

Data Evaluation Record of the Analytical Method for Residues of Broflanilide in Soil

Data Requirement: EPA Guideline: 850.6100

EPA PC Code: 283200

Test material: Broflanilide

DP barcode: 450103

This study was reviewed as part of a joint review with the Health Canada Pest Management Regulatory Agency (PMRA). This preface is a supplement to the attached PMRA review of the study for EFED.

Primary Reviewer: Charles Lee **Date:** June 20, 2018

Evaluation Officer, EAD, PMRA

Secondary Reviewer: Mike Brown **Date:** July 26, 2018

Evaluation Officer, EAD, PMRA

Final EPA Reviewer: Faruque Khan **Signature:**

Senior Fate Scientist, EFED, OCSPP **Date:** November 21, 2019

Executive Summary

This analytical method, No. D1603/01 is designed for the quantitative determination of residues of broflanilide (BAS 450 I) and its metabolites DM-8007, DC-DM-8007, DC-8007 and S(PFP-OH)-8007 in soil LC-MS/MS. The recovery data were acceptable (between 70-120%) and the validated LOQ for residues of broflanilide in soil is 0.001 mg/kg. The LOQ is several orders of magnitude less than the lowest toxicological level of concern in soil. No modifications were made by the independent laboratory. The soil method can be extended for use as a post-registration monitoring method for broflanilide and the transformation products in sediments. This study is classified as **acceptable**.

Table 1. Analytical Method Summary

| Analyte(s) by Pesticide | MRID | | Matrix ¹ | Method Date (dd/mm/yyyy) | Registrant | Analysis | Limit of Quantitation (LOQ) |
|--|--------------------------------|-----------------------------------|---------------------|--------------------------|------------|----------|-----------------------------|
| | Environmental Chemistry Method | Independent Laboratory Validation | | | | | |
| Broflanilide and its Degradates ² | 50211340 50211645 | 50211646 | Soil | 08/22/2018 | BASF | LC/MS/MS | 0.001 mg/kg |

¹ For more details on the soil collection, handling, and characterization can be found in the terrestrial field dissipation study (MRID 50211431)

² Chemical name, code and chemical structure for active and all major transformation products / metabolites provided in **Appendix A-1**.

Analytical Methodology (parent compound and transformation products)**Reports:****MRID 50211340 (ECM)**

Title: Validation of method D1603/01: Method for the determination of residues of BAS 450 I (Reg. No. 5672774) and its metabolites DM-8007 (Reg. No. 5856361), DC-DM-8007 (Reg. No. 5936906), DC-8007 (Reg. No. 5936907) and S(PFP-OH)-8007 (Reg. No. 5959598) in soil by LC-MS/MS (at LOQ of 1ppb).

MRID 50211645 (ECM)

Title: Evaluation of the limit of detection (LOD) for method D1603/01, method for the determination of residue of BAS 450 I (Reg. No. 5672774) and its metabolites DM-8007 (Reg. No.5856361), DC-DM-8007 (Reg. No. 5936906), DC-8007 (Reg. No. 5936907) and S(PFP-OH)-8007 (Reg. No. 5959598) in soil by LC-MS/MS (at LOQ of 1ppb) (Data #1).

MRID 50211646 (ILV)

Title: Independent laboratory validation of the following method entitled: BASF analytical method D1603/01: Method for the determination of residues of BAS 450 I (Reg. No. 5672774) and its metabolites DM-8007 (Reg. No. 5856361), DC-DM-8007 (Reg. No. 5936906), DC-8007 (Reg. No. 5936907) and S(PFP-OH)-8007 (Reg. No. 5959598) in soil by LC-MS/MS (at LOQ of 1ppb).

Table 2. Principle of the method

| Items | Details |
|------------------------|---|
| Details of sample used | Clay (18-24 inch soil depth) and sandy (0-2 inch soil depth) soil samples (USDA Textural Class) were used for the study. The samples were characterized with the percentages of sand, silt and clay, bulk density, cation/anion exchange capacity, moisture, organic carbon/matter, salinity and base saturation |
| Sample preparation | Weigh 5 g of soil into a 50 mL plastic centrifuge tube, add exactly 10 mL of methanol, vortex to mix, shake for 30 minutes at 300 rpm on a mechanical shaker and then centrifuge at 3000 for 5 minutes. Decant supernatant into a 50 mL plastic centrifuge tube. Add exactly 10 mL of methanol:water (70:30, v/v) to soil marc, vortex to mix, shake for 30 minutes at 300 rpm on a mechanical shaker and then centrifuge at 3000 for 5 minutes. Combine both supernatants into the same 50 mL plastic centrifuge tube. Mix well. An aliquot of the solution is diluted with methanol:water (50:50, v/v) – further dilution may be needed for high fortification and high residue samples to fit in the calibration curve. Filter the sample through a 0.45 µm PTFE syringe filter directly into a HPLC injection vial. |

| Items | Details | | | | | | | | | | | | | | | | | | | | | | | | |
|---|--|--------------------|--------------------------|--------------------|--------------------|--------------|------|---------|---------|------------|------|---------|---------|---------|------|---------|---------|---------|------|---------|---------|----------------|------|---------|--------------------------|
| Method for quantitative analysis of parent compound and transformation products | <p>External standard</p> <p>The test/reference standards were synthesized by Mitsui Chemical Agro, Inc. (MCAG, Tokyo, Japan). Japan Analytical Chemistry Consultants Co., Ltd., on behalf of MCAG, determined characterization and purity prior to use. MCI-8007, 99.67%; DC-DM-8007, 98.58%; DC-8007, 99.07%; DM-8007, 98.84%; S(PFP-OH)-8007, 99.06%</p> | | | | | | | | | | | | | | | | | | | | | | | | |
| Method for identification of parent compound/transformation products | <p>LC-MS/MS method</p> <p>Column: BEH C18 column (100 x 2.1 mm, 2.5 µm)</p> <p>Detector: Triple Quad Mass Spectrometer</p> <p>Mobile phase: gradient of water:methanol, each acidified with 0.1% formic acid, 70:30 to 5:95, v/v, over ~6 minutes, flow rate 600 µL/minute</p> <p>Ionization: Electrospray ionization with positive mode</p> <table border="1" data-bbox="646 1035 1425 1535"> <thead> <tr> <th>Analyte</th> <th>Retention time (min)</th> <th>Quantitation [m/z]</th> <th>Confirmation [m/z]</th> </tr> </thead> <tbody> <tr> <td>Broflanilide</td> <td>5.64</td> <td>663→643</td> <td>665→645</td> </tr> <tr> <td>DC-DM-8007</td> <td>5.15</td> <td>545→525</td> <td>547→527</td> </tr> <tr> <td>DC-8007</td> <td>5.46</td> <td>559→539</td> <td>561→541</td> </tr> <tr> <td>DM-8007</td> <td>5.65</td> <td>649→242</td> <td>651→242</td> </tr> <tr> <td>S(PFP-OH)-8007</td> <td>5.30</td> <td>661→641</td> <td>661→621 663→643 (ILV)</td> </tr> </tbody> </table> | Analyte | Retention time (min) | Quantitation [m/z] | Confirmation [m/z] | Broflanilide | 5.64 | 663→643 | 665→645 | DC-DM-8007 | 5.15 | 545→525 | 547→527 | DC-8007 | 5.46 | 559→539 | 561→541 | DM-8007 | 5.65 | 649→242 | 651→242 | S(PFP-OH)-8007 | 5.30 | 661→641 | 661→621 663→643 (ILV) |
| Analyte | Retention time (min) | Quantitation [m/z] | Confirmation [m/z] | | | | | | | | | | | | | | | | | | | | | | |
| Broflanilide | 5.64 | 663→643 | 665→645 | | | | | | | | | | | | | | | | | | | | | | |
| DC-DM-8007 | 5.15 | 545→525 | 547→527 | | | | | | | | | | | | | | | | | | | | | | |
| DC-8007 | 5.46 | 559→539 | 561→541 | | | | | | | | | | | | | | | | | | | | | | |
| DM-8007 | 5.65 | 649→242 | 651→242 | | | | | | | | | | | | | | | | | | | | | | |
| S(PFP-OH)-8007 | 5.30 | 661→641 | 661→621 663→643 (ILV) | | | | | | | | | | | | | | | | | | | | | | |
| Chromatograms of spiked sample, control sample, blank and standard solution | Acceptable chromatograms of the solvent blank, reference standards and fortified sample were provided. | | | | | | | | | | | | | | | | | | | | | | | | |
| Quantitation | Linear regression (1/x weighting). Direct comparison of the sample peak responses to those of external standards. | | | | | | | | | | | | | | | | | | | | | | | | |

| Items | Details |
|---|--|
| Criteria for setting LOD and LOQ | The limit of detection (LOD) is the lowest level of fortification tested of an analyte in the matrix with an acceptable signal to noise ratio (S/N > 3:1). The limit of quantitation (LOQ) is the lowest fortification level that can be reliably quantitated. |
| Stability of parent and transformation products at various stages of analysis | Not stated |
| Special problems encountered and/or precautions to be taken during analysis/handling/storage of samples | Not stated |
| Total time for completion | Not stated |

The method validation data for the parent compound and the metabolites are summarized in **Table 3a** and **Table 3b**.

Table 3a. Method validation: Parent compound and converted products

| Parameter | Parent compound | DC-DM-8007 | DC-8007 | DM-8007 | S(PFP-OH)-8007 |
|--|-----------------|-----------------|-----------------|-----------------|-----------------|
| % Recovery at 0.001 ppm | 103-121 | 86-111 | 99-110 | 95-108 | 101-121 |
| % Recovery at 0.01 ppm | 97-119 | 92-110 | 100-111 | 97-109 | 98-112 |
| Mean % recovery | 106 | 100 | 106 | 103 | 107 |
| RSD % | 5 | 4 | 4 | 5 | 6 |
| Method linearity (ng/mL) | 0.01-0.2 | 0.01-0.2 | 0.01-0.2 | 0.01-0.2 | 0.01-0.2 |
| Correlation coefficient (r) ¹ | 0.9988 / 0.9972 | 0.9980 / 0.9975 | 0.9993 / 0.9995 | 0.9991 / 0.9987 | 0.9958 / 0.9969 |
| LOD (ppb) | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 |
| LOQ (ppb) | 1 | 1 | 1 | 1 | 1 |

¹ Obtained from the quantitation ion / from the confirmation ion

Table 3b. Independent Laboratory validation: Parent compound and converted products

| Parameter | Parent compound | DC-DM-8007 | DC-8007 | DM-8007 | S(PFP-OH)-8007 |
|--|-----------------|-----------------|-----------------|-----------------|-----------------|
| % Recovery at 0.001 ppm | 81.6-104 | 90.0-104 | 88.6-95.8 | 81.6-105 | 82.2-110 |
| % Recovery at 0.01 ppm | 84.4-97.0 | 81.6-94.6 | 81.0-99.0 | 81.4-95.0 | 87.4-103 |
| Mean % recovery | 91.0 | 91.9 | 90.8 | 89.3 | 96.5 |
| RSD % | 5.08 | 4.36 | 4.32 | 6.53 | 7.14 |
| Method linearity (ng/mL) | 0.01-0.2 | 0.01-0.2 | 0.01-0.2 | 0.01-0.2 | 0.01-0.2 |
| Correlation coefficient (r) ¹ | 0.9935 / 0.9969 | 0.9980 / 0.9975 | 0.9979 / 0.9979 | 0.9970 / 0.9972 | 0.9962 / 0.9978 |
| LOD (ppb) | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 |
| LOQ (ppb) | 1 | 1 | 1 | 1 | 1 |

¹ Obtained from the quantitation ion / from the confirmation ion

Comments/Deficiency

The submitted studies have provided analytical methods for the determination of broflanilide and metabolites in soil. No deficiencies have been identified but please note that any transformation products/metabolites present at levels greater than 10% of the initial concentration of the pesticide at any time during the study, as well as those products that have not attained 10% (e.g., 8-9%) but show a continuous increase in concentration up until the termination of the study, are considered to be major. (Also, transformation products/metabolites that are of toxicological concern are considered to be major, even if their maximum concentrations are less than 10% of the initial parent concentration).

Conclusion

An LC-MS/MS method was developed for the determination of broflanilide and its metabolites in soil and was validated in two soils. The recovery data were acceptable (between 70-120%), and the LOQ was determined to be 1 µg/Kg. This method is **acceptable** for use as a post-registration monitoring method.

Appendix A. Chemical structures:

Table A-1. Chemical name, code and chemical structure for active and all major transformation products / metabolites

| Chemical name | Code | Chemical structure |
|--|--|--------------------|
| <i>N</i> -[2-bromo-4-(perfluoropropan-2-yl)-6-(trifluoromethyl)phenyl]-2-fluoro-3-(<i>N</i> -methylbenzamido)benzamide | MLP-8607, BAS 450 I, MCI-8007, Reg. No. 5672774 [broflanilide] | |
| 3-benzamido- <i>N</i> -[2-bromo-4-(perfluoropropan-2-yl)-6-(trifluoromethyl)phenyl]-2-fluorobenzamide | DM-8007, MLP-8473, Reg. No. 5856361 | |
| 3-amino- <i>N</i> -[2-bromo-4-(perfluoropropan-2-yl)-6-(trifluoromethyl)phenyl]-2-fluorobenzamide | DC-DM-8007, MLQ-2111, Reg. No. 5936906 | |
| <i>N</i> -[2-bromo-4-(perfluoropropan-2-yl)-6-(trifluoromethyl)phenyl]-2-fluoro-3-(methylamino)benzamide | DC-8007 | |
| <i>N</i> -[2-bromo-4-(1,1,1,3,3,3-hexafluoro-2-hydroxypropan-2-yl)-6-(trifluoromethyl)phenyl]-2-fluoro-3-(<i>N</i> -methylbenzamido)benzamide | S(PFP-OH)+8007, MLQ-2172, Reg. No. 5959598 | |