery data); Figure 1, p. 22 (calibration curves); Figures 2-6, pp. 23-31 (chromatograms) of MRID 50362703; DER Attachment 2. Q = Quantitation ion transition; C = Confirmatory ion transition.

1 Reported correlation coefficients were reviewer-calculated from r values reported in the study report (Figures 1-7, pp. 29-35 of MRID 50362702; p. 12; Figure 1, p. 22 of MRID 50362703; DER Attachment 2). Significant figures of r² were limited to four.

2 In ECM, the drinking water (pH 7.72, total hardness 2.33 mmol/L (Deutsche Härtegrade, 13.0°d), total organic carbon not reported) obtained from a PTRL Europe laboratory tap in Ulm, Germany, and the surface (river) water (pH 8.09, total hardness 1.26 mmol/L (Deutsche Härtegrade, 7.1°d), total organic carbon 1.30 mg/L) obtained from Brenz River in Herbrechtingen, Germany, were used in the study and characterized by Institute Alpha, Ulm, Germany (p. 14 of MRID 50362702).

3 In the ILV, the water matrix used in the study were provided by the Sponsor, BASF (p. 12; Appendix 2, p. 36 of MRID 50362703). The drinking water (pH 7.2, hardness 25 mg equivalent CaCO₃/L, total organic carbon 3.1 ppm) was characterized by Agvise Laboratories, Northwood, North Dakota. The source of the water was not further described.

- 4 The number of trials was not reported in the ILV; however, the reviewer believed that the ILV successfully validated the ECM in the first trial since the method was performed as written with only minor LC/MS instrument and parameter modifications (pp. 7, 12, 14-15 of MRID 50362703). However, only the parent dimethenamid-P was included as an analyte; M23, M27, and M31 were not included in the ILV trial.
- 5 Based on Figure 22, p. 50 and Figure 34, p. 62 of MRID 50362702.

IV. Method Deficiencies and Reviewer's Comments

1. For the analytical method BASF Registration Document No. 2012/1278546 for dimethenamid-P metabolites M23, M27, and M31, only one set of performance data was submitted, an ECM in MRID 50362702. In ILV MRID 50362703, M23, M27, and M31 were not included as analytes. OCSPP guidelines state that two sets of performance data should be submitted, one for the initial or other internal validation and one for the ILV. An independent laboratory validation of BASF Registration Document No. 2012/1278546 including dimethenamid-P metabolites M23, M27, and M31 should be submitted. These metabolites, however, are not residues of concern.
2. The specificity of the method was not supported by ECM representative chromatograms of M23 due to significant baseline noise around the analyte peak which interfered with analyte integration and attenuation at LOQ in both waters (Figure 22, p. 50; Figure 34, p. 62 of MRID 50362702). Additional sample processing may be required to enhance the specificity of the method for this analyte.
3. The number of ILV trials required to successfully validate the ECM was not reported in the ILV study report (pp. 7, 12, 14-15 of MRID 50362703). OCSPP guidelines state that maximum of three sample sets should be used to validate the ECM.
4. The estimation of LOQ and LOD in the ECM and ILV was not based on scientifically acceptable procedures as defined in 40 CFR Part 136 (p. 12 of MRID 50362702; p. 7 of MRID 50362703). No calculations for the LOQ and LOD were provided in the ECM and ILV. The LOQ and LOD were reported in the ILV from the ECM without justification. Detection limits should not be based on arbitrary values.

V. Reviewer's Comments

1. Matrix effects were studied in the ECM and determined to be insignificant for all analytes (p. 21 of MRID 50362702). Solvent-based calibration standards were used for analysis.
2. The ILV reported that communication between the ILV Study Director and Study Monitor consisted of protocol approval and the Study Monitor being notified of the successful completion of the ILV trial (p. 15 of MRID 50362703). No other communication was described.

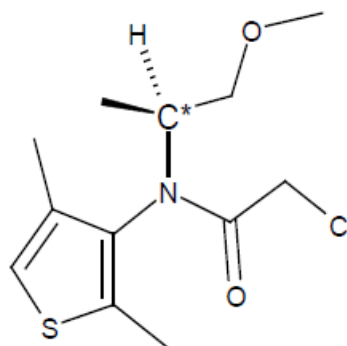
3. In the ECM, the time required to complete the extraction of one set of 12 samples required ca. 1 calendar day of work, including calculation of results (preparation, 2 hours; LC/MS/MS analysis, 5 hours; evaluation, 2 hours; p. 21 of MRID 50362702). The time requirement was not reported in the ILV.
4. Amendments to the ILV report included the correction of the calibration solution solvent and supporting documentation, as well as updates to the BASF Registration Document Number (p. 1; Appendices 4-5, pp. 42-46 of MRID 50362703).

VI. References

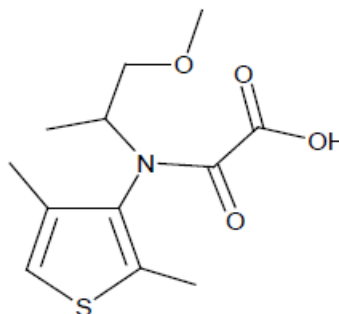
- U.S. Environmental Protection Agency. 2012. Ecological Effects Test Guidelines, OCSPP 850.6100, Environmental Chemistry Methods and Associated Independent Laboratory Validation. Office of Chemical Safety and Pollution Prevention, Washington, DC. EPA 712-C-001.
- 40 CFR Part 136. Appendix B. Definition and Procedure for the Determination of the Method Detection Limit-Revision 1.11, pp. 317-319.

Attachment 1: Chemical Names and Structures**Dimethenamid-P (BAS 656 H; Reg. No. 363851)**

IUPAC Name: (S)-2-chloro-N-(2,4-dimethyl-3-thienyl)-N-(2-methoxy-1-methylethyl)acetamide
CAS Name: Not reported
CAS Number: 163515-14-8
SMILES String: Not found

**M23 (Reg. No. 360715)**

IUPAC Name: N-(2,4-dimethyl-thiophen-3-yl)-N-(2-methoxy-1-methyl-ethyl)-oxalamic acid
CAS Name: Not reported
CAS Number: Not reported
SMILES String: Not found



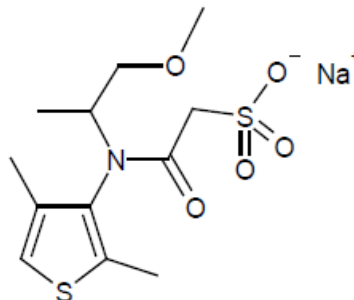
M27 (Reg. No. 360714)

IUPAC Name: Sodium [(2,4-dimethyl-thiophen-3-yl)-(2-methoxy-1-methyl-ethyl)-carbamoyl]-methanesulfonate

CAS Name: Not reported

CAS Number: Not reported

SMILES String: Not found

**M31 (Reg. No. 360712)**

IUPAC Name: [[(2,4-Dimethyl-thiophen-3-yl)-(2-methoxy-1-methyl-ethyl)-carbamoyl]-methanesulfinyl]-acetic acid

CAS Name: Not reported

CAS Number: Not reported

SMILES String: Not found

