

Environmental Chemistry Method (ECM) and Independent Laboratory Validation (ILV) for Determination of Bentazon Residues in Water

Reports: ECM: Validation of analytical method L0141/01 for the LC-MS/MS determination of BAS 351 H (Bentazon) in surface water and groundwater

ILV: Independent Laboratory Validation of Analytical Method Number L0141/01: LC-MS/MS determination of BAS 351 H (Bentazon) in surface water and groundwater

Document No.: [MRIDs 48970702 & 49141201]

Guideline: 850.6100 [U.S.], 8.2.2.3 [Water];

Statements: This study was conducted in accordance with the OECD Principles of Good Laboratory Practice and the GLP Principles of the German "Chemikaliengesetz" (Chemicals Act) and meets the United States Environmental Protection Agency Good Laboratory Practice Standards [40 CFR Part 160 (FIFRA) and Part 792 (TSCA)], with the exception that recognized differences exist between the GLP Principles/Standards of OECD and the Principles/Standards of FIFRA and TSCA.

Classification: The ECM and ILV methods are classified as **Supplemental** for monitoring bentazon parent compound in water. Both methods are upgradable upon provision of the correction of the following deficiencies.

Deficiencies:

- 1). The limit of detection (LOD) was not reported in the ECM.
- 2). The unit for Water should be in liter (L), not in Kg in ECM, e.g. using $\mu\text{g/L}$ instead of $\mu\text{g/Kg}$.
- 3). The mass spectrum images were not included in both ECM and ILV.
- 4). ILV reported the mg/Kg (page 13-14) as ppb, it should be ppm. The clarification is needed for the LOD (or LOQ) unit: mg/Kg or $\mu\text{g/Kg}$.
- 5). ILV reported using the homogenized matrix (water) without explanation of the content of homogenized water matrix. A water source clarification (e.g. surface water or ground water) is needed.

PC Code: 103901

Reviewer: He Zhong, Ph.D.
Biologist

Signature: 
Date: 11/13/2013

Executive Summary

This analytical method (ECM MRID 48970702) is for the quantitative determination of parent bentazon in water using LC-MS/MS. The limit of quantification (LOQ) is 0.03 µg/L (Table 1). The method limit of detection (LOD) is not reported in the ECM. The ILV (MRID 49141201) has reported an estimated LOD as 0.006 µg/L and confirmed ECM's LOQ limits as 0.03 µg/L. The ILV did not specify the ECM study deficiency. The LOQ (0.03 ppb) is below the level of concerns (LOCs) for Molluscs, Pacific oyster (*Crassostrea gigas*), (NOAEC = 7.04 ppb, ECOTOX Ref# 111918) and aquatic algae, Marine diatom (*Chaetoceros gracilis*), (EC50 = 60 ppb, ECOTOX Ref# 114118).

Table 1. Analytical Method Summary

Analyte(s)	MRID		Matrix	Method Date (m/d/y)	Registrant	Analysis	Limit of Detection ¹ (µg/L)	Limit of Quantitation (µg/L)
	ECM	ILV						
Bentazon	48970702	49141201	Water	11/16/2012	BASF	LC-MS/MS	0.006	0.03

¹The limit of detection (LOD) is only reported in Independent Laboratory Validation (ILV MRID 49141201) as 20% of limit of quantitation (LOQ) but not reported in Environmental Chemistry Method (ECM MRID 48970702)

I. Principle of the Method

A 10 g aliquot of the water sample is adjusted to pH 2 and extracted by ENV solid phase extraction (SPE) cartridge using methanol. The methanol extracts are evaporated to dryness and the residues are re-dissolved in water-methanol (50:50, v:v). An aliquot of the final volume is measured for LC-MS/MS. The final determination of BAS 351 H (Bentazon) was performed by LC-MS/MS in negative ion mode. One MRM parent-daughter ion (m/z 239.0 → 132.0 and m/z 239.0 → 197.0 (for confirmatory purposes) was monitored for quantitation.

II. Recovery Findings

The mean recoveries and the relative standard deviations (RSD) of bentazon were within guideline requirements (mean 70-120%; RSD ≤20%) for ECM (**Table 2**) and ILV (**Table 3**).

Table 2. Validation of Analytical Method (ECM) Recoveries for Bentazon in Surface and Ground Water

Analyte	Matrix	Fortification Level (µg/Kg)	Number of Tests	Recovery Range (%)	Mean Recovery (%)	% RSD
Bentazon Quantitation m/z 239/132	Surface Water	0.03	5	70.7-82	77.5	6.6
		0.3	5	83.3-86.7	85.7	1.7
	Ground Water	0.03	5	89.3-104.0	94.0	7.3
		0.3	5	80.0-84.0	82.1	1.9
Bentazon Confirmation m/z 239/197	Surface Water	0.03	5	70.0-80.7	76.1	6.6
		0.3	5	82.7-87.3	85.3	2.0
	Ground Water	0.03	5	90.7-104.7	94.8	7.0
		0.3	5	81.3-85.3	82.8	1.8

Table 3. Independent Lab Validation (ILV) Method Recoveries for Bentazon in Homogenized Water

Analyte	Matrix	Fortification Level (µg/L)	Number of Tests	Recovery Range (%)	Mean Recovery (%)	CV (%)
Bentazon Quantitation m/z 239/132	Homogenized Water	0.03	5	74-102	90	12.2
		0.3	5	77-107	94	13.4
Bentazon Confirmation m/z 239/197	Homogenized Water	0.03	5	72-95	84	12.1
		0.3	5	70-97	88	12.6

III. Method Characteristics

The ECM method characteristics and ILV confirmation are listed in Tables 4 and 5.

Table 4. Bentazon ECM Method Characteristics and ILV Method Confirmation

	Bentazon (ECM)	Bentazon (ILV)
Limit of Quantitation (LOQ)	0.03 µg/L	0.03 ² µg/L
Limit of Detection (LOD)	N/A	0.006 µg/L
Linearity (¹ calibration curve r ² and concentration range)	r ² = 0.999 0.022 – 1.0 µg/L	r ² = 0.992 0.025 – 0.5 µg/L
Repeatable	Yes	Yes
Reproducible	Yes	Yes
Specific	Yes	Yes

¹calibration curve is based on linear regression ($y=a+bx$) and r-values are reported in ECM/ILV method and r²-values are calculated based on the r-values.

²Reported value is 0.03 mg/Kg, which may be a type error for the unit, it should be µg/Kg based on the review on other lines of evidences.

Linearity is established in the calibration ($y=a+bx$) using external standards. The correlation coefficient of the calibration curves was above 0.999. The **limit of quantification** (LOQ) is 0.03 µg/L. The method in general satisfies the **repeatability** criteria with mean recoveries are in the range of 70-120% and RSDs are ≤20%. **Reproducibility** is satisfactory with the independent validation confirmed the LOQ(s) established by the initial validation. This method using LCMS/MS demonstrated excellent **specificity** by selecting the following daughter and parent ions (Table 5). However, both ECM and ILV method did not include the Mass Spectrum Graph to confirm the method specificity for the parent and daughter ions.

Table 5. Method Specificity—LC-MS/MS Parent and Daughter ions

Analyte	Parent ion	Daughter ion
Bentazon	132	197

IV. Method Deficiencies and Reviewer's Comments

ECM:

The limit of detection (LOD) is not reported. The water unit should be reported as liter (L) not as kilogram (Kg), e.g.: using $\mu\text{g/L}$ instead of $\mu\text{g/Kg}$. The calibration levels for the calibration curve were not reported. The images for mass spectrum were not included for the visual view of the primary and secondary ions.

ILV:

The ILV failed to disclose the content of the homogenized water matrix samples. A clarification of the water sample source is needed. The mg/Kg (page 13-14) is equivalent to ppm not ppb. It may be a typo since the $\mu\text{g/Kg}$ (ppb) is reported in the ECM. The registrant should provide EPA with a clarification of the correct "unit". The images for mass spectrum were not included for the visual view of the primary and secondary ions.

V. References

Grant, J. 2012. Independent Laboratory Validation of Analytical Method Number L0141/01: LC-MS/MS determination of BAS 351 H (Bentazon) in surface water and groundwater MRID 49141201

Penning, H. 2009. Validation of analytical method L0141/01 for the LC-MS/MS determination of BAS 351 H (Bentazon) in surface water and groundwater MRID 48970702