

## 1. INTRODUCTION

### 1.1 Scope of the Method

BASF Analytical Method No. L0109/02 was developed to determine Dimethenamid-P (656 H) and its metabolites M23, M27 and M31 in soil and sediment using LC-MS/MS at BASF Crop Protection in Limburgerhof, Germany. This report represents the validation by an independent laboratory, Critical Path Services, LLC in Garnet Valley, Pennsylvania.

The independent lab validation was conducted using two fortification levels, at the LOQ (0.005 mg/kg) and ten times the LOQ (0.05 mg/kg) for soil and sediment. Five replicates were analyzed for each fortification level. Additionally, one reagent blank and two replicates of unfortified samples were examined.

### 1.2 Principle of the Method

Soil and sediment samples (5 g) were fortified, using a micropipette, with stock fortification solutions containing all analytes and then extracted with methanol/water (60:40, v/v). An aliquot of resulting solution was analyzed to determine the residues of BAS 656 H and its metabolites M23, M27 and M31 using LC-MS/MS. The transitions for BAS 656 H at  $m/z$  276  $\rightarrow$  244 and at  $m/z$  276  $\rightarrow$  168 were monitored in positive mode for primary and confirmation quantification, respectively. The transitions for M23 at  $m/z$  270  $\rightarrow$  198 and at  $m/z$  270  $\rightarrow$  166 were monitored in negative mode for primary and confirmation quantification, respectively. The transitions for M27 at  $m/z$  320  $\rightarrow$  121 and at  $m/z$  320  $\rightarrow$  80 were monitored in negative mode for primary and confirmation quantification, respectively. The transitions for M31 at  $m/z$  346  $\rightarrow$  240 and at  $m/z$  346  $\rightarrow$  198 were monitored in negative mode for primary and confirmation quantification, respectively.

### 1.3 Specificity

To demonstrate the specificity of the analytical method, one primary and a confirmatory mass transition were monitored for BAS 656 H and each of its metabolites M23, M27 and M31. The method was able to accurately determine residues of BAS 656 H and its metabolites M23, M27 and M31, and no interference was observed at the retention time of the analyte peaks. The product ion spectra of each analyte to justify the transitions being monitored can be found in the validation of this method (Reference 1)

## 2. REFERENCE SUBSTANCE AND SAMPLING HISTORY

### 2.1 Test Systems

The test systems in this study were soil and sediment.

The control samples were provided by BASF. The soil and sediment samples were both received on June 8, 2017. Upon arrival at the laboratory, the samples were opened, inspected, and checked against enclosed shipping forms. The test systems were received

frozen and were stored under frozen conditions at all times, unless necessary for laboratory analysis.

## 2.2 Test and Reference Substances

The standard substances were stored in at room temperature, as stated on the certificates of analysis, until use. BASF has retained reserve samples of these chemicals, and has documentation specifying the location of the synthesis and characterization information available at BASF Crop Protection, Research Triangle Park, North Carolina.

The BAS 656 H (Batch No. BEAU201204), M23 (Batch No. L82-104), M27 (Batch No. 01311-28) and M31 (Batch No. L81-132) reference substances were provided by the sponsor and received on June 7, 2017 in good condition. The certificates of analysis for all substances are presented in Appendix B. A summary of the reference substances is presented below.

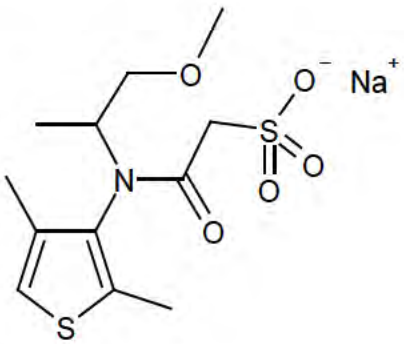
BAS Code	BAS 656 H	
Common Name	Dimethenamid-P	
Chemical Name	(S)-2-chloro-N-(2,4-dimethyl-3-thienyl)-N-(2-methoxy-1-methylethyl)acetamide	
BASF Reg. No.	363851	
CAS No.	163515-14-8	
Lot No.	BEAU201204	
Purity (Certificate)	96.4%	
Expiration	May 01, 2026	
Molecular Formula	C <sub>12</sub> H <sub>18</sub> ClNO <sub>2</sub> S	
Molecular Weight	275.8	

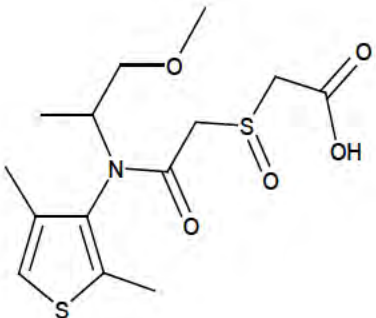
Common Name	M23	
Chemical Name	{{(2,4-dimethylthiophen-3-yl)[(2S)-1-methoxypropan-2-yl]amino}(oxo)acetic acid	
BASF Reg. No.	5886780 (S-enantiomer of Reg No. 360715)	
CAS No.	None	
Lot No.	L82-104	
Purity (Certificate)	99.9% <sup>(1)</sup>	
Expiration	July 01, 2017 <sup>(2)</sup>	
Molecular Formula	C <sub>12</sub> H <sub>17</sub> NO <sub>4</sub> S	
Molecular Weight	271.3	

(1) Updated COA: 95.8%

(2) Updated COA: June 01, 2019

Note: 99.9% purity used for all standard calculations

Common Name	M27	
Chemical Name	Sodium [(2,4-dimethyl-thiophen-3-yl)-(2-methoxy-1-methyl-ethyl)-carbamoyl]-methanesulfonate	
BASF Reg. No.	360714	
CAS No.	None	
Lot No.	01311-28	
Purity (Certificate)	97.1%	
Expiry	July 01, 2022	
Molecular Formula	C <sub>12</sub> H <sub>18</sub> NNaO <sub>5</sub> S <sub>2</sub>	
Molecular Weight	343.4	

Common Name	M31	
Chemical Name	[[[(2,4-Dimethyl-thiophen-3-yl)-(2-methoxy-1-methyl-ethyl)-carbamoyl]-methanesulfinyl]-acetic acid	
BASF Reg. No.	360712	
CAS No.	None	
Lot No.	L81-132	
Purity (Certificate)	100%	
Expiry	July 01, 2019	
Molecular Formula	C <sub>14</sub> H <sub>21</sub> NO <sub>5</sub> S <sub>2</sub>	
Molecular Weight	347.5	

### 2.3 Test System

Control soil and sediment were provided and homogenized by BASF. The soil and sediment were sent from BASF Crop Protection, Inc. on June 7, 2017 and received by Critical Path Services, LLC on June 8, 2017 in good condition.

The control samples were logged in to CPS records and given a unique sample number GS-17-92-1 (soil, BASF ID 22R04) and GS-17-92-2 (sediment, BASF ID CM17-025).

### 3. ANALYTICAL METHOD

BASF Analytical Method L0109/02 "Determination of Dimethenamid-P and its Metabolites Reg. No. 360715 (M23), Reg. No. 360714 (M27) and Reg. No. 360712 (M31) in Soil and Sediment by HPLC-MS/MS" was used for the analysis of the samples.

The residues of Dimethenamid-P (BAS 656 H) and its metabolites M23, M27 and M31 are extracted from 5 grams of soil or sediment by adding 20 mL of 60:40 methanol:water (v/v) and by shaking for 30 minutes on a mechanical shaker. After centrifugation, the supernatant is decanted into a 50-mL plastic centrifuge tube. Twenty mL of 60:40 methanol:water (v/v) are added to the soil or sediment matrix and the sample is shaken for another 30 minutes. After centrifugation, the supernatants are combined into the same 50-mL plastic centrifuge tube and diluted to 50 mL with water. The sample is diluted further in a separate container with another 50 mL of water. The sample extracts are then analyzed by HPLC-MS/MS. Instrument parameters are described in Table 26.

The primary (quantitative) and secondary (confirmatory) transition ions monitored are presented below:

Analyte	Transition ( <i>m/z</i> )		Polarity	RT (min)
	Primary	Secondary		
BAS 656 H	276 → 244	276 → 168	Positive	7.9
M23	270 → 198	270 → 166	Negative	6.6
M27	320 → 121	230 → 80	Negative	6.1
M31	346 → 240	346 → 198	Negative	6.4

