

Environmental Chemistry Method (ECM) and Independent Laboratory Validation (ILV) for Determination of Terbufos, Terbufos Sulfoxide and Terbufos Sulfone Residues in Soil and Sediment

Reports: ECM: Terbufos, Terbufos Sulfoxide and Terbufos Sulfone: Validation of Analytical Methodology for the Determination of Residues in Soil and Sediment

ILV: Independent Laboratory Validation of Methodology for the Determination of Residues of Terbufos, Terbufos Sulfoxide and Terbufos Sulfone in Soil (Sandy Loam and Clay Loam) and Sediment (Sandy Silt Loam)

Document No.: [MRIDs 49057601 & 48978101]

Guideline: 850.6100 [U.S.], 8.2.2.3 [Soil and Sediment];


Statements: The study was conducted in compliance with the following Good Laboratory Practice (GLP) Standards: UK (1999 No. 3106 and amended in 2004 No. 994), OECD (as revised in 1997) and EC Commission Directive (2004/10/EC, Official Journal No. L 50/44). These principles of GLP are accepted by the regulatory authorities of the USA and Japan on the basis of intergovernmental agreements. No claim of confidentiality is made for any information contained in this study on the basis of its falling within the scope of FIFRA § 10(d)(I)(A), (B), or (C).

Classification: The ECM is classified as **Supplemental** for monitoring Terbufos, Terbufos Sulfoxide and Terbufos Sulfone in Soil and Sediment. The ILV is classified as **Supplemental** and the method is upgradable if the omitted information is provided.

Deficiencies: 1). ECM: The ion transition is m/z 289>199 and the transition m/z 289>103 does not exist in mass spectrum provided. 2). ILV: fails to provides the mass spectrum confirmation image and fails to confirm the ion transition m/z for ECM. 3). ILV reports the wrong fortification level in $\mu\text{g/L}$ in the summary table and the correct one should be same as the ECM (mg/Kg).

PC Code: 105001

Reviewer: He Zhong, Ph.D.
Biologist

Signature: 
Date: 9/5/2013

Executive Summary

This analytical method, MRIDs **49057601 & 48978101**, is for the quantitative determination of parent Terbufos, and two degradates, Terbufos Sulfoxide and Terbufos Sulfone in soil and sediment using LC-MS/MS (see Table 1). The method is quantitative for parent Terbufos, Terbufos Sulfoxide and Terbufos Sulfone at the limit of detection (LOD) of 0.002 mg/Kg and limit of quantification (LOQ) of 0.01mg/Kg. The Agency does not have terrestrial plant data or earthworm data to assess the lowest toxicological level of concern. As a reference, terbufos toxicity for rat (NOAEL = 0.07 mg ai/Kg bw/day, MRID 43649402), and for avian (NOAEL = 1 ppm, MRID 00085177). The extraction time was clarified by ILV method that the procedure needs to be conducted within a maximum period of 2.5 hours. The ILV has confirmed the LOD and LOQ limits. No study deficiency was identified by the independent laboratory.

Table 1. Analytical Method Summary

Analyte(s) by Pesticide	MRID		Matrix	Method Date (m/d/y)	Registrant	Analysis	Limit of Detection (mg/Kg)	Limit of Quantitation (mg/Kg)
	Environmental Chemistry Method	Independent Laboratory Validation						
Terbufos	49057601	48978101	Soil and Sediment	28/5/2012	AMVAC	LC- MS/MS	0.002	0.01
Terbufos Sulfoxide	49057601	48978101	Soil and Sediment	28/5/2012	AMVAC	LC- MS/MS	0.002	0.01
Terbufos Sulfone	49057601	48978101	Soil and Sediment	28/5/2012	AMVAC	LC- MS/MS	0.002	0.01

I. Principle of the Method

Soil and sediment samples (sandy clay soil, sandy soil and sediment) fortified with known amount of terbufos, terbufos sulfoxide and terbufos sulfone (0.01 and 0.1 mg/Kg) were extracted with methanol:water (90:10 v:v), and cleaned up using solid phase extraction (SPE) cartridges, prior to reconstitution in acetonitrile:water (60:40 v:v). Quantitation was performed using liquid chromatography with tandem mass spectrometric detection (LC-MS/MS). The ion transition monitored were m/z 289>233, m/z 305>187 and m/z 321>171 for terbufos, terbufos sulfoxide and terbufos sulfone respectively, which were monitored for quantitation purpose. For residue confirmation, alternative ion transitions, m/z 289>199, m/z 305>243 and m/z 321>265 were also monitored respectively.

II. Recovery Findings

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

Table 2. Initial Validation Method Recoveries for Terbufos, Terbufos Sulfoxide and Terbufos Sulfone in Soil and Sediment

Analyte	Matrix	Fortification Level (mg/L)	Number of Tests	Recovery Range (%)	Mean Recovery (%)	CV (%)
Terbufos Quantitation	Sandy Clay Soil	0.01	5	76-87	80	5.2
		0.1	5	78-90	82	6.5
	Sandy Soil	0.01	5	70-84	75	7.9
		0.1	5	73-83	77	4.7
	Sediment	0.01	5	70-78	73	4.0
		0.1	5	76-83	79	3.5
Terbufos Confirmation	Sandy Clay Soil	0.01	5	74-84	80	5.2
		0.1	5	70-84	77	6.9
	Sandy Soil	0.01	5	79-90	85	5.5
		0.1	5	71-79	74	4.8
	Sediment	0.01	5	72-79	75	3.7
		0.1	5	78-84	81	3.7
Terbufos Sulfoxide Quantitation	Sandy Clay Soil	0.01	5	78-85	83	3.4
		0.1	5	88-95	91	3.4
	Sandy Soil	0.01	5	87-91	89	1.7
		0.1	5	94-96	95	0.9
	Sediment	0.01	5	83-90	86	3.1
		0.1	5	89-95	93	2.5
Terbufos Sulfoxide Confirmation	Sandy Clay Soil	0.01	5	85-94	90	3.8
		0.1	5	86-96	90	4.4
	Sandy Soil	0.01	5	90-98	95	3.4
		0.1	5	92-95	94	1.2
	Sediment	0.01	5	77-87	81	4.8
		0.1	5	91-98	94	2.6
Terbufos Sulfone Quantitation	Sandy Clay Soil	0.01	5	78-86	83	3.6
		0.1	5	87-93	89	2.8
	Sandy Soil	0.01	5	84-89	86	2.2
		0.1	5	89-95	92	2.4
	Sediment	0.01	5	83-90	86	3.5
		0.1	5	90-95	93	2.2
Terbufos Sulfone Confirmation	Sandy Clay Soil	0.01	5	78-85	82	3.6
		0.1	5	84-92	88	3.3
	Sandy Soil	0.01	5	82-92	87	4.6
		0.1	5	90-94	91	1.8
	Sediment	0.01	5	81-92	87	4.7
		0.1	5	86-94	90	3.3

Table 3. Independent Lab Validation Method Recoveries for Terbufos, Terbufos Sulfoxide and Terbufos Sulfone in Soil and Sediment

Analyte	Matrix	Fortification Level (mg/L)	Number of Tests	Recovery Range (%)	Mean Recovery (%)	CV (%)
Terbufos Quantitation	Sandy Clay Soil	0.01	5	74.1-92.0	83.5	7.9
		0.1	5	77.2-87.5	81.4	4.6
	Sandy Soil	0.01	5	72.3-88.6	80.0	7.2
		0.1	5	77.8-83.4	81.5	2.7
	Sediment	0.01	5	68.1-94.4	79.7	12.1
		0.1	5	73.6-81.1	78.8	4.0
Terbufos Confirmation	Sandy Clay Soil	0.01	5	69.7-84.4	79.0	7.2
		0.1	5	70.2-87.5	77.1	8.5
	Sandy Soil	0.01	5	69.3-87.7	76.4	9.1
		0.1	5	73.0-81.0	78.0	5.2
	Sediment	0.01	5	67.5-90.5	78.3	12.3
		0.1	5	69.0-83.2	76.6	9.1
Terbufos Sulfoxide Quantitation	Sandy Clay Soil	0.01	5	90.5-98.2	93.5	3
		0.1	5	82.9-91.0	85.5	3.9
	Sandy Soil	0.01	5	90.1-94.3	91.4	1.8
		0.1	5	82.6-86.1	84.6	1.7
	Sediment	0.01	5	87.3-99.1	94.2	4.7
		0.1	5	77.2-87.3	84.6	5.0
Terbufos Sulfoxide Confirmation	Sandy Clay Soil	0.01	5	88.6-96.4	91.5	3.6
		0.1	5	84.0-95.7	89.1	5.1
	Sandy Soil	0.01	5	84.6-91.6	88.0	3.4
		0.1	5	84.4-89.3	87.2	2.4
	Sediment	0.01	5	85.0-93.9	88.9	4.2
		0.1	5	77.4-85.4	83.0	3.9
Terbufos Sulfone Quantitation	Sandy Clay Soil	0.01	5	88.2-102.2	94.4	6.4
		0.1	5	89.6-97.7	91.6	3.7
	Sandy Soil	0.01	5	77.6-83.4	81.2	2.7
		0.1	5	87.3-91.6	89.0	1.8
	Sediment	0.01	5	86.3-98.8	93.4	6.5
		0.1	5	87.4-93.9	90.7	3.1
Terbufos Sulfone Confirmation	Sandy Clay Soil	0.01	5	90.4-101.1	94.9	4.4
		0.1	5	89.4-100.8	93.9	4.7
	Sandy Soil	0.01	5	81.3-90.0	84.5	4.8
		0.1	5	85.9-89.6	87.8	2.0
	Sediment	0.01	5	85.8-94.2	89.8	44
		0.1	5	86.3-88.9	88.2	1.2

III. Method Characteristics

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

Table 4. ECM Method Characteristics

	Terbufos	Terbufos Sulfoxide	Terbufos Sulfone
Limit of Quantitation (LOQ)	0.01 mg/Kg	0.01 mg/Kg	0.01 mg/Kg
Limit of Detection (LOD)	0.002 mg/Kg	0.002 mg/Kg	0.002 mg/Kg
Linearity (¹ calibration curve r^2 and concentration range)	$r^2 = 0.999$ 0.1 – 10 µg/L	$r^2 = 0.999$ 0.1 – 10 µg/L	$r^2 = 0.999$ 0.1 – 10 µg/L
Repeatable	Yes	Yes	Yes
Reproducible	Yes	Yes	Yes
Specific	Yes	Yes	Yes

¹calibration curve is based on linear regression ($y=a+bx$) and r -values are reported in ECM method and r^2 -values are calculated based on the r -values.

Table 5. ILV Method Characteristics Confirmation

	Terbufos	Terbufos Sulfoxide	Terbufos Sulfone
Limit of Quantitation (LOQ)	0.01 mg/Kg	0.01 mg/Kg	0.01 mg/Kg
Limit of Detection (LOD)	0.002 mg/Kg	0.002 mg/Kg	0.002 mg/Kg
Linearity (¹ calibration curve r^2 and concentration range)	$r^2 = 0.997$ 0.1 – 10 µg/L	$r^2 = 0.992$ 0.1 – 10 µg/L	$r^2 = 0.995$ 0.1 – 10 µg/L
Repeatable	Yes	Yes	Yes
Reproducible	Yes	Yes	Yes
Specific	Yes	Yes	Yes

¹calibration curve is based on linear regression ($y=a+bx$) and r -values are reported in ILV method and r^2 -values are calculated based on the r -values.

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 0.01 mg/L. The method in general satisfies the **repeatability** criteria with mean recoveries are in the range of 70-120% and RSDs are $\leq 20\%$. **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 6). However, ILV method did not include the Mass Spectrum Graph to confirm the method specificity for the parent and daughter ions

Table 6. Method Specificity—LC-MS/MS Parent and Daughter ions

Analyte	Parent ion	Daughter ion
Terbufos	233	289 or 199
Terbufos Sulfoxide	187	305 or 243
Terbufos Sulfone	171	321 or 265

IV. Method Deficiencies and Reviewer's Comments

- 1). The ECM did not specify the parent ions for terbufos (Fig 7), terbufos sulfoxide (Fig 8) and terbufos sulfone (Fig 9).
- 2). The ion transition monitored cannot be verified at 289>103 from the mass spectrum image provided by the ECM.
- 3). ILV fails to report the correct fortification level (0.1 and 1 µg/L) in summary table in Page 14 and accuracy and precision table in Page 25. The correct values are 0.01 and 0.1 mg/Kg
- 4). ILV fails to include the mass spectrum confirmation image in the report and did not verify the ion transition for ECM.

V. References

- Brewin, S. 2012. Terbufos, Terbufos Sulfoxide and Terbufos Sulfone: Validation of Analytical Methodology for the Determination of Residues in Soil and Sediment MRID 49057601
- Pawula, M. 2012. Independent Laboratory Validation of Methodology for the Determination of Residues of Terbufos, Terbufos Sulfoxide and Terbufos Sulfone in Soil (Sandy Loam and Clay Loam) and Sediment (Sandy Silt Loam). MRID 48978101