Designation: D8025 – 16

Standard Test Method for Determination of Select Pesticides in Water by Multiple Reaction Monitoring Liquid Chromatography Tandem Mass Spectrometry[1]

This standard is issued under the fixed designation D8025; the number immediately following the designation indicates the year of original adoption or, in the case of revision, the year of last revision. A number in parentheses indicates the year of last reapproval. A superscript epsilon (ε) indicates an editorial change since the last revision or reapproval.

1. Scope

1.1 This procedure covers a method for analysis of selected pesticides in a water matrix by filtration followed with liquid chromatography/electrospray ionization tandem mass spectrometry analysis. The samples are prepared in 20 % methanol, filtered, and analyzed by liquid chromatography/tandem mass spectrometry. This method was developed for an agricultural run-off study, not for low level analysis of pesticides in drinking water. This method may be modified for lower level analysis. The analytes are qualitatively and quantitatively determined by this method. This method adheres to multiple reaction monitoring (MRM) mass spectrometry.

1.2 A full collaborative study to meet the requirements of Practice D2777 has not been completed. This standard contains single-operator precision and bias based on single-operator data. Publication of standards that have not been fully validated is done to make the current technology accessible to users of standards, and to solicit additional input from the user community.

1.3 A reporting limit check sample (RLCS) is analyzed during every batch to ensure that if an analyte was present in a sample at or near the reporting limit it would be positively identified and accurately quantitated within set quality control limits. A method detection limit (MDL) study was not done for this method, the method detection limits would be much lower than the reporting limits in this method and would be irrelevant. A RLCS was determined to be more applicable for this standard. If this method is adapted to report much lower or near the MDL then a MDL study would be warranted.

1.4 *Units*—The values stated in SI units are to be regarded as standard. No other units of measurement are included in this standard.

1.5 The Reporting Range for the target analytes are listed in Table 1.

1.5.1 The reporting limit in this test method is the minimum value below which data are documented as non-detects. The reporting limit is calculated from the concentration of the Level 1 calibration standard as shown in Table 6 after taking into account an 8 mL water sample volume and a final diluted sample volume of 10 mL (80 % water/20 % methanol).

1.6 *This standard does not purport to address all of the safety concerns, if any, associated with its use. It is the responsibility of the user of this standard to establish appropriate safety and health practices and determine the applicability of regulatory limitations prior to use.*

2. Referenced Documents

2.1 *ASTM Standards:*[2]

D1129 Terminology Relating to Water
D1193 Specification for Reagent Water
D2777 Practice for Determination of Precision and Bias of Applicable Test Methods of Committee D19 on Water
D3856 Guide for Management Systems in Laboratories Engaged in Analysis of Water
D4841 Practice for Estimation of Holding Time for Water Samples Containing Organic and Inorganic Constituents
D5847 Practice for Writing Quality Control Specifications for Standard Test Methods for Water Analysis
E2554 Practice for Estimating and Monitoring the Uncertainty of Test Results of a Test Method Using Control Chart Techniques

2.2 *Other Document:*

EPA publication SW-846 Test Methods for Evaluating Solid Waste, Physical/Chemical Methods[3]

3. Terminology

3.1 *Definitions:*

[1] This test method is under the jurisdiction of ASTM Committee D19 on Water and is the direct responsibility of Subcommittee D19.06 on Methods for Analysis for Organic Substances in Water.

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[2] For referenced ASTM standards, visit the ASTM website, www.astm.org, or contact ASTM Customer Service at service@astm.org. For *Annual Book of ASTM Standards* volume information, refer to the standard's Document Summary page on the ASTM website.

[3] Available from National Technical Information Service (NTIS), U.S. Department of Commerce, 5285 Port Royal Road, Springfield, VA, 22161 or at http://www.epa.gov/epawaste/hazard/testmethods/index.htm

TABLE 1 Reporting Range

Analyte	Reporting Ranges, (ng/L)
2,4-D	250–10 000
Acetochlor	250–10 000
Alachlor	250–10 000
Aldicarb	250–10 000
Atrazine	62.5–2 500
Desethylatrazine	62.5–2 500
Desisopropylatrazine	125–5 000
Azoxystrobin	31.2–1 250
Bentazon	250–10 000
Carbaryl	250–10 000
Chlorpyrifos	250–10 000
Clopyralid	25 000–1 000 000
Clothianidin	62.5–2 500
Diazinon	62.5–2 500
Dicamba	12 500–500 000
Fipronil	250–10 000
Imidacloprid	62.5–2 500
Malathion	125–5 000
Methomyl	250–10 000
Metolachlor	62.5–2 500
Metribuzin	125–5 000
Picloram	6 250–250 000
Propiconazole	62.5–2 500
Simazine	62.5–2 500
Tebuconazole	62.5–2 500
Thiamethoxam	62.5–2 500
Triclopyr	1 250–5 000

3.1.1 For definitions of terms used in this standard, refer to Terminology D1129.

3.2 *Definitions of Terms Specific to This Standard:*

3.2.1 *batch QC, n*—all the quality control samples and standards included in an analytical procedure.

3.2.2 *independent reference material, IRM, n*—a material of known purity and concentration obtained either from the National Institute of Standards and Technology (NIST) or other reputable supplier.

3.2.2.1 *Discussion*—The IRM shall be obtained from a different lot of material than is used for calibration.

3.2.3 *reporting limit, RL, n*—the minimum concentration below which data are documented as non-detects.

3.2.4 *reporting limit check sample, RLCS, n*—a sample used to ensure that if the analyte was present at the reporting limit, it would be confidently identified.

3.3 *Acronyms:*

3.3.1 *CCC, n*—Continuing Calibration Check

3.3.2 *CRW, n*—Chicago River Water

3.3.3 *IC, n*—Initial Calibration

3.3.4 *LC, n*—Liquid Chromatography

3.3.5 *LCS/LCSD, n*—Laboratory Control Sample/Laboratory Control Sample Duplicate

3.3.6 *MDL, n*—Method Detection Limit

3.3.7 *MeOH, n*—Methanol

3.3.8 *mM, n*—millimolar, 1×10^{-3} moles/L

3.3.9 *MRM, n*—Multiple Reaction Monitoring

3.3.10 *MS/MSD, n*—Matrix Spike/Matrix Spike Duplicate

3.3.11 *NA, adj*—Not Available

3.3.12 *ND, n*—non-detect

3.3.13 *P&A, n*—Precision and Accuracy

3.3.14 *ppt, n*—parts-per-trillion

3.3.15 *QA, adj*—Quality Assurance

3.3.16 *QC, adj*—Quality Control

3.3.17 *RL, n*—Reporting Limit

3.3.18 *RLCS, n*—Reporting Limit Check Sample

3.3.19 *RSD, n*—Relative Standard Deviation

3.3.20 *RT, n*—Retention Time

3.3.21 *SDS, n*—Safety Data Sheets

3.3.22 *SRM, n*—Single Reaction Monitoring

3.3.23 *SS, n*—Surrogate Standard

3.3.24 *TC, n*—Target Compound

3.3.25 *VOA, n*—Volatile Organic Analysis

4. Summary of Test Method

4.1 The operating conditions presented in this standard have been successfully used in the determination of the select pesticides in water; however, this standard is intended to be performance based and alternative operating conditions can be used to perform this method provided data quality objectives are attained.

4.2 For pesticide analysis, samples are shipped to the lab on ice and analyzed within 14 days of collection. A sample (8 mL) is transferred to an amber VOA vial, an isotopically labeled pesticide surrogate mix is added to all samples followed by a pesticide spike solution which is added only to the Reporting Limit Check Samples, Laboratory Control and Matrix Spike samples before the addition of methanol. Then 2 mL of methanol is added to each sample and hand shaken or vortexed for 1 minute. The samples are then filtered through a PTFE membrane syringe driven filter unit and then analyzed by LC/MS/MS. All concentrations reported only to the reporting limit.

TABLE 2 Gradient Conditions for Neutral Liquid Chromatography

Time (min)	Flow (mL/min)	Percent 95 % Water: 5 % Methanol	Percent Methanol	Percent 200 mM Ammonium Formate (95 % Water: 5 % Methanol)
0	0.3	95	0	5
1.5	0.3	95	0	5
9	0.3	0	95	5
12	0.3	0	95	5
13	0.3	95	0	5
16	0.3	95	0	5

4.3 The analysis of the sample requires two separate analysis methods, one using the LC gradient conditions in Table 2 with the Methanol/Water/Ammonium Formate and the second using the LC gradient conditions in Table 3 with the Methanol/Water/Formic acid. Each analysis set is to be analyzed separately in two different complete sample sequences which includes the exact same samples and may even include the same calibration level standards. The only analytes reported from the formic acid run conditions are 2,4-D, Clopyralid, Dicamba, Picloram, Triclopyr, 2,4-D (Ring-D3) and Dicamba-D3, the rest of the analytes are reported from the ammonium formate analysis run.

4.4 The pesticides are identified by comparing the single reaction monitoring (SRM) transition and its confirmatory SRM transitions if correlated to the known standard SRM transition (Tables 4 and 5) and quantitated utilizing an external calibration. The final report issued for each sample lists the concentration of pesticides, if detected, or RL, if not detected, in ng/L and surrogate recovery.

5. Significance and Use

5.1 Pesticides may be used in various agricultural and household products. These products may enter waterways at low levels through run-off or misuse near water resources. Hence, there is a need for quick, easy and robust method to determine pesticide concentration in water matrices for understanding the sources and concentration levels in affected areas.

5.2 This method has been single-laboratory validated in reagent water and surface waters (Tables 12-14).

6. Interferences

6.1 All glassware is washed in hot water with detergent and rinsed in hot water followed by distilled water. The glassware is then dried and heated in an oven at 250°C for 15 to 30 minutes. All glassware is subsequently rinsed or sonicated, or both, with acetone, n-propanol, acetonitrile, or a combination thereof.

6.2 All reagents and solvents should be pesticide residue purity or higher to minimize interference problems.

6.3 Matrix interferences may be caused by contaminants in the sample. The extent of matrix interferences can vary considerably depending on variations in the sample matrices.

7. Apparatus

7.1 *LC/MS/MS System:*

7.1.1 *Liquid Chromatography System*[4]—A complete LC system is required in order to analyze samples, this should include a sample injection system, an autosampler, a solvent pumping system capable of mixing solvents, a sample compartment capable of maintaining required temperature and a temperature controlled column compartment. A LC system that is capable of performing at the flows, pressures, controlled temperatures, sample volumes, and requirements of the standard shall be used.

7.1.2 *Analytical Column*[5]—A reverse phase C18 particle column was used to develop this test method. Any column that achieves adequate resolution may be used. The retention times and order of elution may change depending on the column used and need to be monitored.

7.2 *Tandem Mass Spectrometer System*[6]—A MS/MS system capable of multiple reaction monitoring (MRM) analysis or any system that is capable of meeting the requirements in this standard shall be used. Electrospray ionization is utilized for this standard.

7.3 *Adjustable Volume Pipettes*—10, 20, 100 and 1000 µL and 5 and 10 mL.

7.3.1 *Discussion*—Any pipette may be used providing the data generated meets the performance of the standard.

7.3.2 *Pipette Tips*—Polypropylene pipette tips free of release agents or low retention coating of various sizes.

7.4 *Class A Volumetric Glassware.*

7.5 *Filtration Device:*

7.5.1 *Hypodermic Syringe*—A luer-lock tip glass syringe capable of holding a syringe driven filter unit.

7.5.2 A 10 mL Lock Tip Glass Syringe size is recommended since a 10 mL prepared sample size is used in this test method. If a smaller volume syringe is used, do not wash out the syringe

[4] A Waters Acquity (a trademark of the Waters Corporation, Milford, MA) UPLC H-Class System, or equivalent, has been found suitable for use.

[5] A Waters Acquity (a trademark of the Waters Corporation, Milford, MA) UPLC BEH C18, 2.1×100 mm and 1.7 µm particle size column, or equivalent, has been found suitable for use. It was used to develop this test method and generate the precision and bias data presented in Section 16.

[6] A Waters Xevo (a trademark of the Waters Corporation, Milford, MA) TQ-S triple quadrupole mass spectrometer, or equivalent, has been found suitable for use.

TABLE 3 Gradient Conditions for Acidic Liquid Chromatography

Time (min)	Flow (mL/min)	Percent 95 % Water: 5 % Methanol	Percent Methanol	Percent 4 % Formic Acid (95 % Water: 5 % Methanol)
0	0.3	95	0	5
1.5	0.3	95	0	5
6	0.3	0	95	5
9	0.3	0	95	5
10	0.3	95	0	5
13	0.3	95	0	5

TABLE 4 SRM Ions and Analyte-Specific Mass Spectrometer Parameters

Chemical	Primary/ Confirmatory	MRM Transition	ESI Mode Neg/Pos	Cone (V)	Collision (eV)
2,4-D[A]	Primary	218.9→160.9	Neg	20	14
	Confirmatory	220.9→162.9			14
Acetochlor	Primary	270.1→224.1	Pos	20	10
	Confirmatory	270.1→148			18
Alachlor	Primary	270.1→162	Pos	30	12
	Confirmatory	270.1→238.1			18
Aldicarb	Primary	213→88.9	Pos	20	16
	Confirmatory	213→116			12
Atrazine	Primary	216.1→174	Pos	20	18
	Confirmatory	216.1→95.9			24
Desethylatrazine	Primary	188→146	Pos	30	16
	Confirmatory	188→78.8			22
Desisopropylatrazine	Primary	174→96	Pos	30	16
	Confirmatory	174→78.8			16
Azoxystrobin	Primary	404.2→372.2	Pos	20	14
	Confirmatory	404.2→344.2			24
Bentazon	Primary	239→197	Pos	20	20
	Confirmatory	239→175			20
Carbaryl	Primary	202.1→145	Pos	10	10
	Confirmatory	202.1→127			25
Chlorpyrifos	Primary	350→197.9	Pos	30	20
	Confirmatory	350→322			11
Clopyralid[A]	Primary	189.9→145.9	Neg	5	10
	Confirmatory	191.9→147.9			10
Clothianidin	Primary	250→169	Pos	20	15
	Confirmatory	250→131.9			11
Diazinon	Primary	305.1→169	Pos	30	20
	Confirmatory	305.1→153			20
Dicamba[A]	Primary	218.9→174.9	Neg	10	8
	Confirmatory	220.9→176.9			8
Fipronil	Primary	435→330	Pos	30	15
	Confirmatory	435→318			23
Imidacloprid	Primary	256.1→209.1	Pos	30	17
	Confirmatory	256.1→175			23
Malathion	Primary	331.1→127	Pos	20	12
	Confirmatory	331.1→285			6
Methomyl	Primary	163→87.9	Pos	10	10
	Confirmatory	163→105.9			11
Metolachlor	Primary	284.1→252.1	Pos	30	16
	Confirmatory	284.1→176.1			25
Metribuzin	Primary	215.1→187.1	Pos	30	16
Picloram[A]	Primary	240.9→196.9	Neg	10	11
	Confirmatory	238.9→194.9			11
Propiconazole	Primary	342.1→158.9	Pos	30	22
	Confirmatory	342.1→205			17
Simazine	Primary	202→132	Pos	40	17
	Confirmatory	202→124			17
Tebuconazole	Primary	308.2→70	Pos	40	20
	Confirmatory	308.2→125			28
Thiamethoxam	Primary	292.1→211.1	Pos	20	12
	Confirmatory	292.1→131.9			20
Triclopyr[A]	Primary	253.9→195.9	Neg	10	12
	Confirmatory	253.9→217.9	Neg	10	6
		Surrogates			
2,4-D (Ring-D3)[A]	Primary	221.9→163.8	Neg	20	14
Atrazine (ethyl-D5)	Primary	221.1→179	Pos	20	17
Desethylatrazine (iso-propyl-D7)	Primary	195→146.9	Pos	20	18
Desisopropylatrazine (ethyl-D5)	Primary	179→100.9	Pos	30	18
Bentazon -D7	Primary	246.1→182	Pos	20	20
Carbofuran (Ring-13C6)	Primary	228.1→171	Pos	20	12
Clothianidin -D3	Primary	253→131.9	Pos	20	15
Diazinon (Diethyl-D10)	Primary	315.2→170	Pos	30	20
Dicamba -D3[A]	Primary	223.9→179.9	Neg	10	8
Imidacloprid -D4	Primary	260.1→213.1	Pos	30	15
Methomyl (Acetohydroxamate-13C2 15N)	Primary	166→90.8	Pos	10	8
Simazine (Diethyl-D10)	Primary	212.1→134	Pos	40	18
Tebuconazole (tert-Butyl-D9)	Primary	317.2→69.9	Pos	40	20
Thiamethoxam -D3	Primary	295→214.1	Pos	30	12

[A] Indicates analyzed under acidic LC conditions.

TABLE 5 SRM Ions, Retention times and SRM Ion Ratios

Chemical	Primary/ Confirmatory	MRM Transition	Retention Time Minutes	Primary/Confirmatory SRM Area Ratio
2,4-D[A]	Primary	218.9→160.9	7.6	1.5
	Confirmatory	220.9→162.9		
Acetochlor	Primary	270.1→224.1	10.3	2.5
	Confirmatory	270.1→148		
Alachlor	Primary	270.1→162	10.3	0.4
	Confirmatory	270.1→238.1		
Aldicarb	Primary	213→88.9	8	2.1
	Confirmatory	213→116		
Atrazine	Primary	216.1→174	9.2	3.6
	Confirmatory	216.1→95.9		
Desethylatrazine	Primary	188→146	7.6	5.4
	Confirmatory	188→78.8		
Desisopropylatrazine	Primary	174→96	6.6	1.3
	Confirmatory	174→78.8		
Azoxystrobin	Primary	404.2→372.2	9.6	3.7
	Confirmatory	404.2→344.2		
Bentazon	Primary	239→197	6.4	1.1
	Confirmatory	239→175		
Carbaryl	Primary	202.1→145	8.8	3.6
	Confirmatory	202.1→127		
Chlorpyrifos	Primary	350→197.9	11.4	1.8
	Confirmatory	350→322		
Clopyralid[A]	Primary	189.9→145.9	5.2	1.5
	Confirmatory	191.9→147.9		
Clothianidin	Primary	250→169	6.9	1.6
	Confirmatory	250→131.9		
Diazinon	Primary	305.1→169	10.6	1.6
	Confirmatory	305.1→153		
Dicamba[A]	Primary	218.9→174.9	7.1	1.5
	Confirmatory	220.9→176.9		
Fipronil	Primary	435→330	10.3	5.7
	Confirmatory	435→318		
Imidacloprid	Primary	256.1→209.1	6.9	1.1
	Confirmatory	256.1→175		
Malathion	Primary	331.1→127	9.9	1.2
	Confirmatory	331.1→285		
Methomyl	Primary	163→87.9	6.1	1.8
	Confirmatory	163→105.9		
Metolachlor	Primary	284.1→252.1	10.3	2.4
	Confirmatory	284.1→176.1		
Metribuzin	Primary	215.1→187.1	8.5	NA
Picloram[A]	Primary	240.9→196.9	5.9	1
	Confirmatory	238.9→194.9		
Propiconazole	Primary	342.1→158.9	10.6	7.2
	Confirmatory	342.1→205		
Simazine	Primary	202→132	7.6	5.4
	Confirmatory	202→124		
Tebuconazole	Primary	308.2→70	10.5	11.2
	Confirmatory	308.2→125		
Thiamethoxam	Primary	292.1→211.1	6.3	4
	Confirmatory	292.1→131.9		
Triclopyr[A]	Primary	253.9→195.9	7.8	2.9
	Confirmatory	253.9→217.9		
Surrogates				
2,4-D (Ring-D3)[A]	Primary	221.9→163.8	7.6	NA
Atrazine (ethyl-D5)	Primary	221.1→179	9.2	NA
Desethylatrazine (iso-propyl-D7)	Primary	195→146.9	7.6	NA
Desisopropylatrazine (ethyl-D5)	Primary	179→100.9	6.6	NA
Bentazon -D7	Primary	246.1→182	6.4	NA
Carbofuran (Ring-13C6)	Primary	228.1→171	8.6	NA
Clothianidin -D3	Primary	253→131.9	6.9	NA
Diazinon (Diethyl-D10)	Primary	315.2→170	10.6	NA
Dicamba -D3[A]	Primary	223.9→179.9	7.1	NA
Imidacloprid -D4	Primary	260.1→213.1	6.9	NA
Methomyl (Acetohydroxamate-13C2, 15N)	Primary	166→90.8	6.1	NA
Simazine (Diethyl-D10)	Primary	212.1→134	7.6	NA
Tebuconazole (tert-Butyl-D9)	Primary	317.2→69.9	10.5	NA
Thiamethoxam -D3	Primary	295→214.1	6.3	NA

[A] Indicates analyzed under acidic LC conditions.

or change filters while filtering the same sample if multiple refills of the syringe are required in order to filter the 10 mL prepared sample.

7.5.3 *Filter Unit*[7]—PTFE filter units were used to filter the samples.

7.6 *Vials*—2-mL autosampler vials (LC vials) with pre-slit PTFE/silicone septa or equivalent.

7.7 *Sonicator.*

7.8 *Oven*—Capable to achieve 250°C.

7.9 *VOA Vials*—Amber, 40 mL.

8. Reagents and Materials

8.1 *Purity of Reagents*—High Performance Liquid Chromatography (HPLC) pesticide residue analysis and spectrophotometry grade chemicals shall be used in all tests. Unless indicated otherwise, it is intended that all reagents shall conform to the Committee on Analytical Reagents of the American Chemical Society.[8] Other reagent grades may be used provided they are first determined to be of sufficiently high purity to permit their use without affecting the accuracy of the measurements.

8.2 *Purity of Water*—Unless otherwise indicated, references to water shall be understood to mean reagent water conforming to Type 1 of Specification D1193. It shall be demonstrated that this water does not contain contaminants at concentrations sufficient to interfere with the analysis.

8.3 All prepared solutions are routinely replaced every year if not previously discarded for quality control failure.

8.4 *Gases*—Ultrapure nitrogen and argon.

8.5 Formic Acid (CAS # 64-18-6)

8.6 Acetonitrile (CAS # 75-05-8)

8.7 Methanol (CAS # 67-56-1)

8.8 Ammonium Formate (CAS # 540-69-2)

8.9 2-Propanol (isopropyl alcohol, CAS # 67-63-0)

8.10 2,4-Dichlorophenoxyacetic acid (2,4-D, CAS # 94-75-7)

8.11 Acetochlor (CAS # 34256-82-1)

8.12 Alachlor (CAS # 15972-60-8)

8.13 Aldicarb (CAS # 116-06-3)

8.14 Atrazine (CAS # 1912-24-9)

8.15 Desethylatrazine (CAS # 6190-65-4)

8.16 Desisopropylatrazine (CAS # 1007-28-9)

8.17 Azoxystrobin (CAS # 131860-33-8)

8.18 Bentazon (CAS # 25057-89-0)

8.19 Carbaryl (CAS # 63-25-2)

8.20 Chlorpyrifos (CAS # 2921-88-2)

8.21 Clopyralid (CAS # 1702-17-6)

8.22 Clothianidin (CAS # 210880-92-5)

8.23 Diazinon (CAS # 333-41-5)

8.24 Dicamba (CAS # 1918-00-9)

8.25 Fipronil (CAS # 120068-37-3)

8.26 Imidacloprid (CAS # 138261-41-3)

8.27 Malathion (CAS # 121-75-5)

8.28 Methomyl (CAS # 16752-77-5)

8.29 Metolachlor (CAS # 51218-45-2)

8.30 Metribuzin (CAS # 21087-64-9)

8.31 Picloram (CAS # 1918-02-1)

8.32 Propiconazole (CAS # 60207-90-1)

8.33 Simazine (CAS # 122-34-9)

8.34 Tebuconazole (CAS # 107534-96-3)

8.35 Thiamethoxam (CAS # 153719-23-4)

8.36 Triclopyr (CAS # 55335-06-3)

8.37 *Isotopically Labeled Pesticide Standards (Surrogates)*—There are not isotopically labeled surrogates for every target analyte. The labeled surrogate only mimics its unlabeled target analyte. The isotopically labeled carbofuran was chosen to mimic carbaryl. (Note—P&A data show that the labeled carbofuran is not a good surrogate for carbaryl even though they are structurally similar.) Surrogates may be added or deleted from the below list if new ones become available or if the existing ones are not readily available. The surrogate list is long and expensive to maintain. If surrogates are not available at the time of analysis it will be mentioned in the case narrative that accompanies the data, if extra surrogates are added this will also be mentioned in the case narrative. (CAS #'s are for the unlabeled native analyte).

8.37.1 2,4-Dichlorophenoxyacetic acid (2,4-D (Ring-D3), CAS # 94-75-7)

8.37.2 Atrazine (ethyl-D5, CAS # 1912-24-9)

8.37.3 Desethylatrazine (iso-propyl-D7, CAS # 6190-65-4)

8.37.4 Desisopropylatrazine (ethyl-D5, CAS # 1007-28-9)

8.37.5 Bentazon (D7, CAS # 25057-89-0)

8.37.6 Carbofuran (Ring-13C6, CAS # 1563-66-2)

8.37.7 Clothianidin (D3, CAS # 210880-92-5)

8.37.8 Diazinon (diethyl-D10, CAS # 333-41-5)

8.37.9 Dicamba (D3, CAS # 1918-00-9)

8.37.10 Imidacloprid (D4, CAS # 138261-41-3)

8.37.11 Methomyl (Acetohydroxamate-13C2, 15N, CAS # 16752-77-5)

8.37.12 Simazine (diethyl-D10, CAS # 122-34-9)

8.37.13 Tebuconazole (tert-Butyl-D9, CAS # 107534-96-3)

8.37.14 Thiamethoxam (D3, CAS # 153719-23-4)

[7] A Millipore IC Millex-LG PTFE/0.2µm membrane syringe driven membrane filter unit (Millex is a trademark of Merck KGAA, Darmstadt, Germany) has been found suitable for use for this method, any filter unit may be used that meets the performance of this method may be used.

[8] *Reagent Chemicals, American Chemical Society Specifications*, American Chemical Society, Washington, DC. For suggestions on the testing of reagents not listed by the American Chemical Society, see *Analar Standards for Laboratory Chemicals*, BDH Ltd., Poole, Dorset, U.K., and the United States *Pharmacopeia and National Formulary*, U.S. Pharmacopeial Convention, Inc. (USPC), Rockville, MD.

9. Hazards

9.1 Normal laboratory safety applies to this method. Analysts should wear safety glasses, gloves, and lab coats when working in the lab. Analysts should review the Safety Data Sheets (SDS) for all reagents used in this method.

10. Sampling and Preservation

10.1 Grab samples are collected in amber glass containers with Teflon™ lined caps, such as, 40 mL amber VOA vials. As part of the overall quality assurance program for this test method, field blanks exposed to the same field conditions as samples are collected and analyzed according to this standard to assess the potential for field contamination, refer to Guide D3856 as a guide for sampling. This test method is based upon an 8 mL sample size per analysis. If different sample sizes are used, spiking solution amounts may need to be modified. EPA publication SW-846 may be used as a sampling guide. Samples shall be shipped with a trip blank and at less than 6ºC.

10.2 Once received the sample temperature is taken and should be less than 6ºC. If the receiving temperature is greater than 6ºC, the sample temperature is noted in the case narrative accompanying the data. Samples should be stored refrigerated between 0ºC and 6ºC from the time of collection until analysis.

10.3 The samples should be analyzed within 14 days of collection. No holding time study has been done on water matrices tested in this test method. Holding time may vary depending on the matrix and individual laboratories should determine the holding time in their matrix, refer to Practice D4841.

11. Preparation of LC/MS/MS

11.1 *LC Chromatograph Operating Conditions:*

11.1.1 Injections of all standards and samples are made at a 25 or 50 µL volume. Other injection volumes may be used to optimize conditions. Standards and sample extracts shall be in a 80:20 water:methanol solution. In the case of extreme concentration differences amongst samples, it is wise to analyze a blank after a concentrated sample and before a dilute sample to minimize carry-over of analytes from injection to injection. However, there should not be carry-over between samples. The LC utilized to develop this test method has a flow through LC needle design. The gradient conditions for the two liquid chromatography analysis runs are shown in Tables 2 and 3. The primary SRM transition chromatograms at the lowest calibration level are shown in the Appendix, Figs. X1.1-X1.5.

11.2 *LC Auto Sampler Conditions:*

11.2.1 *Needle Wash Solvent*—60 % acetonitrile/40 % 2-propanol. 8 second wash time before and after injection. Instrument manufacturer's specifications should be followed in order to eliminate sample carry-over.

11.2.2 *Temperatures*—Column, 35°C; Sample compartment, 15°C.

11.2.3 *Seal Wash*—Solvent: 50 % methanol/50 % water; Time: 5 minutes.

11.3 *Mass Spectrometer Parameters:*

11.3.1 To acquire the maximum number of data points per SRM channel while maintaining adequate sensitivity, the tune parameters may be optimized according to the instrument used. Each peak requires at least 10 scans per peak for adequate quantitation. Variable parameters regarding SRM transitions, and cone and collision energies are shown in Table 4. Mass spectrometer parameters used in the development of this method are listed below.

11.3.2 The instrument is set in the Electrospray source setting. The values for the following parameters are shown here for information only. These conditions should be checked and optimized when required.

Methanol/Water/Ammonium Formate Analysis Run Conditions

Capillary Voltage: 1 kV in both ESI modes
Cone: Variable depending on analyte
Source Offset (V) 10
Source Temperature: 150°C
Desolvation Gas Temperature: 500°C
Desolvation Gas Flow: 900 L/hr
Cone Gas Flow: 150 L/hr
Collision Gas Flow: 0.15 mL/min
Low Mass Resolution 1: 2.7
High Mass Resolution 1: 14.7
Ion Energy 1: 0.5
Entrance Energy: 1
Collision Energy: Variable depending on analyte
Exit Energy: 1
Low Mass Resolution 2: 2.8
High Mass resolution 2: 14.7
Ion Energy 2: 1.5
Gain: 1.0
Multiplier: 535
Inter-Scan Delay: 0.003 seconds
Polarity Switching Inter-scan Delay: 0.020 seconds

Methanol/Water/Formic Acid Analysis Run Conditions

Capillary Voltage: Positive mode 2 kV, Negative mode 0.75 kV
Cone: Variable depending on analyte
Source Offset (V) 10
Source Temperature: 150°C
Desolvation Gas Temperature: 300°C
Desolvation Gas Flow: 1000 L/hr
Cone Gas Flow: 300 L/hr
Collision Gas Flow: 0.15 mL/min
Low Mass Resolution 1: 2.7
High Mass Resolution 1: 14.7
Ion Energy 1: 0.5
Entrance Energy: 1
Collision Energy: Variable depending on analyte
Exit Energy: 1
Low Mass Resolution 2: 2.8
High Mass resolution 2: 14.7
Ion Energy 2: 1.5
Gain: 1.0
Multiplier: 535
Inter-Scan Delay: 0.003 seconds
Polarity Switching Inter-scan Delay: 0.020 seconds

12. Calibration and Standardization

12.1 The mass spectrometer shall be calibrated as per manufacturer's specifications before analysis. Analytical values satisfying test method criteria have been achieved using the following procedures. Prepare all solutions in the lab using Class A volumetric glassware.

12.2 *Calibration and Standardization*—To generate a calibration curve, analyze seven calibration standards of the pesticide compounds prior to sample analysis as shown in Table 6. Calibration stock standard solution is prepared from the target and surrogate spike solutions directly to ensure

TABLE 6 Concentrations of Calibration Standards (ng/L)

Pesticide and Surrogate Concentrations (ng/L)	LV1	LV2	LV3	LV4	LV5	LV6	LV7
Azoxystrobin	25	50	100	200	400	800	1 000
Atrazine, Desethylatrazine, Clothianidin, Diazinon, Imidacloprid, Metolachlor, Propiconazole, Simazine, Tebuconazole, Thiamethoxam, Atrazine (ethyl-D5), Desethylatrazine (iso-propyl-D7), Clothianidin-D3, Diazinon (diethyl-D10), Imidacloprid-D4, Simazine (diethyl-D10), Tebuconazole (tert-Butyl-D9), Thiamethoxam-D3	50	100	200	400	800	1 600	2 000
Desisopropylatrazine, Malathion, Metribuzin, Desisopropylatrazine (ethyl-D5)	100	200	400	800	1 600	3 200	4 000
2,4-D, Acetochlor, Alachlor, Aldicarb, Bentazon, Carbaryl, Chlorpyrifos, Fipronil, Methomyl, 2,4-D (Ring-D3), Bentazon-D7, Carbofuran (Ring-13C6), Methomyl (Acetohydroxamate-13C2, 15N)	200	400	800	1600	3 200	6 400	8 000
Triclopyr	1 000	2 000	4 000	8 000	16 000	32 000	40 000
Picloram	5 000	10 000	20 000	40 000	80 000	160 000	200 000
Dicamba, Dicamba-D3	10 000	20 000	40 000	80 000	160 000	320 000	400 000
Clopyralid	20 000	40 000	80 000	160 000	320 000	640 000	800 000

consistency. Stock standard Solution A containing the pesticides is prepared at Level 7 concentration and aliquots of that solution are diluted to prepare Levels 1 through 6. The following steps will produce 1 mL calibration standards with the concentration values shown in Table 6. The analyst is responsible for recording initial component weights carefully when working with pure materials and correctly carrying the weights through the dilution calculations.

12.2.1 At a minimum, five calibration levels are required when using a linear calibration curve and six calibration levels are required when using a quadratic calibration curve. An initial seven point curve may be used to allow for the dropping of the lower level calibration point if the individual laboratory's instrument can't achieve low detection limits. This should allow for at least a five or six point calibration curve to be obtained.

12.2.2 Calibration stock standard Solution A (Level 7, Table 6) is prepared from the target and surrogate spike solutions directly to ensure consistency. 1.25 mL of the surrogate spike and 1.25 mL of the pesticide Target Spike Solution is added to a 50 mL volumetric flask and brought up to 50 mL volume with 80:20 water and methanol solution. This stock standard Solution A (Level 7, Table 6) is diluted to prepare Levels 1 through 6 as shown in Tables 6 and 7. The preparation of the Level 7 standard can be accomplished using appropriate volumes and concentrations of stock solutions as per a particular laboratory's standard procedure.

12.2.3 Aliquots of Solution A are then diluted with 80:20 water:methanol to prepare the 1 mL desired calibration levels in 2 mL amber glass LC vials, as described in Table 7. The calibration vials shall be used within 24 hours to ensure optimum results. Calibration standards are not filtered.

12.2.4 Inject each standard and obtain its chromatogram. An external calibration technique is used to monitor the primary and confirmatory SRM transitions of the pesticides and surrogates. Calibration software is utilized to conduct the quantitation of the analytes using the primary SRM transition. The ratios of the primary/confirmatory SRM transitions area counts are given in Table 5 and will vary depending on the individual tuning conditions. The primary/confirmatory SRM transitions area ratio shall be within 35 % of the individual labs' accepted primary/confirmatory SRM transitions area ratio. The primary SRM transition of the analytes are used for quantitation and the confirmatory SRM transitions for confirmation. This gives added confirmation by isolating the parent ion, forming product ions via fragmentation, and relating it to the retention time in the calibration standard. Metribuzin and the surrogates only have a primary SRM transition.

12.2.5 Depending on sensitivity and matrix interference issues dependent on sample type, a confirmatory SRM transition may be substituted as the primary SRM transition for quantitation during analysis. This shall be explained in a narrative accompanying the data. New primary/confirmatory ion ratios will then be determined if switching the SRM transitions used to quantitate and confirm. The new primary/confirmatory SRM transitions area ratio is required to be within 35 % of the individual labs' new primary/confirmatory SRM transitions area ratio.

12.2.6 The calibration software manual should be consulted to use the software correctly. The quantitation method is set as

TABLE 7 Preparation of Calibration Standards

Solution	LV1	LV2	LV3	LV4	LV5	LV6	LV7
A[A]	25 µL	50 µL	100 µL	200 µL	400 µL	800 µL	1000 µL
B[B]	975 µL	950 µL	900 µL	800 µL	600 µL	200 µL	0 µL

[A] Solution A: Level 7 stock solution prepared according to section 12.2 and at Table 6 concentrations.
[B] Solution B: 80 % Water : 20 % Methanol.

an external calibration using the peak areas in ng/L units. Concentrations may be calculated using the data system software to generate linear regression or quadratic calibration curves. Forcing the calibration curve through the origin (X=0, Y=0) is not recommended.

12.2.7 Linear calibration may be used if the coefficient of determination, r^2, is ≥ 0.98 for the analyte. The point of origin is excluded and a fit weighting of 1/X is used in order to give more emphasis to the lower concentrations. If one of the calibration standards other than the high or low point causes the r^2 of the curve to be <0.98, this point shall be re-injected or a new calibration curve shall be regenerated. Each calibration point used to generate the curve shall have a calculated percent deviation less than 30 % from the generated curve. If the low or high point(s), or both, are excluded, minimally a five point curve is acceptable but the reporting range shall be modified to reflect this change.

12.2.8 Quadratic calibration may be used if the coefficient of determination, r^2, is ≥0.99 for the analyte. The point of origin is excluded, and a fit weighting of 1/X is used in order to give more emphasis to the lower concentrations. If one of the calibration standards causes the curve to be <0.99, this point shall be re-injected or a new calibration curve shall be regenerated. If the low or high point(s), or both, are excluded, minimally a six point curve is acceptable but the reporting range shall be modified to reflect this change. Each calibration point used to generate the curve shall have a calculated percent deviation less than 30 % from the generated curve.

12.2.9 The retention time window of the SRM transitions shall be within 5 % of the retention time of the analyte in a midpoint calibration standard. If this is not the case, re-analyze the calibration curve to determine if there was a shift in retention time during the analysis and the sample needs to be re-injected. If the retention time is still incorrect in the sample, refer to the analyte as an unknown.

12.2.10 A midpoint calibration check standard shall be analyzed at the end of each batch of 30 samples or within 24 hours after the initial calibration curve was generated, the criteria in the individual labs' quality system may be more restrictive pertaining to the number of samples. This end calibration check should come from the same calibration standard solution that was used to generate the initial curve. The results from the end calibration check standard shall have a percent deviation less than 30 % from the calculated concentration for the target analyte. If the results are not within these criteria, corrective action including re-occurrence minimization is performed and either all samples in the batch are re-analyzed against a new calibration curve or the affected results are qualified with an indication that they do not fall within the performance criteria of the test method. If the analyst inspects the vial containing the end calibration check standard and notices that the sample evaporated affecting the concentration or other anomaly, a new end calibration check standard may be made and analyzed. If this new end calibration check standard has a percent deviation less than 30 % from the calculated concentration for the target analyte, the results may be reported unqualified.

12.3 If a laboratory has not performed the test before or if there has been a major change in the measurement system, for example, new analyst, new instrument, etc., an instrument qualification study including reporting limit check sample (RLCS), calibration range determination and precision and bias determination shall be performed to demonstrate laboratory capability.

12.3.1 Analyze at least four replicates of a spiked water sample containing the pesticides at a prepared sample concentration in the calibration range of Levels 3–6. A Level 4 prepared sample concentration was used to set the QC acceptance criteria in this method. The matrix and chemistry should be similar to the matrix used in this test method. Each replicate shall be taken through the complete analytical test method including any sample manipulation and preparation steps.

12.3.2 Calculate the mean (average) percent recovery and relative standard deviation (RSD) of the four values and compare to the acceptable ranges of the QC acceptance criteria for the Initial Demonstration of Performance in Table 8.

12.3.3 This study should be repeated until the single operator precision and mean recovery are within the limits in Table 8. If a concentration other than the recommended concentration is used, refer to Practice D5847 for information on applying the F test and t test in evaluating the acceptability of the mean and standard deviation.

12.3.3.1 The QC acceptance criteria for the Initial Demonstration of Performance in Table 8 were generated from the single-laboratory data shown in the Precision and Bias Section 16. Data from reagent water and surface water are shown in the Precision and Bias Section 16. It is recommended that each laboratory determine in-house QC acceptance criteria which meet or exceed the criteria in this standard. References generating QC acceptance criteria are ASTM Practices D2777, D5847, E2554 and Method 8000 in EPA publication SW-846.

12.4 *Surrogate Spiking Solution:*

12.4.1 A surrogate spiking solution containing fourteen isotopically-labeled pesticides (listed in Section 8) are added to all samples. 50 µL of a methanolic solution containing the surrogates and concentrations are listed in Table 9, Concentrations in Surrogate Spike Solution, is added to all 8 mL samples to achieve the concentration in the sample listed in Table 9, Concentration in Water Sample.

12.4.2 The result obtained for the surrogates shall fall within the limits in Table 8.

12.4.3 There are fourteen surrogates for this analysis. The isotopically-labeled surrogate represents the unlabeled native analyte. Carbofuran (Ring-13C6) represents carbaryl in this standard. No qualifications based on surrogate recovery need to be made for the analytes that do not have representative surrogates. It is left to the analyst's judgment to qualify data based upon non-representative surrogates. The user of the data must also make decisions based on all QC available. If the result is not within these limits, sample analysis is halted until corrective action resolving the problem has been performed. Impacted samples in the batch are either re-analyzed, or the results are flagged with a qualifier stating that they do not fall within the performance criteria of the test method.

12.5 *Method Blank:*

TABLE 8 QC Acceptance Criteria

NOTE 1—Table 8 data is preliminary until a multi-lab validation study is completed.

Analyte	Spiked Sample Conc. (ng/L)	Initial Demonstration of Performance			Laboratory Control Sample	
		Recovery (%)		Precision	Recovery (%)	
		Lower Limit	Upper Limit	Maximum % RSD	Lower Limit	Upper Limit
2,4-D	2 000	70	130	30	70	130
Acetochlor	2 000	70	130	30	70	130
Alachlor	2 000	70	130	30	70	130
Aldicarb	2 000	70	130	30	70	130
Atrazine	500	70	130	30	70	130
Desethylatrazine	500	70	130	30	70	130
Desisopropylatrazine	1 000	70	130	30	70	130
Azoxystrobin	250	70	130	30	70	130
Bentazon	2 000	70	130	30	70	130
Carbaryl	2 000	50	130	30	50	130
Chlorpyrifos	2 000	50	130	30	50	130
Clopyralid	200 000	70	130	30	70	130
Clothianidin	500	70	130	30	70	130
Diazinon	500	70	130	30	70	130
Dicamba	100 000	70	130	30	70	130
Fipronil	2 000	60	130	30	60	130
Imidacloprid	500	70	130	30	70	130
Malathion	1 000	40	130	30	40	130
Methomyl	2 000	70	130	30	70	130
Metolachlor	500	70	130	30	70	130
Metribuzin	1 000	70	130	30	70	130
Picloram	50 000	70	130	30	70	130
Propiconazole	500	50	130	30	50	130
Simazine	500	70	130	30	70	130
Tebuconazole	500	70	130	30	70	130
Thiamethoxam	500	70	130	30	70	130
Triclopyr	10 000	70	130	30	70	130
Surrogates	NA	NA	NA	NA	NA	NA
2,4-D (Ring-D3)	2 000	70	130	30	70	130
Atrazine (ethyl-D5)	500	70	130	30	70	130
Desethylatrazine (iso-propyl-D7)	500	70	130	30	70	130
Desisopropylatrazine (ethyl-D5)	1 000	70	130	30	70	130
Bentazon -D7	2 000	70	130	30	70	130
Carbofuran (Ring-13C6)	2 000	70	130	30	70	130
Clothianidin-D3	500	70	130	30	70	130
Diazinon-(diethyl-D10)	500	70	130	30	70	130
Dicamba-D3	100 000	70	130	30	70	130
Imidacloprid-D4	500	70	130	30	70	130
Methomyl (Acetohydroxamate-13C2, 15N)	2 000	70	130	30	70	130
Simazine (diethyl-D10)	500	70	130	30	70	130
Tebuconazole (tert-Butyl-D9)	500	70	130	30	70	130
Thiamethoxam-D3	500	70	130	30	70	130

TABLE 9 Surrogate Spike Concentrations

Surrogate	Concentration in Surrogate Spike Solution (µg/L)	Concentration in Water Sample (ng/L)
Atrazine (ethyl-D5), Desethylatrazine (iso-propyl-D7), Clothianidin-D3, Diazinon (diethyl-D10), Imidacloprid-D4, Simazine (diethyl-D10), Tebuconazole (tert-Butyl-D9), Thiamethoxam-D3	80	500
Desisopropylatrazine (ethyl-D5)	160	1000
2,4-D (Ring-D3), Bentazon-D7, Carbofuran (Ring-13C6), Methomyl (Acetohydroxamate-13C2, 15N)	320	2000
Dicamba-D3	16000	100000

12.5.1 A method blank for every 30 samples is prepared in 8 mL of reagent water, which is taken through the sample preparation Section 13, to investigate for contamination during sample preparation. The concentration of target analytes in the blank shall be at less than 25 % of the reporting limit or the data shall be qualified as having a blank issue and the reporting limit shall be raised to at least 3 times above the blank contamination concentration.

12.6 *Reporting Limit Check Sample (RLCS):*

12.6.1 Each batch or within the 24 hour analysis window a reporting limit check sample shall be analyzed. The reporting limit check sample is processed like a Laboratory Control Sample just spiked at or near (1–2 times) the reporting limit. The concentration of the RLCS may be reported below the reporting limit since the spike is at or near the reporting limit. This sample is to ensure if the analytes were present at the reporting limit that they would be identified. The recovery limits for the RLCS are 35 to 150 %, if any analytes are outside of these limits the QC exceedance is explained in a narrative

accompanying the data or the batch is re-prepared and analyzed. A continued failure shall be explained, investigated and should be corrected.

12.6.2 To prepare the RLCS, 8 mL of reagent water is added to a 40 ml VOA vial. The sample is spiked with 6.25 µL of the target spike solution (see section 12.7). The sample is then prepared as described in Section 13.

12.7 *Laboratory Control Sample (LCS):*

12.7.1 Analyze at least one LCS with the pesticides at a mid-level prepared sample concentration. The concentration of pesticides at a prepared sample concentration in the calibration range of Levels 3–6 should be used. The LCS is prepared following the analytical method and analyzed with each batch of 30 samples or less. Each MS/MSD or LCS/LCSD sample is spiked with target pesticides (listed in Section 8) to achieve the concentrations in Table 10, Concentrations in Water Sample. For example, 50 µL of a methanolic pesticide Target Spike Solution shown in Table 10 is spiked into each 8 mL water sample. (The target analyte spiking solution is prepared from intermediate solutions which are prepared from neat standards.) The concentrated stock standard concentration can vary when preparing from neat material. To prepare the LCS, 8 mL of reagent water is added to a 40 mL VOA vial. The sample is spiked with 50 µL of a target spike solution and then taken through the sample preparation Step in Section 13.

12.7.2 The result obtained for the LCS shall fall within the limits in Table 8.

12.7.3 If the result is not within these limits, sample analysis is halted until corrective action resolving the problem has been performed. Impacted samples in the batch are either re-analyzed, or the results are flagged with a qualifier stating that they do not fall within the performance criteria of the test method.

12.8 *Matrix Spike (MS):*

12.8.1 To check for interferences in the specific matrix being tested, perform a MS on at least one sample from each batch of 30 or fewer samples by spiking the sample with a known concentration of pesticides and following the analytical method. The target spike solution from the LCS section 12.7 is used for the spike solution. Spike 50 µL of this stock solution into 8 mL of the site sample to yield the various concentrations in the spike sample as shown in section 12.7. The sample is then taken through the sample preparation Step in Section 13.

TABLE 10 Target Spike Concentrations

Analyte	Concentration in Target Spike Solution (µg/L)	Concentration in Water Sample (ng/L)
Azoxystrobin	40	250
Atrazine, Desethylatrazine, Clothianidin, Diazinon, Imidacloprid, Metolachlor, Propiconazole, Simazine, Tebuconazole, Thiamethoxam	80	500
Desisopropylatrazine, Malathion, Metribuzin	160	1000
2,4-D, Acetochlor, Alachlor, Aldicarb, Bentazon, Carbaryl, Chlorpyrifos, Fipronil, Methomyl	320	2000
Triclopyr	1600	10000
Picloram	8000	50000
Dicamba	16000	100000
Clopyralid	32000	200000

12.8.2 If the spiked concentration plus the background concentration exceeds that of the Level 7 calibration standard, the sample shall be diluted using 80 % water/20 % methanol to a level near the midpoint of the calibration curve.

12.8.3 Calculate the percent recovery of the spike (P) using Eq 1:

$$P = 100\frac{|A(V_S + V) - BV_S|}{CV} \quad (1)$$

where:

A = concentration found in spiked sample,
B = concentration found in unspiked sample,
C = concentration of analyte in spiking solution,
V_S = volume of sample used,
V = volume of spiking solution added, and
P = percent recovery.

12.8.4 The percent recovery of the spike shall fall within the limits in Table 11. If the percent recovery is not within these limits, a matrix interference may be present. Under these circumstances either all samples in the batch may be analyzed by a test method not affected by the matrix interference, or the results shall be qualified indicating that they do not fall within the performance criteria of the test method. It has been found that in some cases the matrix spike concentration may be minimal compared to the concentration in the native sample. If this is the case, the sample may be spiked at a higher level or the generated data may be reported explaining in the narrative accompanying the data that the spike was negligible compared to the native concentration found in the sample.

12.8.5 The matrix spike/matrix spike duplicate (MS/MSD) limits in Table 11 were generated by a single-laboratory study using the data in the Precision and Bias Section 16. The limits in Table 11 are preliminary until a multi-lab validation study is completed. The matrix variation between different waters may have a tendency to generate significantly wider control limits than those generated for this Standard. It is recommended that each laboratory determine in-house QC acceptance criteria meeting or exceeding the criteria stated in this standard.

12.8.5.1 Each laboratory should generate its own in-house QC acceptance criteria after the analysis of 15–20 matrix spike samples of a particular water matrix. References on generating QC acceptance criteria are ASTM Practices D5847, D2777, E2554 and Method 8000 in EPA publication SW-846.

12.9 *Duplicate:*

12.9.1 To check the precision of sample analyses, analyze a sample in duplicate with each batch of 30 or fewer samples. If the sample contains the analyte at a level greater than 5 times the reporting limit of the method, the sample and duplicate may be analyzed unspiked; otherwise, a matrix spike/matrix spike duplicate should be used.

12.9.2 Calculate the relative percent difference (RPD) between the duplicate values or MS/MSD values as shown in Eq 2. Compare to the RPD limit in Table 11.

$$\text{RPD} = \frac{|[\text{MS}] - [\text{MSD}]|}{([\text{MS}] + [\text{MSD}])/2} \times 100 \quad (2)$$

TABLE 11 MS/MSD QC Acceptance Criteria

NOTE 1—Table 11 data is preliminary until a multi-lab validation study is completed.

Analyte	Spiked Sample Conc. (ng/L)	MS/MSD		
		Recovery (%)		Precision
		Lower Limit	Upper Limit	Maximum % RPD
2,4-D	2 000	70	130	30
Acetochlor	2 000	70	130	30
Alachlor	2 000	70	130	30
Aldicarb	2 000	70	130	30
Atrazine	500	70	130	30
Desethylatrazine	500	70	130	30
Desisopropylatrazine	1 000	70	130	30
Azoxystrobin	250	70	130	30
Bentazon	2 000	70	130	30
Carbaryl	2 000	50	130	30
Chlorpyrifos	2 000	50	130	30
Clopyralid	200 000	70	130	30
Clothianidin	500	70	130	30
Diazinon	500	70	130	30
Dicamba	100 000	70	130	30
Fipronil	2 000	60	130	30
Imidacloprid	500	70	130	30
Malathion	1 000	40	130	30
Methomyl	2 000	70	130	30
Metolachlor	500	70	130	30
Metribuzin	1 000	70	130	30
Picloram	50 000	70	130	30
Propiconazole	500	50	130	30
Simazine	500	70	130	30
Tebuconazole	500	70	130	30
Thiamethoxam	500	70	130	30
Triclopyr	10 000	70	130	30

where:

RPD = relative percent difference,
MS = measured concentration in the matrix spike QC sample (to calculate duplicate RPD use—sample concentration), and
MSD = measured concentration in the matrix spike duplicate QC sample (to calculate duplicate RPD use—sample duplicate concentration).

12.9.3 If the result exceeds the precision limit (Table 11), the batch shall be re-analyzed or the results are flagged with a qualifier stating that they do not fall within the performance criteria of the test method.

13. Procedure

13.1 This test method is based upon an 8 mL sample size per analysis. The samples shall be analyzed within 14 days of collection. If samples are received or stored above 6ºC, or are not analyzed within 14 days of collection, it is noted in the case narrative that accompanies the data.

13.2 Each batch of samples (30 or less) shall contain at least a method blank, a laboratory control sample, matrix spike, duplicate and a reporting limit check sample at a minimum.

13.3 In the laboratory, 8 mL sample is placed in a 40 mL amber VOA vial. All samples are spiked with surrogates as described in Section 12. The laboratory control, reporting limit check and matrix spike samples are then spiked with the target compounds as described in Section 12. The samples are then shaken in order to mix the spike solutions throughout the sample.

13.4 To all samples, 2 ml of methanol is added and hand shaken/vortexed for ~1 minute. After mixing, the sample is filtered through a PTFE filter syringe driven filter unit to remove particulates in the samples. An aliquot of the solution, approximately 1 mL, is transferred to a LC vial and a cap is applied. The final volume of the solution is 10 mL for quantitation purposes.

13.4.1 The syringe shall be cleaned between each filtration. It is the analyst's responsibility to ensure that the syringe is clean.

13.5 The analysis of the sample requires two separate analysis methods, one using the LC gradient conditions in Table 2 with the Methanol/Water/Ammonium Formate Analysis Run Conditions in Section 11 and the second using the LC gradient conditions in Table 3 with the Methanol/Water/Formic Acid Run Conditions in Section 11. Each analysis set is to be analyzed separately in two different complete sample sequences which includes the exact same samples and may even include the same calibration level standards. The only analytes reported from the formic acid run conditions are 2,4-D, Clopyralid, Dicamba, Picloram, Triclopyr, 2,4-D (Ring-D3) and Dicamba-D3, the rest of the analytes are reported from the ammonium formate analysis run. It is important to condition the column using the desired run conditions before analysis in order to flush out the previous modifier. A 25 µL injection volume has been found appropriate for the ammonium formate analysis run and a 50 µL injection volume has been found suitable for the formic acid analysis run.

NOTE 1—It has been determined, from experience with this analysis, that at least an hour of flushing with the desired mobile phase is required

when changing between different mobile phases to achieve optimum conditions.

13.6 Each analysis sequence may begin once a passing calibration curve is generated. An order of analysis may be method blank(s), reporting limit check, laboratory control sample(s), sample(s), duplicate(s) and matrix spike sample(s) followed by an end calibration check standard.

14. Calculation or Interpretation of Results

14.1 For quantitative and qualititaive analysis of the pesticides, the SRM transitions are identified by comparison of retention times in the sample to those of the standards. Most of the pesticides (Except for Metribuzin and the surrogates) are identified by comparing the sample primary SRM transition and its confirmatory SRM transitions if correlated to the known standard SRM transitions. The primary/confirmatory SRM ion ratios shall meet the criteria set in the quantitation method by ±35 %. The primary/confirmatory SRM ion ratios is the average of the individual levels primary/confirmatory SRM ion ratios in the calibration curve on the day of analysis. These ratios will vary depending on the instrument acquisition parameters and must be checked for every sample batch. External calibration curves are used to calculate the amounts of pesticides. Calculate the concentration in ng/L for each pesticide. A pesticide may be reported if present at or above the reporting limit. If the concentration of the analyte is determined to be above the calibration range, the sample is diluted with a solution of 80 % water/ 20 % methanol to obtain a concentration near the mid-point of the calibration range and re-analyzed.

14.1.1 If the ion ratios do not meet the criteria for the pesticides that have a secondary SRM transition listed, the compound is determined to be an unknown.

14.2 *Example Calculation of Sample Concentration Reported:*

14.2.1 The concentration of sample is calculated using Eq 3.

$$\frac{V_f}{V_i}\,(C_u) = C_f \tag{3}$$

where:

V_f = final volume (10 mL–8 mL sample plus 2 mL methanol),
V_i = initial volume (8 mL),
C_u = uncorrected concentration (concentration from instrument, uncorrected for volume variation), and
C_f = final concentration (corrected for dilution).

15. Report

15.1 Determine the results in units of ng/L (ppt) in a water sample. Calculate the concentration in the sample using the linear or quadratic calibration curve generated. All data that do not meet the specifications in the test method shall be appropriately qualified.

16. Precision and Bias

16.1 The determination of precision and bias was conducted by US EPA Region 5 Chicago Regional Laboratory (CRL) and generated applicable data to determine the precision and bias as described in Practice D2777 for a single laboratory validation study.

16.2 This test method was tested by CRL on reagent water, lake and river water. The samples were spiked with the pesticides and surrogates as described in Section 12. Tables 12-14 contain the recoveries for the pesticides and surrogates in the water matrices tested.

17. Quality Control

17.1 A crucial part of a test method is quality control. A laboratory should follow their in-house QA/QC procedures and should meet or exceed the criteria given in this test method. The quality-control criteria are given in the various test method sections. Section 10 contains the sampling and preservation requirements and Section 12 contains the majority of quality-control requirements when following this test method. Section 12 includes requirements for calibration, precision and bias study to demonstrate laboratory capability, initial demonstration of performance, surrogate, method blank, reporting limit check, laboratory control, matrix spike and duplicate sample requirements. An IRM should be incorporated into the analysis periodically to verify that standard concentrations are comparable between sources. The IRM criteria should be based upon the laboratories QA/QC policies and the individual data quality objectives.

18. Keywords

18.1 2,4-dichlorophenoxyacetic acid; acetochlor; alachlor; aldicarb; atrazine; azoxystrobin; bentazon; carbaryl; chlorpyrifos; clopyralid; clothianidin; desethylatrazine; desisopropylatrazine; diazinon; dicamba; fipronil; imidacloprid; liquid chromatography; malathion; mass spectrometry; methomyl; metolachlor; metribuzin; pesticides; picloram; propiconazole; simazine; tebuconazole; thiamethoxam; triclopyr; water

TABLE 12 Precision and Accuracy Study in ASTM Type I Water

Table 12a. Precision and Accuracy Study for Pesticides in ASTM Type I Water

Sample	Methomyl	Desisopropl-atrazine	Desethyl-atrazine	Simazine	Carbaryl	Aldicarb	Metribuzin	Atrazine	Clothianidin
	Concentration (ng/L)								
MB1	<RL	<RL	<RL	<RL	<RL	<RL	<RL	<RL	<RL
MB2	<RL	<RL	<RL	<RL	<RL	<RL	<RL	<RL	<RL
P&A1	1787	923	447	437	1572	1771	865	446	440
P&A2	1780	930	453	442	1575	1777	873	448	431
P&A3	1672	870	428	417	1331	1721	832	423	417
P&A4	1752	927	448	431	1525	1816	889	448	451
P&A5	1726	919	452	435	1074	1774	868	448	449
P&A6	1738	905	442	429	1532	1771	857	440	437
Spike Level (ng/L)	2000	1000	500	500	2000	2000	1000	500	500
Average Recovery (ng/L)	1742.5	912.3	445.0	431.8	1434.8	1771.7	864.0	442.2	437.5
% Average Recovery	87.1	91.2	89.0	86.4	71.7	88.6	86.4	88.4	87.5
Standard Deviation	41.8	22.5	9.2	8.6	198.5	30.2	19.0	9.9	12.5
RSD (%)	2.4	2.5	2.1	2.0	13.8	1.7	2.2	2.2	2.9

Table 12b. Precision and Accuracy Study for Pesticides in ASTM Type I Water

Sample	Alachlor	Metolachlor	Thiamethoxam	Diazinon	Tebuconazole	Malathion	Propiconazole	Chlorpyrifos	Azoxystrobin
	Concentration (ng/L)								
MB1	<RL	<RL	<RL	<RL	<RL	<RL	<RL	<RL	<RL
MB2	<RL	<RL	<RL	<RL	<RL	<RL	<RL	<RL	<RL
P&A1	1700	430	431	422	425	731	351	1371	213
P&A2	1701	431	436	429	419	713	349	1421	213
P&A3	1654	414	412	404	404	589	337	1365	198
P&A4	1715	431	445	428	416	693	345	1397	213
P&A5	1678	427	428	420	410	421	340	1332	210
P&A6	1681	424	413	420	418	694	336	1358	209
Spike Level (ng/L)	2000	500	500	500	500	1000	500	2000	250
Average Recovery (ng/L)	1688.2	426.2	427.5	420.5	415.3	640.2	343.0	1374.0	209.3
% Average Recovery	84.4	85.2	85.5	84.1	83.1	64.0	68.6	68.7	83.7
Standard Deviation	21.7	6.6	13.0	9.0	7.4	118.2	6.3	31.1	5.8
RSD (%)	1.3	1.5	3.0	2.1	1.8	18.5	1.8	2.3	2.8

Table 12c. Precision and Accuracy Study for Pesticides in ASTM Type I Water

Sample	2,4-D	Dicamba	Picloram	Triclopyr	Clopyralid	Imidacloprid	Acetochlor	Bentazon	Fipronil
	Concentration (ng/L)								
MB1	<RL	<RL	<RL	<RL	<RL	<RL	<RL	<RL	<RL
MB2	<RL	<RL	<RL	<RL	<RL	<RL	<RL	<RL	<RL
P&A1	2081	103205	44522	10160	173933	440	1822	1756	1524
P&A2	2145	107729	44733	10516	183408	438	1782	1799	1477
P&A3	1990	102034	44372	9638	171250	415	1674	1676	1479
P&A4	2093	106461	45688	10421	178997	442	1797	1751	1657
P&A5	2117	110105	46892	10514	179919	437	1765	1740	1470
P&A6	2093	109273	45443	10014	177720	438	1760	1700	1551
Spike Level (ng/L)	2000	100000	50000	10000	200000	500	2000	2000	2000
Average Recovery (ng/L)	2086.5	106467.8	45275.0	10210.5	177537.8	435.0	1766.7	1737.0	1526.3
% Average Recovery	104.3	106.5	90.6	102.1	88.8	87.0	88.3	86.9	76.3
Standard Deviation	52.5	3255.6	947.6	346.0	4354.8	10.0	50.7	43.6	71.5
RSD (%)	2.5	3.1	2.1	3.4	2.5	2.3	2.9	2.5	4.7

Table 12d. Precision and Accuracy Study for Pesticides in ASTM Type I Water

Sample	Methomyl (Surrogate)	Desisopropyl-atrazine (Surrogate)	Desethyl-atrazine (Surrogate)	Simazine (Surrogate)	Atrazine (Surrogate)	Carbofuran (Surrogate)	Clothianidin (Surrogate)
	Concentration (ng/L)						
MB1	1645	887	423	442	486	1766	450
MB2	1672	862	406	416	620	1596	428
P&A1	1710	873	414	421	426	1652	432
P&A2	1736	864	413	419	437	1639	422
P&A3	1576	838	402	406	414	1553	415
P&A4	1830	957	445	449	460	1759	456
P&A5	1739	920	441	439	440	1567	448
P&A6	1776	921	445	447	444	1723	452
Spike Level (ng/L)	2000	1000	500	500	500	2000	500
Average Recovery (ng/L)	1710.5	890.3	423.6	429.9	465.9	1656.9	437.9
% Average Recovery	85.5	89.0	84.7	86.0	93.2	82.8	87.6
Standard Deviation	79.2	39.3	17.7	16.3	66.0	84.2	15.5
RSD (%)	4.6	4.4	4.2	3.8	14.2	5.1	3.5

TABLE 12 *Continued*

Table 12e. Precision and Accuracy Study for Pesticides in ASTM Type I Water

Sample	Bentazon (Surrogate)	2,4-D (Surrogate)	Dicamba (Surrogate)	Imidacloprid (Surrogate)	Thiamethoxam (Surrogate)	Diazinon (Surrogate)	Tebuconazole (Surrogate)
	Concentration (ng/L)						
MB1	1805	1816	103551	436	441	427	425
MB2	1702	1747	101053	421	413	403	398
P&A1	1772	1944	97217	428	427	418	409
P&A2	1740	2030	101434	422	430	421	398
P&A3	1628	1909	97055	407	411	404	395
P&A4	1912	2159	113447	465	458	452	434
P&A5	1773	2113	108914	452	436	433	415
P&A6	1735	2147	114373	450	436	435	428
Spike Level (ng/L)	2000	2000	10000	500	500	500	500
Average Recovery (ng/L)	1758.4	1983.1	104630.5	435.1	431.5	424.1	412.8
% Average Recovery	87.9	99.2	104.6	87.0	86.3	84.8	82.6
Standard Deviation	82.1	154.7	6840.3	19.3	15.2	16.4	15.1
RSD (%)	4.7	7.8	6.5	4.4	3.5	3.9	3.7

TABLE 13 P&A Study in Lake Water

Table 13a. Precision and Accuracy Study for Pesticides in Lake Water

Sample	Methomyl	Desisopropl-atrazine	Desethyl-atrazine	Simazine	Carbaryl	Aldicarb	Metribuzin	Atrazine	Clothianidin
	Concentration (ng/L)								
Unpsiked - Lake 1	<RL	<RL	53[A]	<RL	<RL	<RL	<RL	58[A]	<RL
Unpsiked - Lake 2	<RL	<RL	53[A]	<RL	<RL	<RL	<RL	59[A]	<RL
Lake - P&A1	1776	949	453	442	881	1842	864	448	434
Lake - P&A2	1767	970	462	459	928	1870	883	462	444
Lake - P&A3	1782	937	449	465	906	1821	864	443	431
Lake - P&A4	1658	916	433	441	867	1762	844	434	414
Lake - P&A5	1759	948	449	447	897	1801	873	450	426
Lake - P&A6	1727	918	438	428	863	1821	839	440	407
Spike Level (ng/L)	2000	1000	500	500	2000	2000	1000	500	500
Average Recovery (ng/L)	1744.8	939.7	447.3	447.0	890.3	1819.5	861.2	446.2	426.0
% Average Recovery	87.2	94.0	89.5	89.4	44.5	91.0	86.1	89.2	85.2
Standard Deviation	46.7	20.6	10.4	13.3	24.8	36.6	16.8	9.6	13.5
RSD (%)	2.7	2.2	2.3	3.0	2.8	2.0	2.0	2.2	3.2

Table 13b. Precision and Accuracy Study for Pesticides in Lake Water

Sample	Alachlor	Metolachlor	Thiamethoxam	Diazinon	Tebuconazole	Malathion	Propiconazole	Chlorpyrifos	Azoxystrobin
	Concentration (ng/L)								
Unpsiked - Lake 1	<RL	<RL	<RL	<RL	<RL	<RL	<RL	<RL	<RL
Unpsiked - Lake 2	<RL	<RL	<RL	<RL	<RL	<RL	<RL	<RL	<RL
Lake - P&A1	1689	440	420	427	413	309	347	1302	212
Lake - P&A2	1761	456	429	441	436	323	362	1362	220
Lake - P&A3	1648	439	420	419	414	307	342	1299	213
Lake - P&A4	1635	430	405	415	409	297	345	1239	205
Lake - P&A5	1719	446	414	427	410	311	349	1326	214
Lake - P&A6	1665	438	412	415	401	296	338	1294	212
Spike Level (ng/L)	2000	500	500	500	500	1000	500	2000	250
Average Recovery (ng/L)	1686.2	441.5	416.7	424.0	413.8	307.2	347.2	1303.7	212.7
% Average Recovery	84.3	88.3	83.3	84.8	82.8	30.7	69.4	65.2	85.1
Standard Deviation	47.3	8.8	8.2	9.9	11.8	10.0	8.2	40.5	4.8
RSD (%)	2.8	2.0	2.0	2.3	2.8	3.2	2.4	3.1	2.3

Table 13c. Precision and Accuracy Study for Pesticides in Lake Water

Sample	2,4-D	Dicamba	Picloram	Triclopyr	Clopyralid	Imidacloprid	Acetochlor	Bentazon	Fipronil
	Concentration (ng/L)								
Unpsiked - Lake 1	<RL	<RL	<RL	<RL	<RL	<RL	<RL	<RL	<RL
Unpsiked - Lake 2	<RL	<RL	<RL	<RL	<RL	<RL	<RL	<RL	<RL
Lake - P&A1	2109	109247	41166	10527	140292	1986	1539	435	1785
Lake - P&A2	2199	113745	42680	10283	131810	2008	1568	443	1860
Lake - P&A3	2040	106082	41093	10212	140497	1943	1567	435	1782
Lake - P&A4	2012	106303	39880	9616	126828	2007	1451	417	1757
Lake - P&A5	2095	102671	40641	10379	128909	1981	1471	425	1794
Lake - P&A6	2029	101309	39589	9824	131041	2056	1593	418	1777
Spike Level (ng/L)	2000	100000	50000	10000	200000	2000	2000	500	2000
Average Recovery (ng/L)	2080.7	106559.5	40841.5	10140.2	133229.5	1996.8	1531.5	428.8	1792.5
% Average Recovery	104.0	106.6	81.7	101.4	66.6	99.8	76.6	85.8	89.6
Standard Deviation	69.4	4513.0	1102.9	348.4	5816.3	37.4	57.6	10.5	35.3
RSD (%)	3.3	4.2	2.7	3.4	4.4	1.9	3.8	2.4	2.0

Table 13d. Precision and Accuracy Study for Pesticides in Lake Water

Sample	Methomyl (Surrogate)	Desisopropyl-atrazine (Surrogate)	Desethyl-atrazine (Surrogate)	Simazine (Surrogate)	Atrazine (Surrogate)	Carbofuran (Surrogate)	Clothianidin (Surrogate)
	Concentration (ng/L)						
Unpsiked - Lake 1	1655	857	418	422	420	1504	426
Unpsiked - Lake 2	1572	851	401	416	414	1462	409
Lake - P&A1	1665	855	410	419	414	1436	407
Lake - P&A2	1625	850	403	414	411	1421	399
Lake - P&A3	1772	927	444	452	451	1549	446
Lake - P&A4	1707	862	422	434	433	1501	415
Lake - P&A5	1674	877	418	427	427	1476	402
Lake - P&A6	1537	778	374	376	385	1308	364
Spike Level (ng/L)	2000	1000	500	500	500	2000	500
Average Recovery (ng/L)	1650.9	857.1	411.3	420.0	419.4	1457.1	408.5
% Average Recovery	82.5	85.7	82.3	84.0	83.9	72.9	81.7
Standard Deviation	74.1	40.8	20.1	21.6	19.1	72.6	23.5
RSD (%)	4.5	4.8	4.9	5.1	4.6	5.0	5.8

TABLE 13 *Continued*

Table 13e. Precision and Accuracy Study for Pesticides in Lake Water							
Sample	Bentazon (Surrogate)	2,4-D (Surrogate)	Dicamba (Surrogate)	Imidacloprid (Surrogate)	Thiamethoxam (Surrogate)	Diazinon (Surrogate)	Tebuconazole (Surrogate)
	Concentration (ng/L)						
Unpsiked - Lake 1	1846	1881	111651	417	410	409	401
Unpsiked - Lake 2	1900	1901	113120	413	408	400	383
Lake - P&A1	1932	1979	102955	413	406	411	402
Lake - P&A2	1895	2035	104073	405	407	411	390
Lake - P&A3	1989	2157	109894	452	442	435	425
Lake - P&A4	1966	2022	103938	417	421	416	406
Lake - P&A5	1959	2048	102544	420	409	420	398
Lake - P&A6	1801	1795	91513	371	374	373	348
Spike Level (ng/L)	2000	2000	100000	500	500	500	500
Average Recovery (ng/L)	1911.0	1977.3	104961.0	413.5	409.6	409.4	394.1
% Average Recovery	95.6	98.9	105.0	82.7	81.9	81.9	78.8
Standard Deviation	63.9	114.0	6847.4	22.1	18.8	17.8	22.3
RSD (%)	3.3	5.8	6.5	5.4	4.6	4.4	5.7

[A] Below Reporting Limit—Average Subtracted from P&A Data.

TABLE 14 P&A Study in River Water

Table 14a. Precision and Accuracy Study for Pesticides in River Water

Sample	Methomyl	Desisopropl-atrazine	Desethyl-atrazine	Simazine	Carbaryl	Aldicarb	Metribuzin	Atrazine	Clothianidin
	Concentration (ng/L)								
Unpsiked - River 1	<RL	<RL	<RL	<RL	<RL	<RL	<RL	26[A]	<RL
Unpsiked - River 2	<RL	<RL	<RL	<RL	<RL	<RL	<RL	27[A]	<RL
River - P&A1	1517	760	397	385	1119	1646	741	405	345
River - P&A2	1540	766	399	388	1162	1663	750	404	344
River - P&A3	1531	800	411	398	1201	1752	766	411	357
River - P&A4	1568	811	417	404	1214	1710	781	422	369
River - P&A5	1672	861	437	418	1248	1817	812	440	398
River - P&A6	1599	835	428	414	1189	1741	797	435	391
Spike Level (ng/L)	2000	1000	500	500	2000	2000	1000	500	500
Average Recovery (ng/L)	1571.2	805.5	414.8	401.2	1188.8	1721.5	774.5	419.5	367.3
% Average Recovery	78.6	80.6	83.0	80.2	59.4	86.1	77.5	83.9	73.5
Standard Deviation	57.4	39.1	15.8	13.4	44.4	62.7	27.4	15.4	23.0
RSD (%)	3.7	4.9	3.8	3.3	3.7	3.6	3.5	3.7	6.3

Table 14b. Precision and Accuracy Study for Pesticides in River Water

Sample	Alachlor	Metolachlor	Thiamethoxam	Diazinon	Tebuconazole	Malathion	Propiconazole	Chlorpyrifos	Azoxystrobin
	Concentration (ng/L)								
Unpsiked - River 1	<RL	<RL	<RL	<RL	69[B]	<RL	38[A]	<RL	<RL
Unpsiked - River 2	<RL	<RL	<RL	<RL	69[B]	<RL	38[A]	<RL	<RL
River - P&A1	1525	406	373	396	369	433	306	1124	196
River - P&A2	1567	403	369	400	379	462	318	1153	199
River - P&A3	1583	413	381	411	385	469	331	1184	203
River - P&A4	1611	422	384	417	407	478	342	1233	206
River - P&A5	1640	426	402	421	417	493	347	1222	212
River - P&A6	1638	426	401	422	410	472	344	1286	210
Spike Level (ng/L)	2000	500	500	500	500	1000	500	2000	250
Average Recovery (ng/L)	1594.0	416.0	385.0	411.2	394.5	467.8	331.3	1200.3	204.3
% Average Recovery	79.7	83.2	77.0	82.2	78.9	46.8	66.3	60.0	81.7
Standard Deviation	44.6	10.1	13.9	11.0	19.4	20.0	16.4	58.7	6.2
RSD (%)	2.8	2.4	3.6	2.7	4.9	4.3	4.9	4.9	3.0

Table 14c. Precision and Accuracy Study for Pesticides in River Water

Sample	2,4-D	Dicamba	Picloram	Triclopyr	Clopyralid	Imidacloprid	Acetochlor	Bentazon	Fipronil
	Concentration (ng/L)								
Unpsiked - River 1	<RL	<RL	<RL	<RL	<RL	<RL	<RL	45[A]	<RL
Unpsiked - River 2	<RL	<RL	<RL	<RL	<RL	<RL	<RL	45[A]	<RL
River - P&A1	1957	98031	32280	10727	100603	1799	1497	357	1655
River - P&A2	2015	102322	32480	11175	87346	1821	1552	370	1665
River - P&A3	2061	102105	33262	11298	92199	1786	1627	373	1679
River - P&A4	2152	100001	35073	11388	99443	1861	1701	380	1760
River - P&A5	2190	101793	35540	11831	97205	1835	1663	402	1745
River - P&A6	2100	104542	34334	12077	101018	1793	1584	402	1731
Spike Level (ng/L)	2000	100000	50000	10000	200000	2000	2000	500	2000
Average Recovery (ng/L)	2079.2	101465.7	33828.2	11416.0	96302.3	1815.8	1604.0	380.7	1705.8
% Average Recovery	104.0	101.5	67.7	114.2	48.2	90.8	80.2	76.1	85.3
Standard Deviation	86.5	2220.6	1361.7	481.0	5449.1	28.7	74.8	18.1	44.9
RSD (%)	4.2	2.2	4.0	4.2	5.7	1.6	4.7	4.8	2.6

Table 14d. Precision and Accuracy Study for Pesticides in River Water

Sample	Methomyl (Surrogate)	Desisopropyl-atrazine (Surrogate)	Desethyl-atrazine (Surrogate)	Simazine (Surrogate)	Atrazine (Surrogate)	Carbofuran (Surrogate)	Clothianidin (Surrogate)
	Concentration (ng/L)						
Unpsiked - River 1	1698	825	409	432	445	1620	384
Unpsiked - River 2	1417	727	360	375	390	1396	342
River - P&A1	1252	606	299	314	328	1170	289
River - P&A2	1344	674	334	351	365	1304	317
River - P&A3	1498	741	363	384	393	1444	337
River - P&A4	1471	740	359	376	389	1408	345
River - P&A5	1455	722	353	362	379	1347	343
River - P&A6	1431	736	359	370	391	1363	344
Spike Level (ng/L)	2000	1000	500	500	500	2000	500
Average Recovery (ng/L)	1445.8	721.4	354.5	370.5	385.0	1381.5	337.6
% Average Recovery	72.3	72.1	70.9	74.1	77.0	69.1	67.5
Standard Deviation	128.7	62.4	30.7	33.1	32.6	127.6	26.9
RSD (%)	8.9	8.7	8.7	8.9	8.5	9.2	8.0

TABLE 14 *Continued*

Table 14e. Precision and Accuracy Study for Pesticides in River Water

Sample	Bentazon (Surrogate)	2,4-D (Surrogate)	Dicamba (Surrogate)	Imidacloprid (Surrogate)	Thiamethoxam (Surrogate)	Diazinon (Surrogate)	Tebuconazole (Surrogate)
	Concentration (ng/L)						
Unpsiked - River 1	2116	2048	116300	410	419	447	424
Unpsiked - River 2	1755	1799	103738	359	377	385	295
River - P&A1	1474	1588	78569	300	302	326	314
River - P&A2	1677	1791	89338	337	348	368	344
River - P&A3	1788	1897	94466	363	379	397	371
River - P&A4	1734	1919	93149	363	373	389	376
River - P&A5	1635	1795	88166	358	353	372	360
River - P&A6	1645	1814	90983	369	362	380	381
Spike Level (ng/L)	2000	2000	100000	500	500	500	500
Average Recovery (ng/L)	1728.0	1831.4	94338.6	357.4	364.1	383.0	358.1
% Average Recovery	86.4	91.6	94.3	71.5	72.8	76.6	71.6
Standard Deviation	184.2	132.3	11308.2	30.9	33.2	33.7	40.5
RSD (%)	10.7	7.2	12.0	8.6	9.1	8.8	11.3

[A] Below Reporting Limit—Average Subtracted from P&A Data.
[B] Average Subtracted from P&A Data.

APPENDIX

(Nonmandatory Information)

X1. FIGURES (CHROMATOGRAMS)

X1.1 See Figs. X1.1-X1.5.

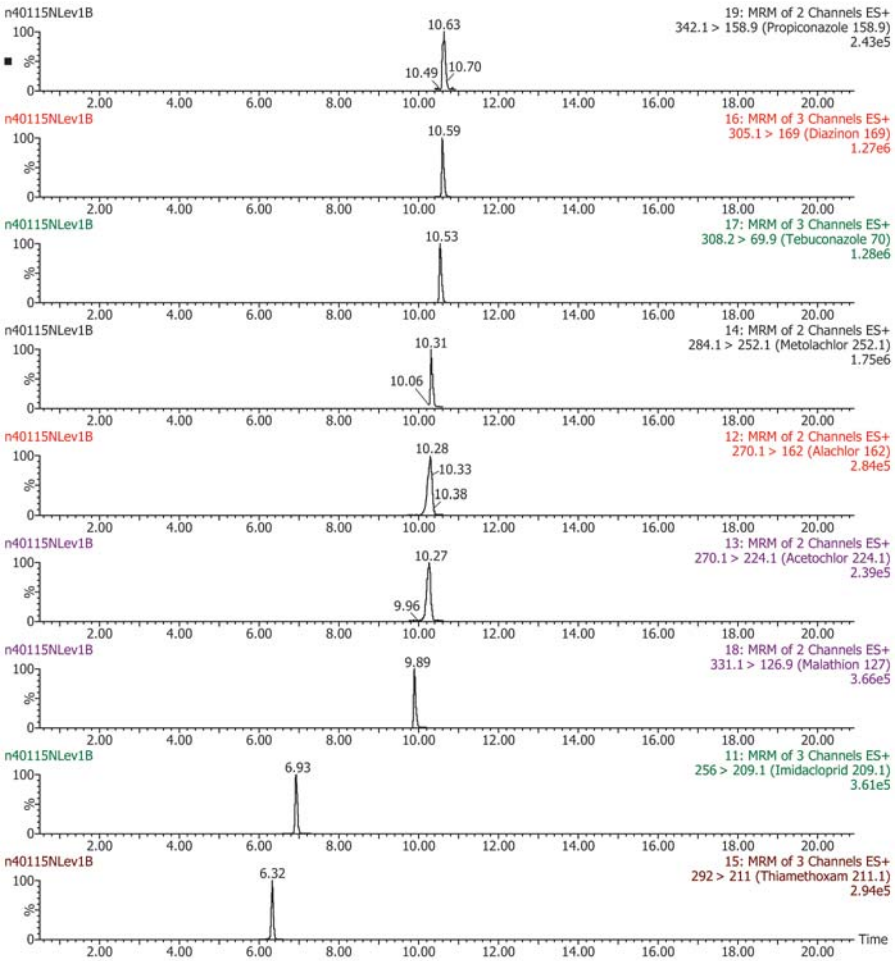

FIG. X1.1 The Primary SRM Transition Chromatograms at the Level 1 Calibration Concentration for Propiconazole, Diazinon, Tebuconazole, Metolachlor, Alachlor, Acetochlor, Malathion, Imidacloprid and Thiamethoxam

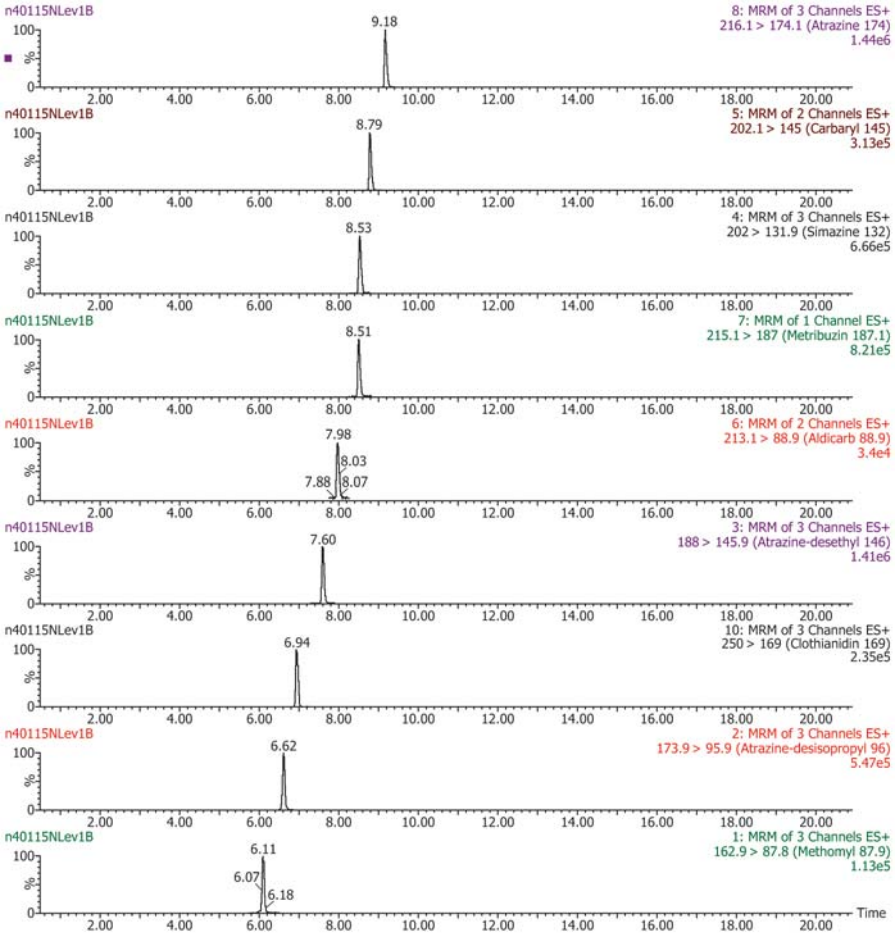

FIG. X1.2 The Primary SRM Transition Chromatograms at the Level 1 Calibration Concentration for Atrazine, Carbaryl, Simazine, Metribuzin, Aldicarb, Atrazine-desethyl, Clothianidin, Atrazine-desisopropyl and Methomyl

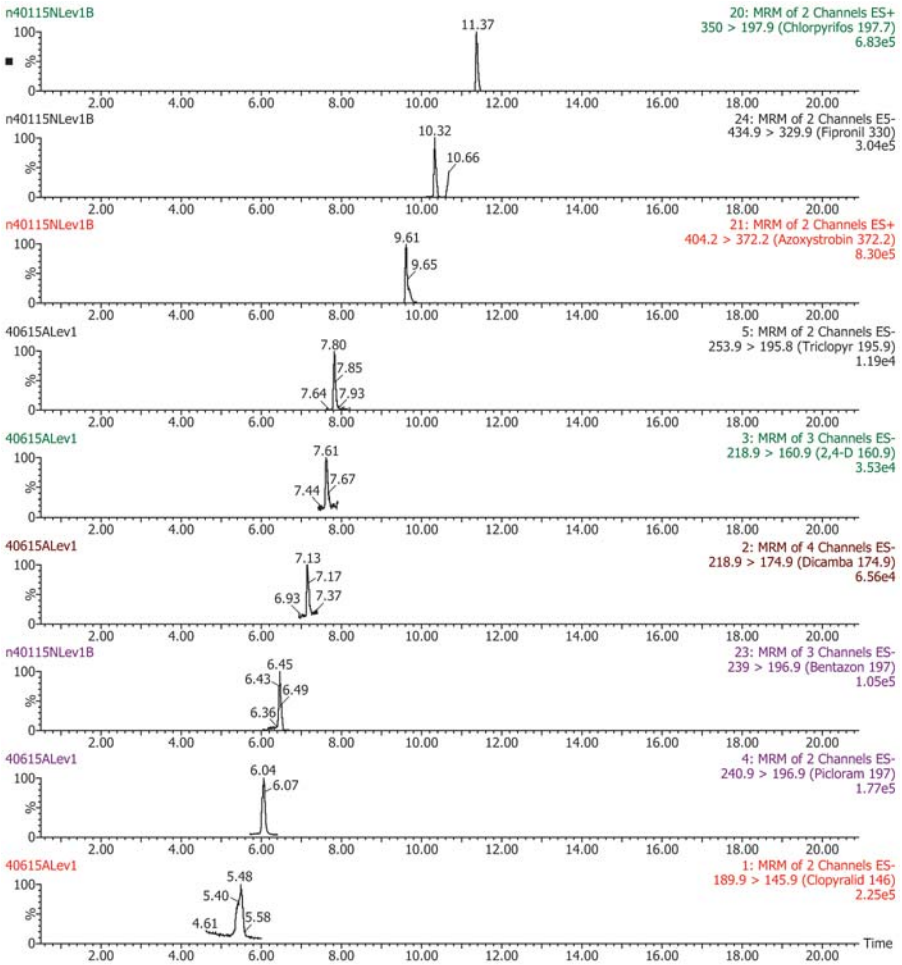

FIG. X1.3 The Primary SRM Transition Chromatograms at the Level 1 Calibration Concentration for Chlorpyrifos, Fipronil, Azoxystrobin, Triclopyr, 2,4-D, Dicamba, Bentazon, Picloram and Clopyralid

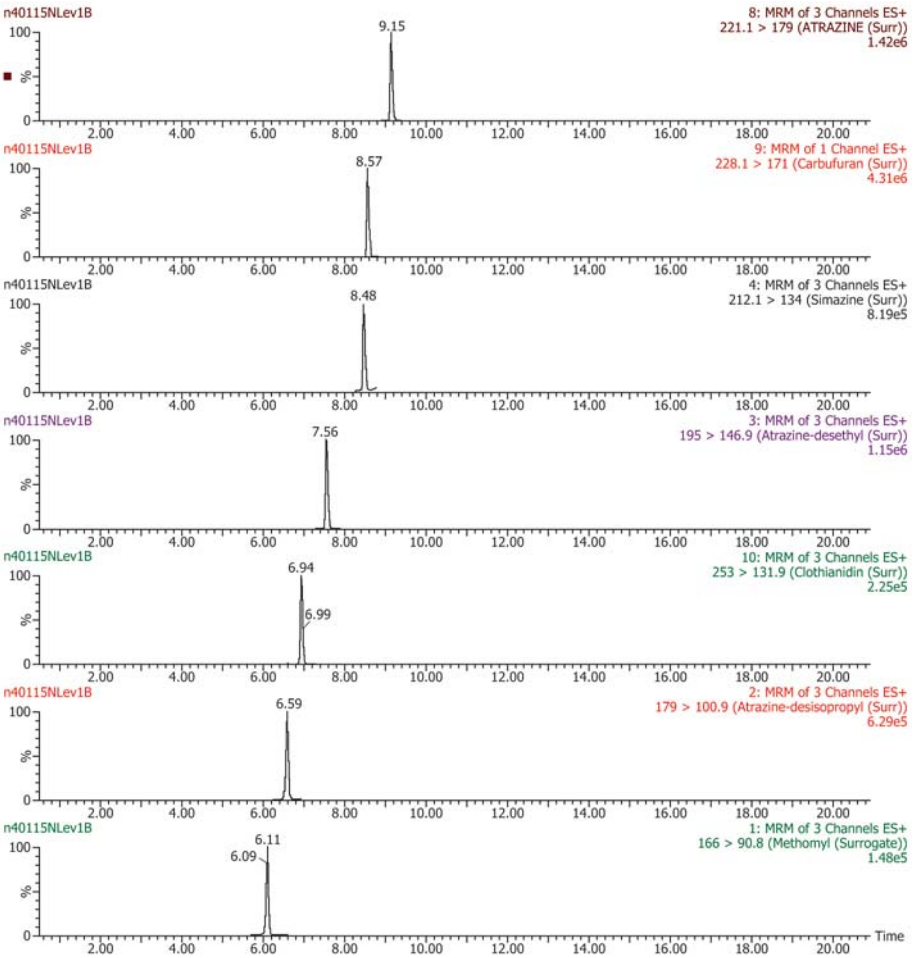

FIG. X1.4 The Primary SRM Transition Chromatograms at the Level 1 Calibration Concentration for the Isotopically Labeled Surrogates of Atrazine, Carbofuran, Simazine, Atrazine-desethyl, Clothianidin, Atrazine-desisopropyl and Methomyl

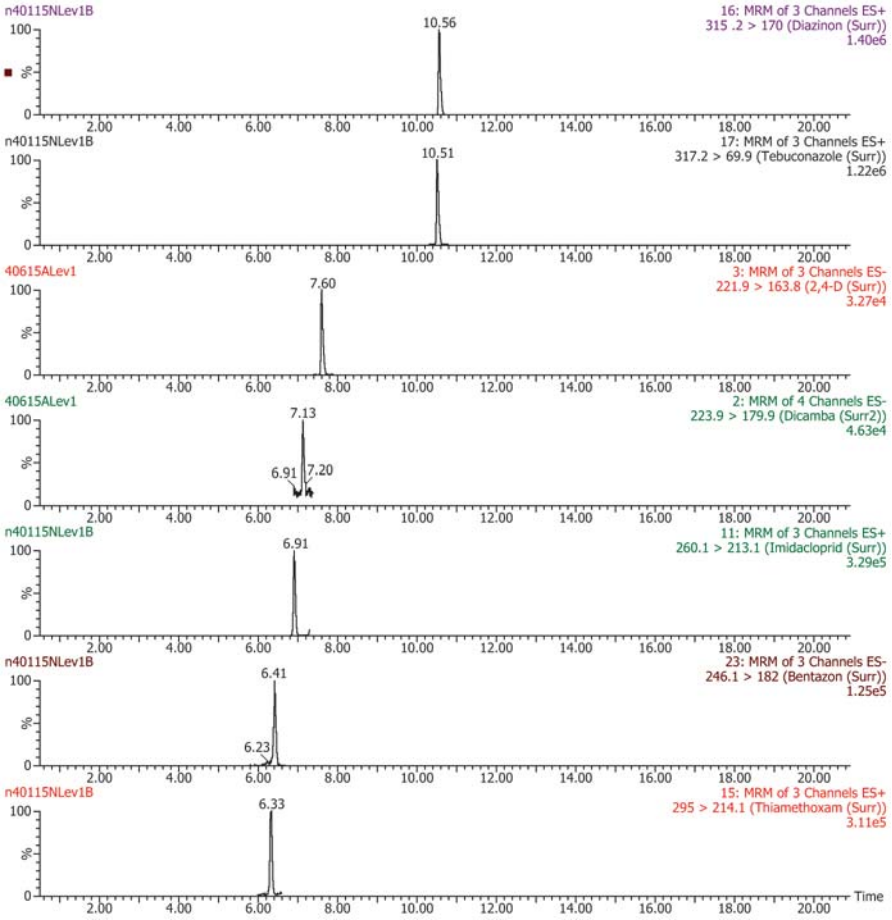

FIG. X1.5 The Primary SRM Transition Chromatograms at the Level 1 Calibration Concentration for the Isotopically Labeled Surrogates of Diazinon, Tebuconazole, 2,4-D, Dicamba, Imidacloprid, Bentazon and Thiamethoxam