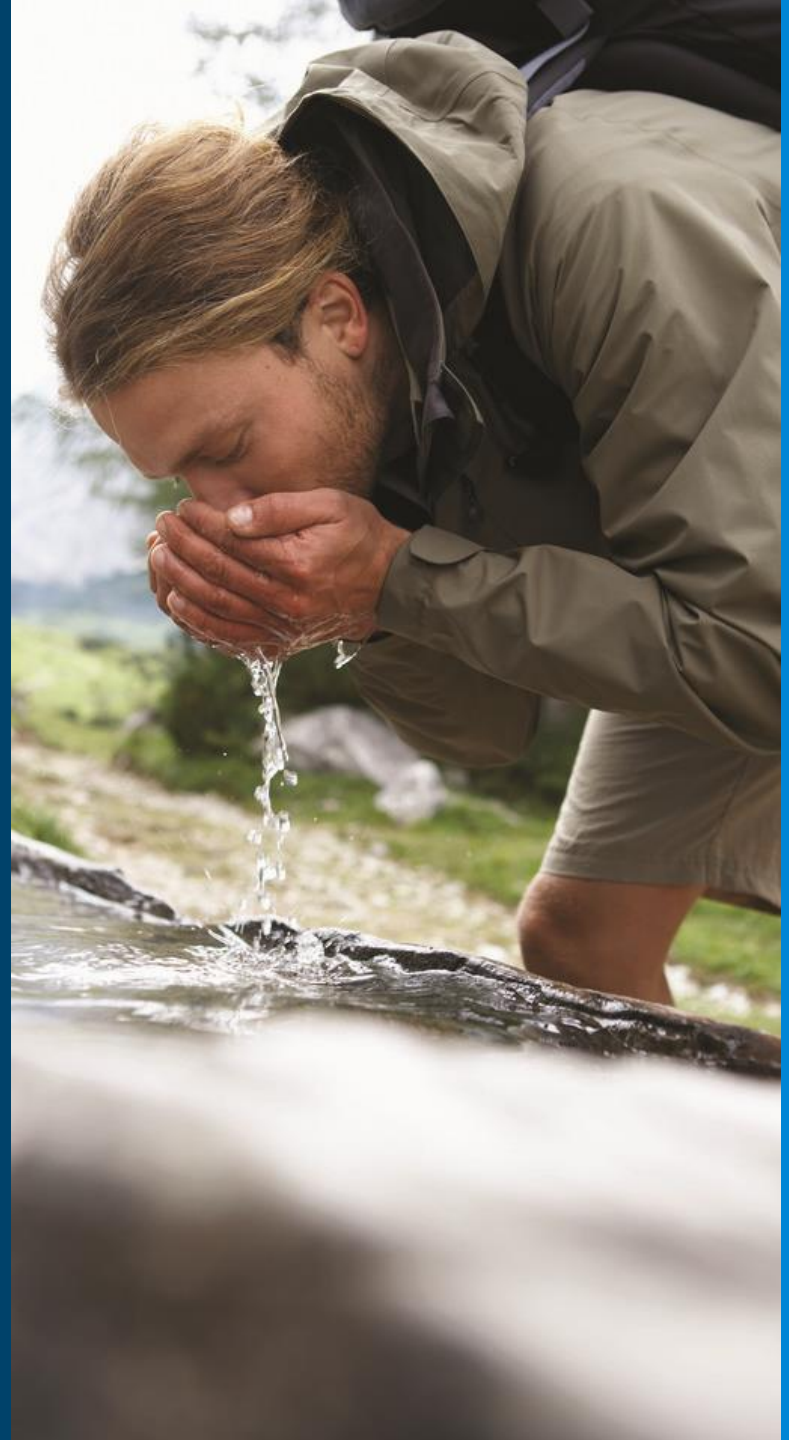
From Legacy to Emerging Pollutants: Mass Spectrometry Solutions to Analyze Dioxins & PFAS in Water

Agilent Lunch Seminar @ NEMC 2021

Tarun Anumol, Ph.D.
Director, Global Environment & Food Markets
Agilent Technologies Inc.

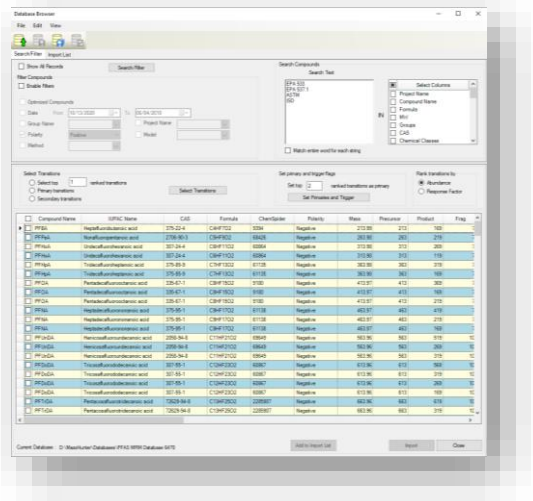


Agilent
Trusted Answers

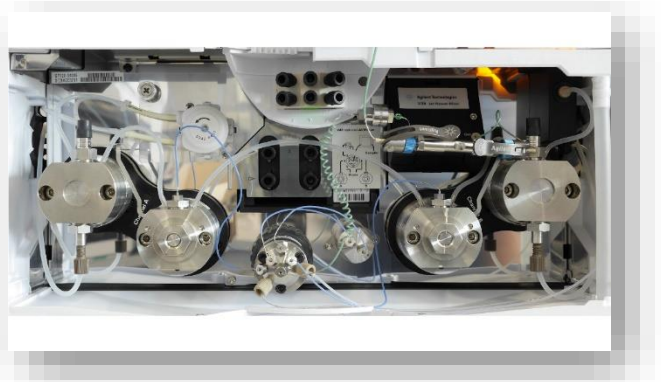
PFAS analysis needs many aspects to Meet Various Laboratory Needs

Individual Products and Services for Regulatory and Individual Method Development

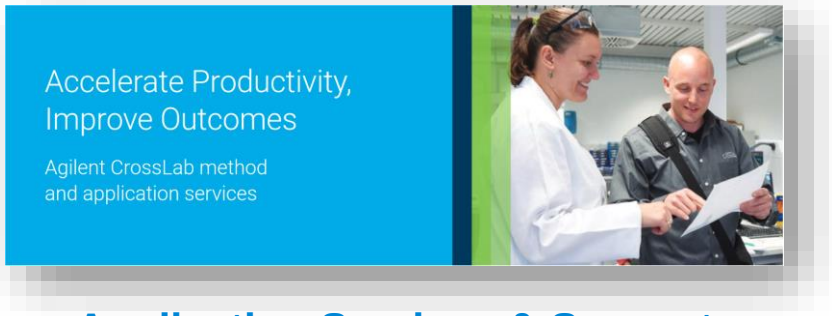
LC-MS/MS instrumentation



PFAS MRM Database



PFC-Free HPLC Conversion Kit



Application Services & Support

PFC-Free Columns and Supplies

www.agilent.com/chem/pfas

PFAS Easy Ordering Guide (5994-2357)

for EPA 537, 533, 8327, ASTM D7979, ISO 21675:2019 and MRM acquisition method

Agilent InfinityLab

Consumables Ordering Guide

Per- and polyfluoroalkyl substances (PFAS) analysis

PFAS have been used in industry and consumer products since the 1940s. Their chain of strong fluorine-carbon bonds makes these chemicals persistent and bio-accumulative over long-term exposure—linking them to health hazards in humans and wildlife.* What's more, PFAS have been widely detected in drinking water, wastewater, ground and surface water, soil, and other complex matrices.

In response, regulatory agencies worldwide are implementing more stringent requirements for monitoring and identifying PFAS. For example:

- The EU water framework directive lists PFOS (a subclass of PFAS) as priority hazardous substances that pose a "significant risk to the aquatic environment."
- The USEPA has established a drinking water advisory level, and has recently announced more stringent standards. Further, many states in the U.S. have regulatory limits for PFAS in water at low ng/L levels, and many countries in Asia including Japan, South Korea, and Australia have guidelines for monitoring PFAS in water too.

You can achieve uncompromising accuracy for rigorous regulatory methods with Agilent PFAS analysis solutions. Our InfinityLab PFC-free HPLC conversion kit includes everything you need to ensure your 1290 Infinity II instruments and 1290 Infinity II high-speed pump are free of PFAS contaminants:

- Tubing
- Inline filter
- Wash bottle head assembly
- Delay column with InfinityLab Quick Connect LC fitting

Agilent's PFAS solution includes products for sample preparation, HPLC columns and solvents as well as PFC-free sample containment and other HPLC supplies you would need to ensure reproducible results with highest quality while eliminating PFAS contamination and reduced the risk of false positive results.

*L. Ahrens, M. Bundschuh. Fate and effects of poly- and perfluoroalkyl substances in the aquatic environment: A review. Environ. Toxicol. Chem., 2014, 33, 1921–1929.

Easy Selection and Ordering Information

This guide provides recommendations for Agilent products by regulatory method, so you can find what you're looking for quickly. To add items to your "My Favorites"* list at the Agilent online store, simply click the MyList links in each header below. Then, enter the quantities for the products you need. Your list will remain under "My Favorites" for your use with future orders.

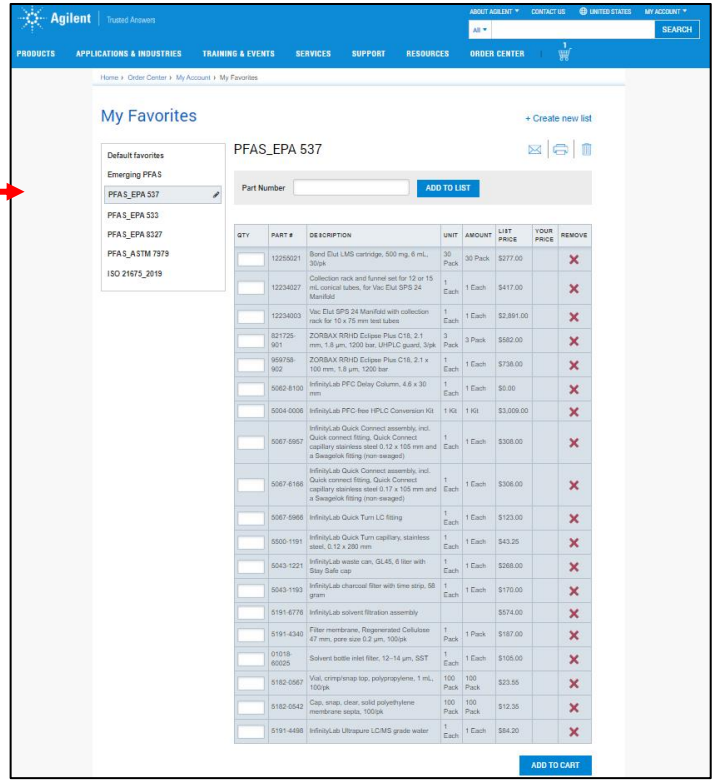
EPA 537 or similar: Determination of PFAS in drinking water by SPE and LC/MS/MS (EPA 537.1 or similar)

View MyList

Product Category	Description	Part Number
Sample preparation	Bond Elut LMS cartridge, 500 mg, 6 mL, 30/pk	12255021
	Collection rack and funnel set for 12 or 15 mL conical tubes, for Vac Elut SPS 24 manifold	12234027
	Vac Elut SPS 24 manifold with collection rack for 10 x 75 mm test tubes	12234003
Guard column	ZORBAX RRHD Eclipse Plus C18 2.1 x 5 mm, 1.8 µm guard	821725-901
Separation column	ZORBAX RRHD Eclipse Plus C18, 2.1 x 100 mm, 1.8 µm	959758-902
PFC-free HPLC conversion kit w/delay column	Agilent InfinityLab PFC-free HPLC conversion kit**	5004-0006
Delay column	InfinityLab PFC Delay Column, 4.6 x 30 mm (replacement)	5062-8100
Other LC supplies	InfinityLab Quick Connect assembly, 0.12 x 105 mm, for column inlet connection on UHPLC	5067-5957
	InfinityLab Quick Connect assembly, 0.17 x 105 mm, for column inlet connection on UHPLC	5067-6166
	InfinityLab Quick Turn fitting, for column outlet	5067-5966
	Quick Turn capillary 0.12 x 280 mm, for connecting column to detector	5500-1191
	Kit of Stay Safe waste caps GL45 with 4 ports and waste can (6 L)	5043-1221
	Charcoal filter with time strip for waste container	5043-1193
	InfinityLab solvent filtration assembly: includes glass funnel (250 mL), membrane holder glass base, glass flask (1 L), and aluminum clamp	5191-6776
	Regenerated cellulose filter membrane (47 mm, 0.20 µm, 100/pk)	5191-4340
	Stainless steel solvent inlet filter, 12–14 µm pore size	01018-60025
Sample containment	Clear snap caps with polyethylene membrane septa (100/pk)	5182-0542
	Polypropylene vials (no caps) 100/pk	5182-0567
Solvents	InfinityLab ultrapure LC/MS water	5191-4498

*First time using "My Favorites"? You will be asked to enter your email address for account verification. If you have an existing Agilent account, you will be able to log in. If you don't have a registered Agilent account, you will need to register for one. This feature is valid only in regions that are e-commerce enabled. All items can also be ordered through your regular sales and distributor channels.

**Although the kit is customer-installable, Agilent offers supplemental installation by a service professional. To add this service to your order, use part number H5949A.



Promotion Code: 1913

Buy 10 or more items from this ordering guide to receive 25% discount

If this is your first time using the Agilent Online Store, you will be asked to enter your email address for account verification. If you don't have a registered Agilent account, you will need to **register for one. The "My List" feature is valid only in regions that are e-commerce enabled. All items can also be ordered through your regular sales and distributor channels.*

Not available in all countries. Please contact your local sales representative for availability.

Sampling Handling & Storage Advances
‘PFAS’ specific vials and caps

- PTFE lining in Caps can have PFAS contamination
- This leads to use of PP style snap top vials that have very poor sealability on piercing or longer-term storage with organic solvent
- This can result in use of 2 or more caps per sample

- Glass vials are thought to adsorb certain PFAS, hence PP vials are preferred

Unique bi-layer of PP and silicone that allows resealing once pierced

No PTFE and free of 26 measured PFAS

1.7 mL fill volume and standard screw top

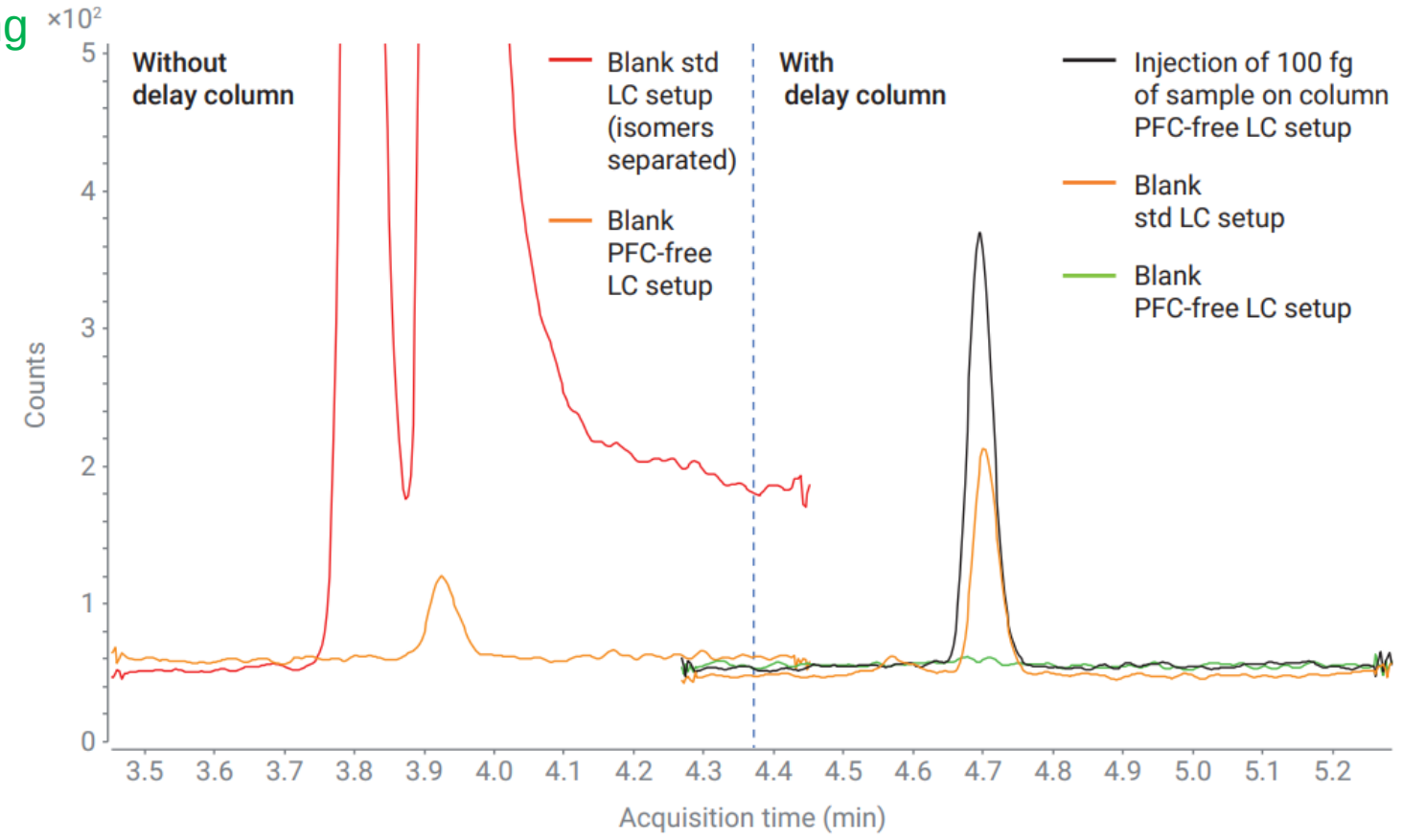
Description	Part number
2 mL screw style clear polypropylene vial (100pk)	5191-8150
9 mm screw style clear polypropylene cap with thin membrane polypropylene / silicone septa (100pk)	5191-8151

PFC Free Kit
Eliminate Background Contamination

Potential Contamination Sources

- Solvents
- Filtration apparatus
- Teflon lined tubing

LC Configuration	PFHpA Background (fg)	PFNA Background (fg)
Standard LC Setup	>3,000	>500
Standard LC Setup With Delay Column	48	48
PFC-free LC Setup	20	37
PFC-free LC Setup With Delay Column	<2 (below detection limit)	<7 (below detection limit)



(P/n: 5004-0006)

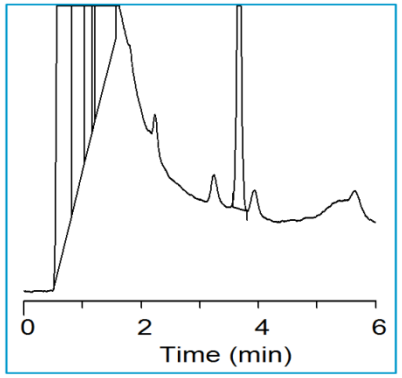
Sample Preparation

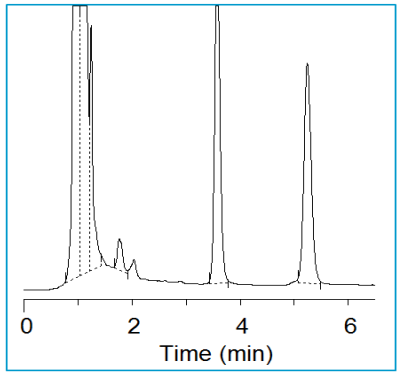
Sample preparation refers to the ways in which samples being treated prior to their analysis. Target analytes are the needle in the haystack of matrix, sample prep helps find the needle in the haystack.

Sample Prep

Sample *without* Sample Prep

Sample with Sample Prep





Analytical choices for PFAS Quantification
LC-MS/MS still seen as gold standard for quantification

LESS SAMPLE PREP

SENSITIVITY

Direct injection

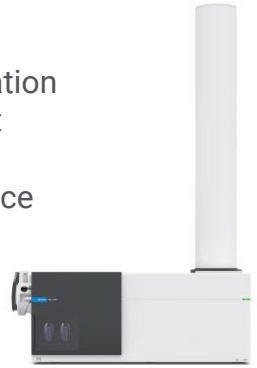
6470 LC/MS/MS
- ✓ Robustness
- ✓ Enhanced sensitivity for Environmental work
- ✓ Gate valve for no-vent capillary cleaning

‘Dilute & shoot’
Large volume injection

6495 LC/MS/MS
- ✓ Ultimate Sensitivity
- ✓ Gate valve for no-vent capillary cleaning

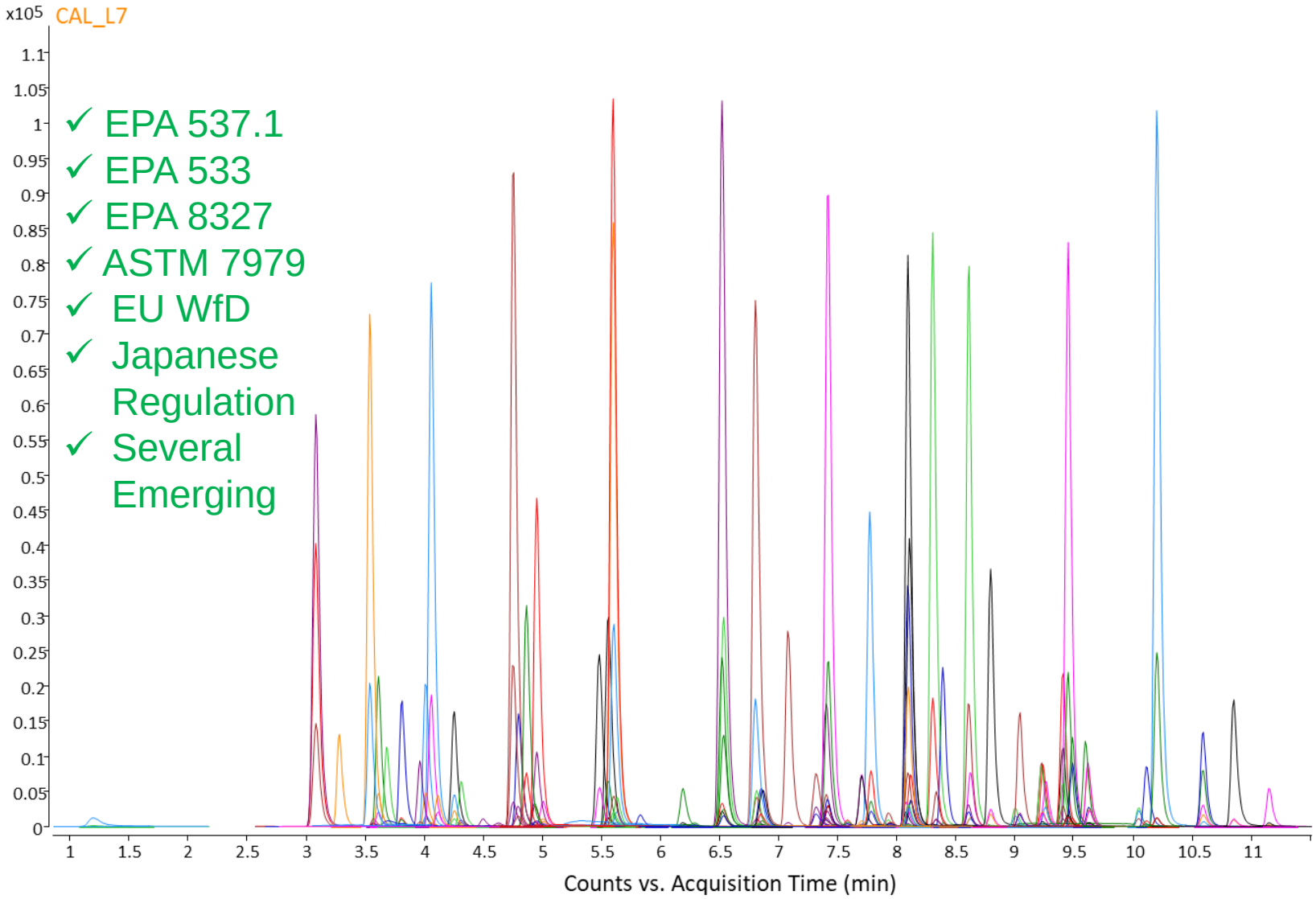
Sample enrichment suggested

ULTIVO
- ✓ Routine Quantification
- ✓ Smallest Footprint LC/MS/MS
- ✓ Guided maintenance



6546 LC-Q/TOF
- ✓ High resolution accurate mass
- ✓ Suspect screening and unknown PFAS

Comprehensive Database & Method – Over 100 PFAS Compounds



- ✓ EPA 537.1
- ✓ EPA 533
- ✓ EPA 8327
- ✓ ASTM 7979
- ✓ EU WfD
- ✓ Japanese Regulation
- ✓ Several Emerging

Native Analytes:

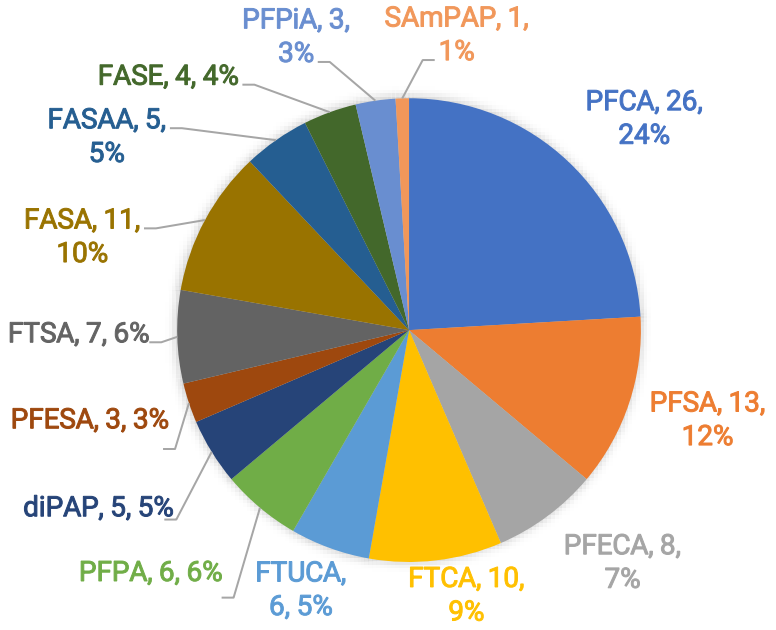
- 71 native PFAS analytes

Surrogates:

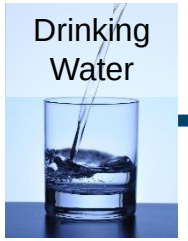
- 33 isotopically labeled PFAS
- 1 native, Cl-PFOPA is used as ISTD for PFPAs

Isotope performance standards:

- 3 isotopically labeled PFAS



PFAS eMethod Workflow overview



250 mL per sample

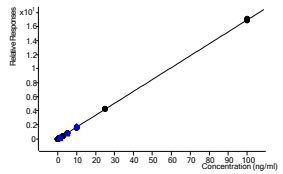
+ SUR

Solid phase extraction (SPE) using SampliQ Weak Anion Exchange (WAX), 6mL, 150 mg

+ IPS

SPE extracts (250x concentration)

Calibration standards (CAL) in 80:20/MeOH:H_2O



Agilent PFC-Free HPLC Conversion Kit

- Agilent 1290 Infinity II UHPLC
- Agilent 6470 LC/TQ

Method assessed on:
- CAL analytical range
- Method sensitivity
- Method recovery
- Method precision

Agilent PFAS Solutions

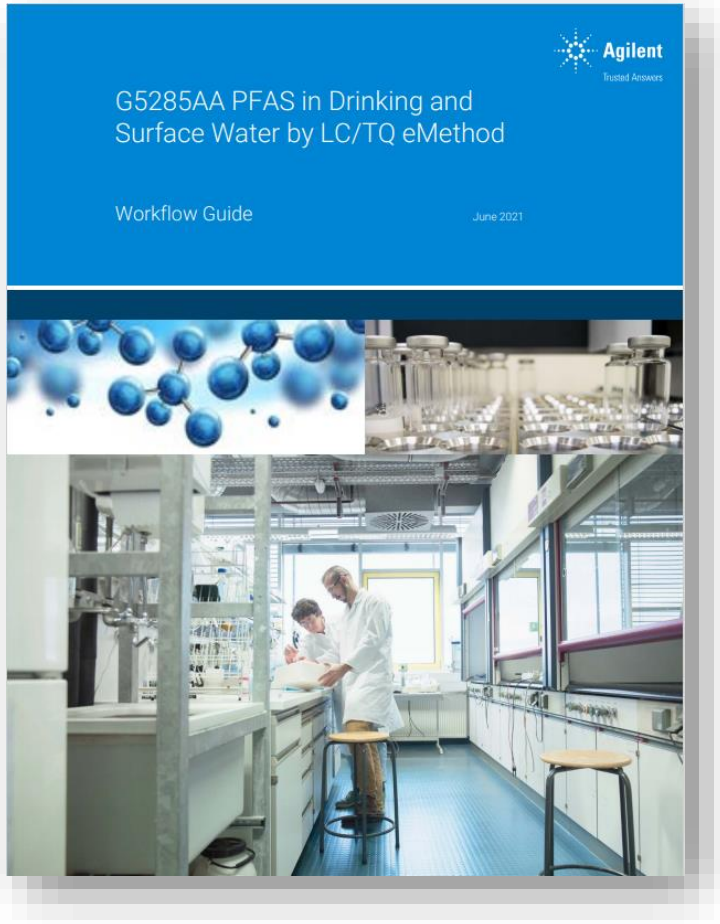
End-to-end Verified Workflow: Turn-key Solution Ready for Immediate Use

PFAS Drinking & Surface Water eMethod

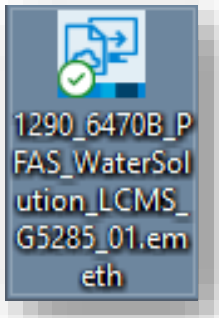
An end-to end, verified solution for the analysis of >100 native & isotopically labeled PFAS in **drinking water** and **surface water** without extensive method development or technical investigation

eMethod Includes:

- Full analysis protocol, from sample prep through reporting
- Optimized MassHunter Acquisition and Quant methods
- Best practices
- Sample preparation training video
- Example calibration data
- Comprehensive ordering information with part number details

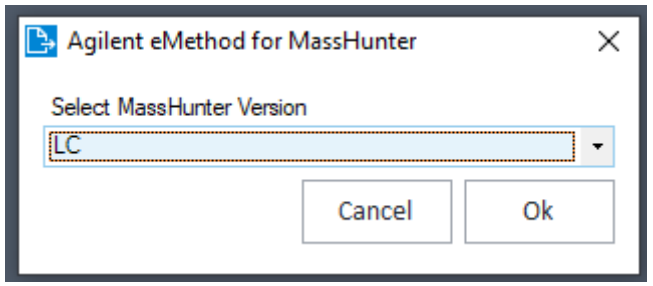


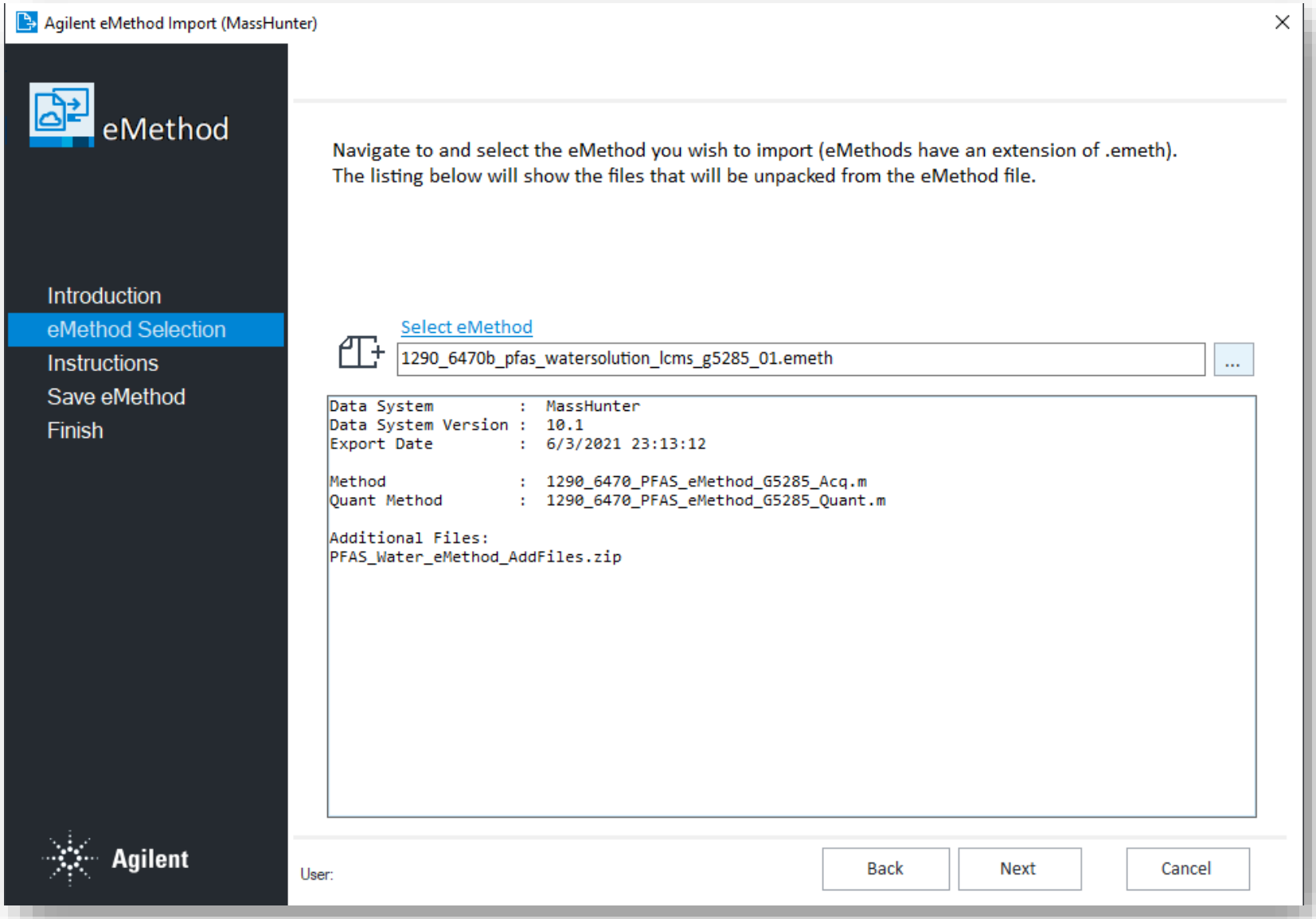
Compatible with1290 + 6470 LC/TQ



1. Double click eMethod icon

Note – Agilent internal version requires selection of LC or GC





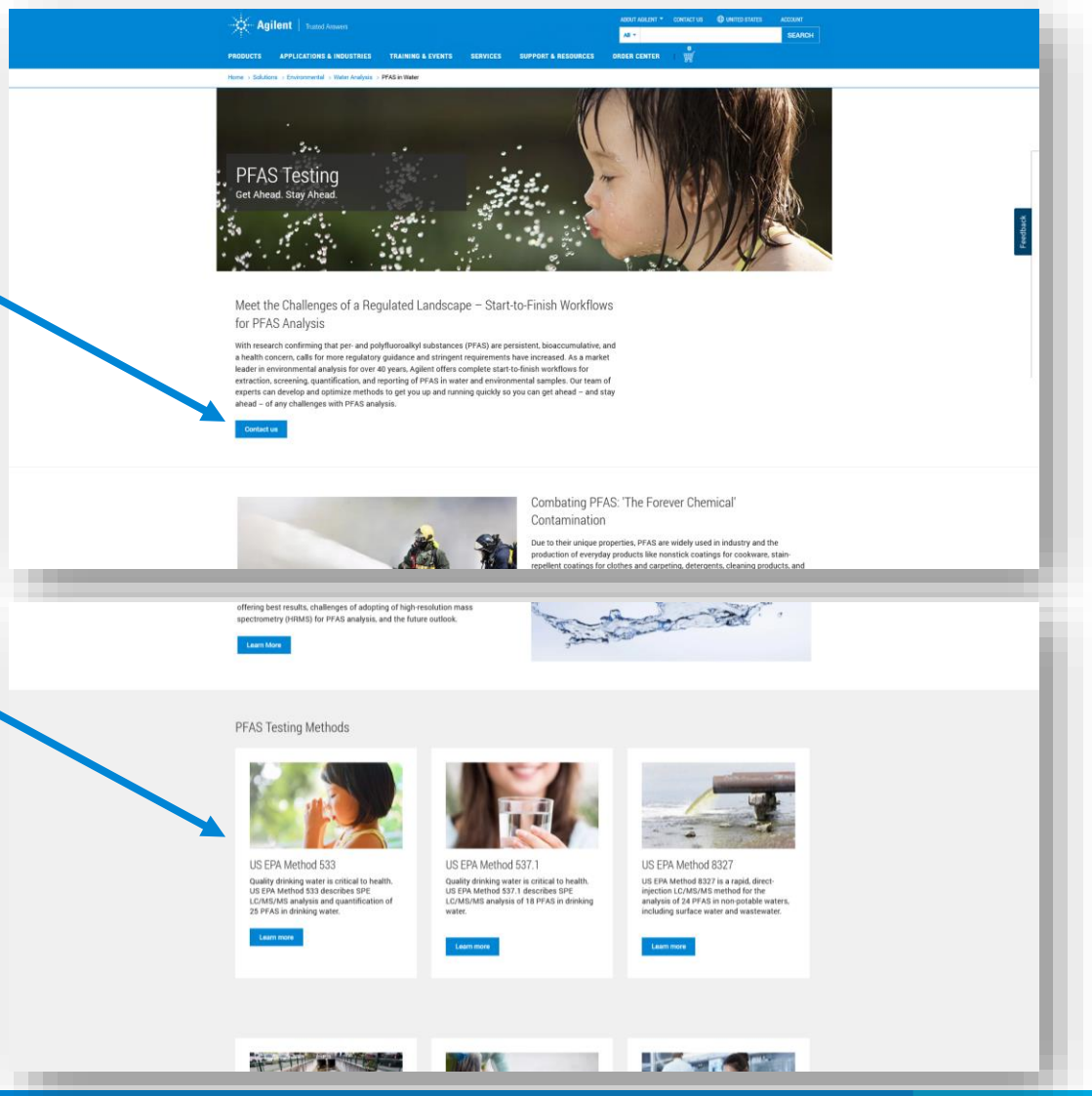
2. Review eMethod detail, click Next

LATEST PFAS TESTING INFORMATION

A one-stop for all PFAS info on regulatory and emerging methods

Links to informational content

Links to individual method details



https://www.agilent.com/en/solutions/environmental/water-analysis/pfas-in-water

Link to PFAS A&I page

Agilent PFAS Talks at NEMC

Watch all talks ON-DEMAND at NEMC

SAMPLE PREPARATION

The Importance of Sorbent Mass to Sample Volume for the Extraction of PFAS from Drinking Water Using Weak Anion Exchange SPE – Matthew Giardina, Agilent Technologies Inc.

TARGETED QUANTIFICATION

Targeted Quantitation of Legacy and Emerging Per- and Polyfluoroalkyl Substances (PFAS) in Water Matrices – Tarun Anumol, Agilent Technologies Inc.

NON-TARGET; HRMS QUANTIFICATION

Accurate Mass QToF - A New Direction in Quantitative PFAS Analysis – Kathy Hunt, Vogon Laboratories

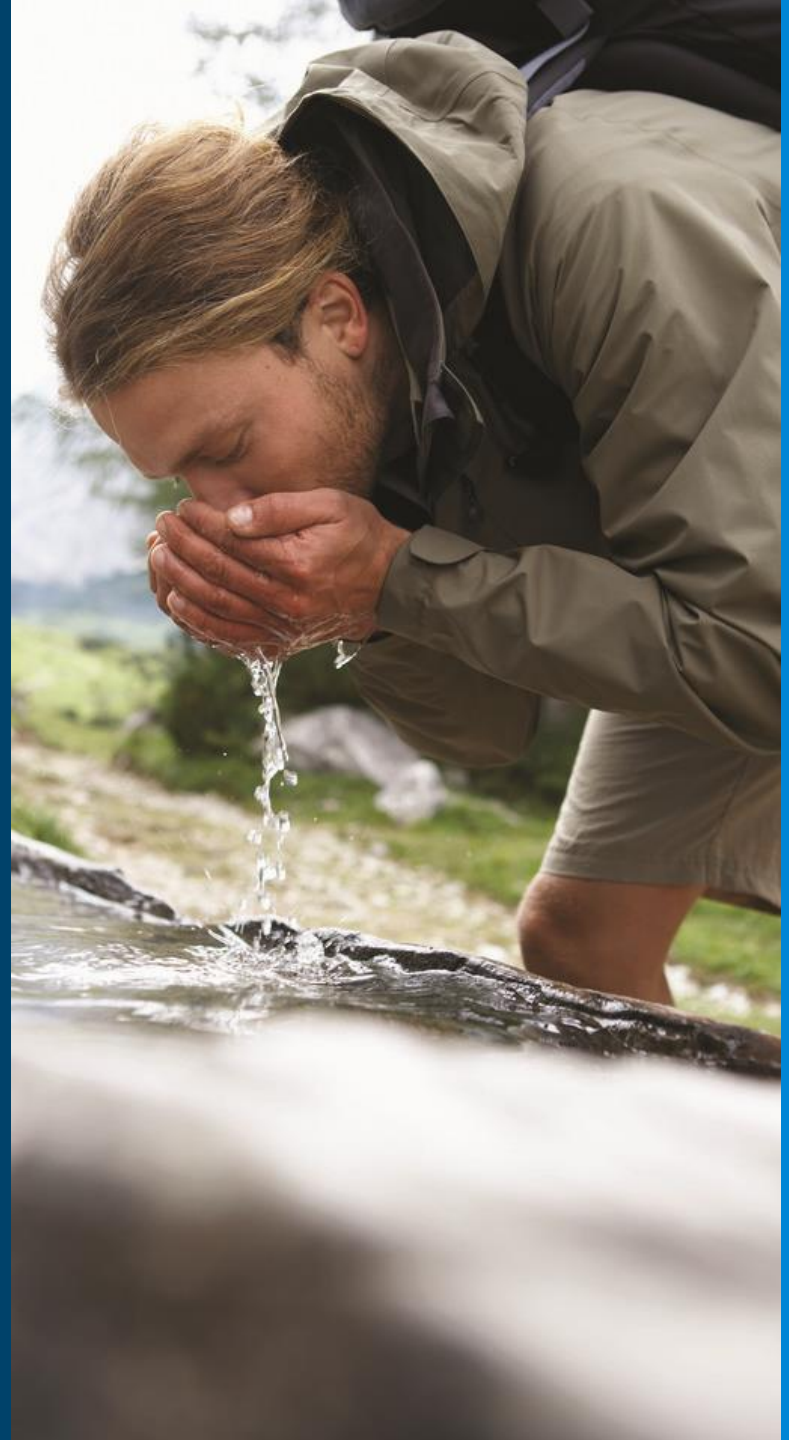
Agilent | Trusted Answers

An Alternate Testing Protocol for EPA 1613B using Agilent Triple Quadrupole GC/MS

Dale R. Walker
GC/MS/MS Application Scientist

Tarun Anumol, Ph.D.
Director, Global Environment & Food Markets

Anastasia Andrianova
GC/MS/MS Application Scientist



Agilent
Trusted Answers

Application Note

Agilent
Trusted Answers

An Alternate Testing Protocol for EPA 1613B using Agilent Triple Quadrupole GC/MS

Determination of 2,3,7,8-substituted tetra- through octa-chlorinated dibenzo-*p*-dioxins and dibenzofurans

Authors

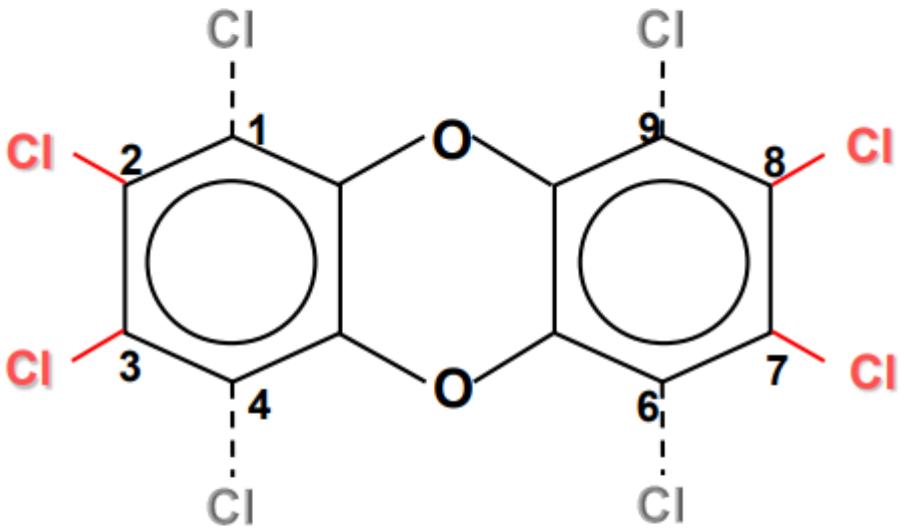
Coreen Hamilton and
Xinhui Xie,
SGS AXYS Analytical Services Ltd.

Tarun Anumol,
Anastasia Andrianova, and
Dale Walker,
Agilent Technologies, Inc.

Abstract

This study provides data used to create an alternate testing protocol for the U.S. Environmental Protection Agency (EPA) to use for Agilent 7010B Triple Quadrupole GC/MS analysis of tetra- through octa-dioxins and furans that is equivalent to EPA Method 1613B. EPA Method 1613B is used for the determination of the 17 toxic tetra- through octa-chlorinated Dibenzo-*p*-Dioxins and Dibenzofurans (CDDs/CDFs) in aqueous, solid, and tissue matrices by isotope dilution gas chromatography/high-resolution mass spectrometry (GC/HRMS) using magnetic sector instruments. Traditionally used for dioxins analysis because of their high sensitivity, GC/HRMS instruments are expensive to maintain, require a highly specialized skill set to operate, and are being phased out by manufacturers. However, current GC/MS/MS (GC/TQ) technology provides many of the specificity and sensitivity advantages of HRMS for the analysis of regulated dioxins and furans, without the cost and complexity, and with added versatility and robustness. This application note describes a method developed in collaboration with SGS AXYS Analytical Services Ltd., SGS AXYS Method 16130, that uses the Agilent 7890B gas chromatograph coupled with an Agilent 7010B Triple Quadrupole GC/MS. Performance factors investigated included sensitivity, linearity, method detection limits (MDLs), recovery, and results compared to reference material. The GC/TQ results met the QA/QC and performance specifications described in Method 1613B for the analysis of polychlorinated dioxins and furans (PCDDs/PCDFs) in environmental matrices. Overall, the GC/TQ method produced accurate data for real-world sample matrices, offering a lower cost, more efficient alternative to GC/HRMS.

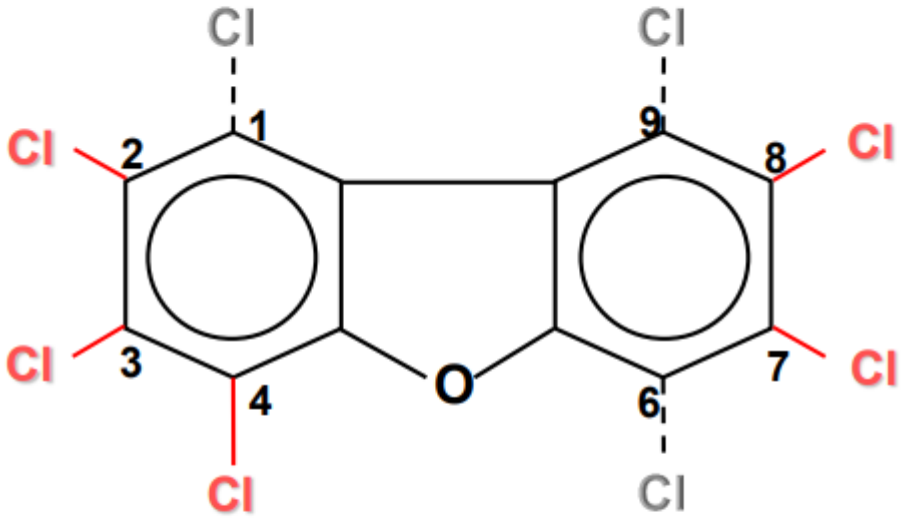
Dioxins (PCDD/Fs)



Dioxins (PCDD)

Total of 75

(2378-TetraCDD)



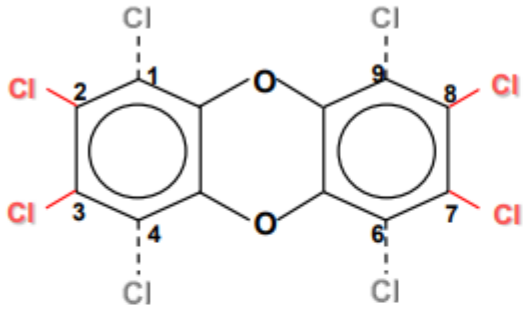
Furans (PCDF)

Total of 135

(23478-PentaCDF)

(17 toxic dioxin congeners)

Numbers of dioxin compounds isomers with different chlorine substituents

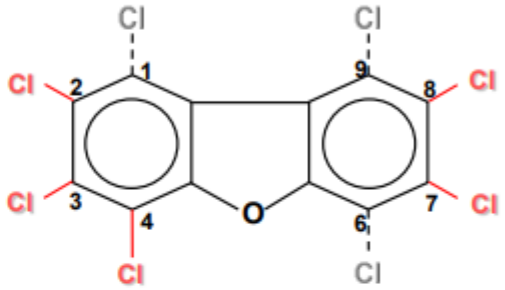


Dioxins (PCDD)

Total of 75

(2378-TetraCDD)

Number of chlorine atoms	Number of PCDD isomers	Number of PCDF isomers
1	2	4
2	10	16
3	14	28
4	22	38
5	14	28
6	10	16
7	2	4
8	1	1
Total	75	135



Furans (PCDF)

Total of 135

(23478-PentaCDF)

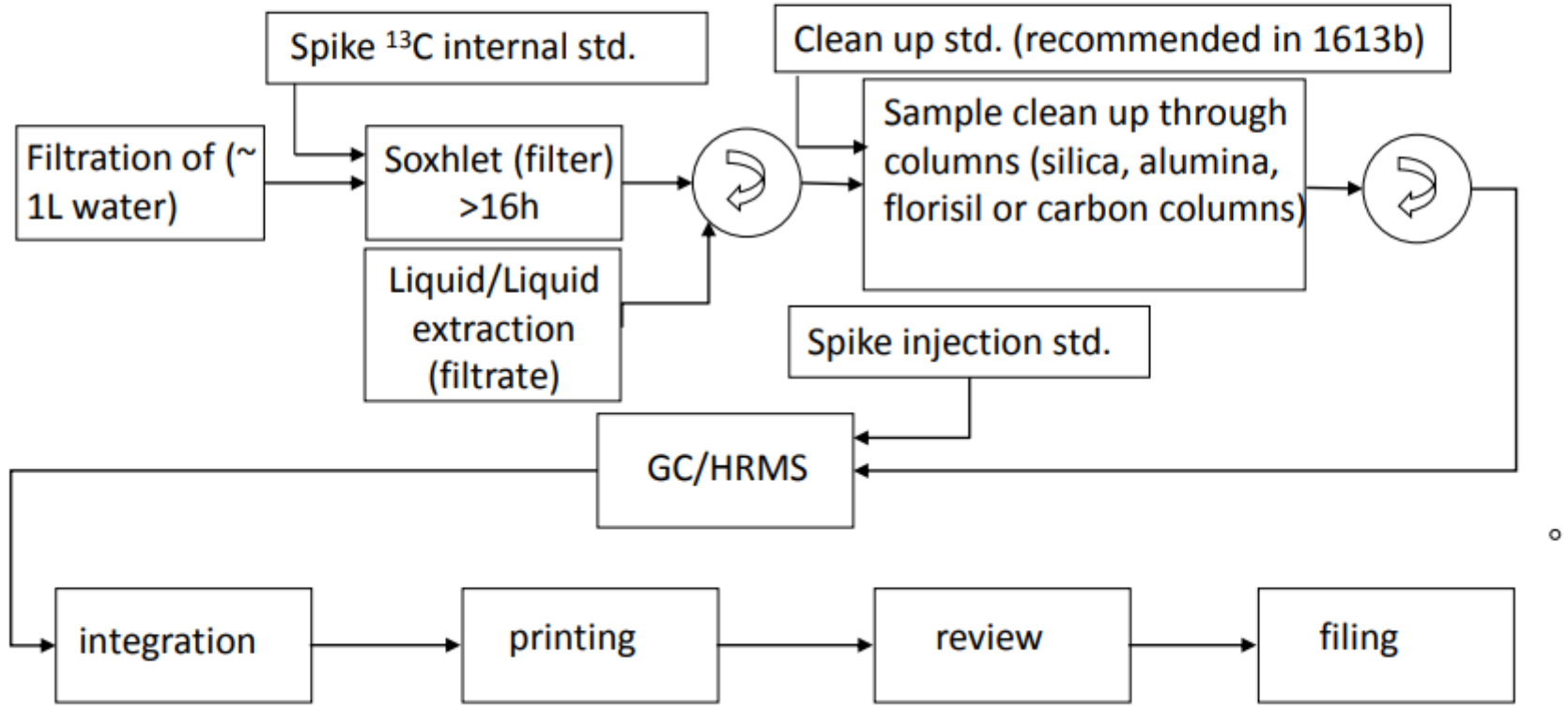
Toxic Equivalent Factors (TEF)

Toxic PCDDs	I-TEF	WHO_{2005}-TEF	Toxic PCDFs	I-TEF	WHO_{2005}-TEF
2378-TetraCDD	1	1	2378-TetraCDF	0.1	0.1
12378-PentaCDD	0.5	**1**	12378-PentaCDF	0.05	**0.03**
123478-HexaCDD	0.1	0.1	23478-PentaCDF	0.5	**0.3**
123678-HexaCDD	0.1	0.1	123478-HexaCDF	0.1	0.1
123789-HexaCDD	0.1	0.1	123678-HexaCDF	0.1	0.1
1234678-HeptaCDD	0.01	0.01	123789-HexaCDF	0.1	0.1
12346789-OctaCDD	0.001	**0.0003**	234678-HexaCDF	0.1	0.1
			1234678-HeptaCDF	0.01	0.01
			1234789-HeptaCDF	0.01	0.01
			12346789-OctaCDF	0.001	**0.0003**

$$TEQ = \sum_{n=1}^{17}([PCDD/F]_i(^{ng}/_L) \times TEF_i)\ (^{ng\ TEQ}/_L)$$

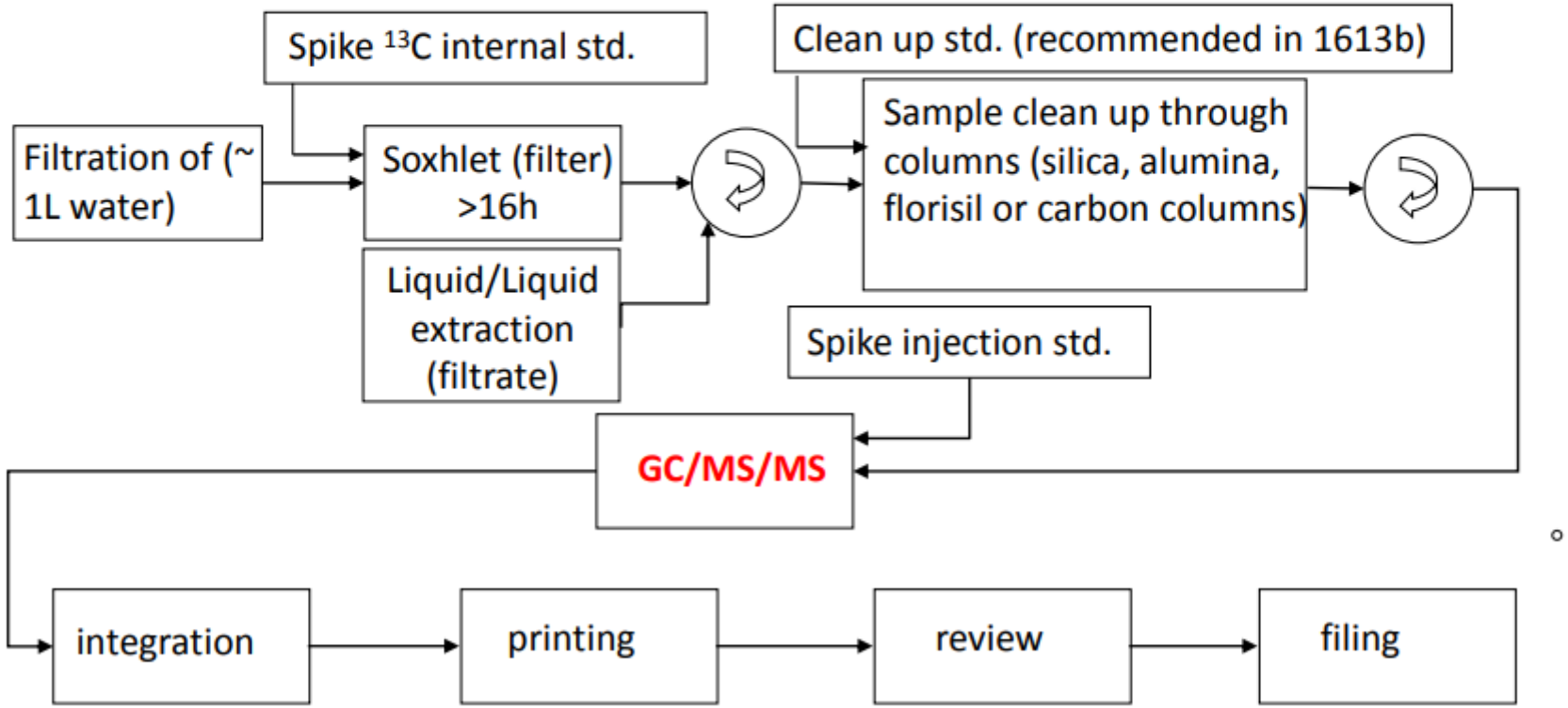
Analytical Strategies (EPA method 1613b):

Note: EPA 1613b is performance-based method



Analytical Strategies (EPA method 1613b):

Note: EPA 1613b is performance-based method



System Verification

Triple Quadrupole GC/MS System Verification - Tune

Instrument Name	8890-7000D / US2106T303	**MS Model**	7000D
Tune Date & Time	7/22/2021 8:56:47 AM	**Source**	EI High Efficiency
Tune File	D:\MassHunter\GCMS\1\7000\atunes.elex_300.tune.xml Modified		

Instrument Actuals		**Vacuum**	
Ionization mode	EI+	Rough Vac	1.03E+2 mTorr
Source Temp.	301 °C	High Vac	8.80E-5 Torr
MS1 Quad Temp.	150 °C	Turbo 1 Speed	100.0 %
MS2 Quad Temp.	150 °C	Turbo 1 Power	19.4 W
Filament Current	100.0 µA		
GC Gas Flow			
Quench Flow	2.250 mL/min	Column 1	1.337 mL/min
Collision Cell	1.500 mL/min	Column 2	0.000 mL/min

MS1 Checktune Results	Value	Limit	Result
Low mass assignment (target 69.00, actual 69.00)	0.00	<= 0.20	OK
Mid mass assignment (target 264.00, actual 263.90)	0.10	<= 0.20	OK
High mass assignment (target 502.00, actual 502.00)	0.00	<= 0.20	OK
Low mass isotope position (target 70.00, actual 70.07)	0.07	<= 0.20	OK
Mid mass isotope position (target 265.00, actual 264.95)	0.05	<= 0.20	OK
High mass isotope position (target 503.00, actual 503.00)	0.00	<= 0.20	OK
Low mass isotope ratio	1.15%	>= 0.5% and <= 1.6%	OK
Mid mass isotope ratio	5.60%	>= 4.2% and <= 6.9%	OK
High mass isotope ratio	10.67%	>= 7.9% and <= 12.3%	OK
Ratio of mid mass to low mass	10.14%	>= 5.0%	OK
Ratio of high mass to low mass	0.80%	>= 0.8%	---
Low mass precursor ratio	0.55%	<= 3.00%	OK
Mid mass precursor ratio	0.00%	<= 6.00%	OK
High mass precursor ratio	1.58%	<= 12.00%	OK
MS2 Checktune Results			
Low mass assignment (target 69.00, actual 68.90)	0.10	<= 0.20	OK
Mid mass assignment (target 264.00, actual 264.00)	0.00	<= 0.20	OK
High mass assignment (target 502.00, actual 501.95)	0.05	<= 0.20	OK
Low mass isotope position (target 70.00, actual 70.00)	0.00	<= 0.20	OK
Mid mass isotope position (target 265.00, actual 265.00)	0.00	<= 0.20	OK
High mass isotope position (target 503.00, actual 503.00)	0.00	<= 0.20	OK
Low mass isotope ratio	1.19%	>= 0.5% and <= 1.6%	OK
Mid mass isotope ratio	5.73%	>= 4.2% and <= 6.9%	OK
High mass isotope ratio	9.62%	>= 7.9% and <= 12.3%	OK
Low mass precursor ratio	0.23%	<= 3.00%	OK
Mid mass precursor ratio	0.13%	<= 6.00%	OK
High mass precursor ratio	0.13%	<= 12.00%	OK
Detector			
EMV	1010	<= 2900	OK
Maximum gain factor	777383	>= 100	OK

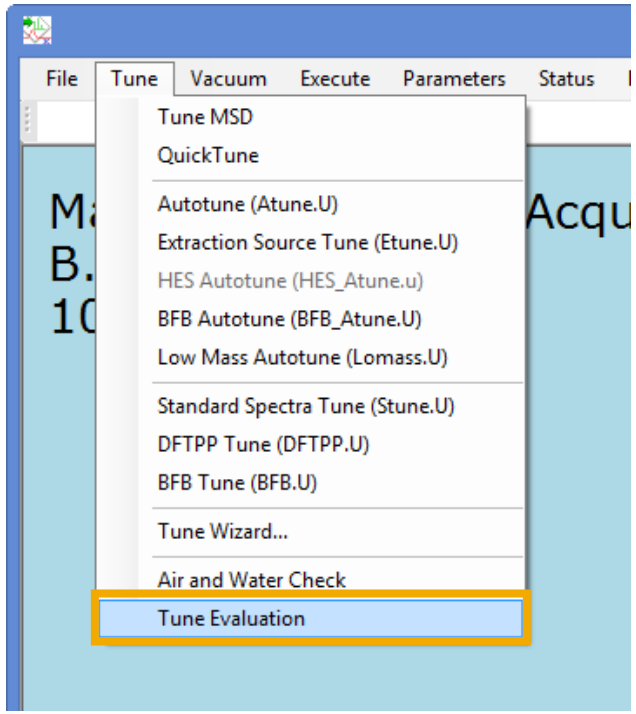
Triple Quadrupole GC/MS System Verification - Tune

Instrument Name	8890-7000D / US2106T303	**MS Model**	7000D
Tune Date & Time	7/22/2021 8:56:47 AM	**Source**	EI High Efficiency
Tune File	D:\MassHunter\GCMS\1\7000\atunes.elex_300.tune.xml Modified		

Air and Water Check	Abundance	Relative Abundance	Limit	Result
PFTBA (69.00)	2884306			
Water	660869	22.91%	<= 20.00%	---
Oxygen	37260	1.29%	<= 2.50%	OK
Nitrogen	138868	4.81%	<= 10.00%	OK

* Nitrogen values are calculated from oxygen abundance

System Verification



System Verification - Tune (Detector Optimization) Portion

Instrument Name	: MH 5977	
DC Polarity	: Positive	
Filament	1	
BasePeak should be 69 or 219		OK
Position of mass 69	69.00	OK
Position of mass 219	219.00	OK
Position of mass 502	502.00	OK
Position of isotope mass 70	70.03	OK
Position of isotope mass 220	220.00	OK
Position of isotope mass 503	503.01	OK
Ratio of mass 70 to mass 69(0.5 - 1.6%)	1.09	OK
Ratio of mass 220 to mass 219(3.2 - 5.4%)	4.32	OK
Ratio of mass 503 to mass 502(7.9 - 12.3%)	10.04	OK
Ratio of 219 to 69 should be > 40% and is	126.00	OK
Ratio of 502 to 69 should be > 2.4% and is	12.76	OK
Mass 69 Precursor (<= 3%)	0.10	OK
Mass 219 Precursor (<= 6%)	0.28	OK
Mass 502 Precursor (<= 12%)	0.49	OK

597x Air and Water Check
Mon Mar 25 11:17:31 2013 Instrument: MH 5977
D:\MASSHUNTER\GCMS\1\5977\atune.u

Testing for a leak in the system		
Ratio of 18 to 69 (<20%)	0.52	OK
Ratio of 28 to 69 (<10%)	2.35	OK
Electron Multiplier Voltage	1576	OK

Tune portion of System Verification passed.

Parameter	Value
Gas Chromatograph	
Model	Agilent 7890B gas chromatograph
Column	Agilent DB-5, 60 m × 0.25 mm, 0.1 µm (p/n 122-5061)
Column Pneumatics	Constant flow, He carrier gas
Injector Mode	Splitless
Injector Liner	Inlet liner, splitless, double taper, deactivated (p/n 5181-3315)
Injection Volume	1.0 µL
Injector Temperature	290 °C
Flow Rate	0.93 mL/min
Temperature Program	90 °C for 2 min, 22 °C/min to 200 °C, 1 °C/min to 215 °C, hold 10 min, 5.2 °C/min to 300 °C, hold 2.7 min
Total Run Time	51.05 min
Equilibration Time	0.1 min
Mass Spectrometer	
Model	Agilent 7010B Triple Quadrupole GC/MS
Ionization Mode	EI, 70 eV
Acquisition Mode	MRM
Filament Current	100 µA
Collision Gas	N_2 at 1.5 mL/min
Quench Gas	He at 2.25 mL/min
GC Interface Temperature	290 °C
Ion Source Temperature	290 °C
Quadrupole 1 Temperature	150 °C
Quadrupole 2 Temperature	150 °C

Table 6. MRM parameters and collision energy.

Segment	Analyte	Precursor ion[1]	Product ion[1]	Dwell	CE[2]
1 Toxic TCDD/TCDF	^{13}C-TCDD	333.9	269.9	50	26
	^{13}C-TCDD	331.9	267.9	50	26
	TCDD	321.9	258.9	100	26
	TCDD	319.9	256.9	100	26
	^{13}C-TCDF	317.9	253.9	50	40
	^{13}C-TCDF	315.9	251.9	50	40
	TCDF	305.9	242.9	100	40
	TCDF	303.9	240.9	100	40
2 Nontoxic TCDD/TCDF[3]	^{13}C-PeCDF	351.9	287.9	25	40
	^{13}C-PeCDF	349.9	285.9	25	40
	PeCDF	339.9	276.9	75	40
	PeCDF	337.9	274.9	75	40
	^{13}C-TCDD	333.9	269.9	25	26
	^{13}C-TCDD	331.9	267.9	25	26
	TCDD	321.9	258.9	75	26
	TCDD	319.9	256.9	75	26
	^{13}C-TCDF	317.9	253.9	25	40
	^{13}C-TCDF	315.9	251.9	25	40
	TCDF	305.9	242.9	75	40
	TCDF	303.9	240.9	75	40
3 PeCDD/PeCDF	^{13}C-PeCDD	367.9	302.9	50	26
	^{13}C-PeCDD	365.9	301.9	50	26
	PeCDD	355.9	292.9	100	26
	PeCDD	353.9	290.9	100	26
	^{13}C-PeCDF	351.9	287.9	50	40
	^{13}C-PeCDF	349.9	285.9	50	40
	PeCDF	339.9	276.9	100	40
	PeCDF	337.9	274.9	100	40

Segment	Analyte	Precursor ion[1]	Product ion[1]	Dwell	CE[2]
4 HxCDD/HxCDF	^{13}C-HxCDD	403.9	339.9	50	25
	^{13}C-HxCDD	401.9	337.9	50	25
	HxCDD	391.8	328.8	100	25
	HxCDD	389.8	326.8	100	25
	^{13}C-HxCDF	387.9	323.9	50	40
	^{13}C-HxCDF	385.9	321.9	50	40
	HxCDF	375.8	312.8	100	40
	HxCDF	373.8	310.8	100	40
5 HpCDD/HpCDF	^{13}C-HpCDD	437.8	373.8	50	24
	^{13}C-HpCDD	435.8	371.8	50	24
	HpCDD	425.8	362.8	100	24
	HpCDD	423.8	360.8	100	24
	^{13}C-HpCDF	421.8	357.8	50	40
	^{13}C-HpCDF	419.8	355.8	50	40
	HpCDF	409.8	346.8	100	40
	HpCDF	407.8	344.8	100	40
6 OCDD/OCDF	^{13}C-OCDD	471.8	407.8	50	24
	^{13}C-OCDD	469.8	405.8	50	24
	OCDD	459.7	396.7	100	24
	OCDD	457.7	394.7	100	24
	^{13}C-OCDF	455.8	391.8	50	40
	^{13}C-OCDF	453.8	389.8	50	40
	OCDF	443.7	380.7	100	40
	OCDF	441.7	378.7	100	40

[1] Wide resolution for precursor and product ions.

[2] Collision energies were adapted from Agilent Food and Feed Analyzer[9].

[3] Segment 2 was added to account for the last eluted TCDD/TCDF and first eluted PeCDD/PeCDF. The retention time of the congeners is too close to TCDD/TCDF in segment 1 and PeCDF in segment 3. The compounds in segment 2 are for investigating nontoxic CDDs/CDFs, and are optional if investigating only the toxic compounds.

Analytes	Primary MRM Transition (*m/z*)	Collision Energy (CE)	Secondary MRM Transition (*m/z*)	CE	Surrogate
1,2,3,4,6,7,8-HpCDF	407.8 → 344.8	36	409.8 → 346.8	36	$^{13}C_{12}$-1,2,3,4,6,7,8-HpCDF
1,2,3,4,7,8,9-HpCDF	407.8 → 344.8	36	409.8 → 346.8	36	$^{13}C_{12}$-1,2,3,4,7,8,9-HpCDF
OCDF	441.7 → 378.8	35	443.7 → 380.8	35	$^{13}C_{12}$-OCDD
Cleanup Standard					
$^{37}Cl_4$-2,3,7,8-TCDD	327.9 → 262.9	33	-		$^{13}C_{12}$-1,2,3,4-TCDD
Labeled Surrogates					**Recovery Calculated Using**
$^{13}C_{12}$-2,3,7,8-TCDD	331.9 → 268.0	24	333.9 → 270.0	24	$^{13}C_{12}$-1,2,3,4-TCDD
$^{13}C_{12}$-1,2,3,7,8-PeCDD	367.9 → 303.9	25	365.9 → 301.9	25	$^{13}C_{12}$-1,2,3,4-TCDD
$^{13}C_{12}$-1,2,3,4,7,8-HxCDD	401.9 → 337.9	25	403.9 → 339.9	25	$^{13}C_{12}$-1,2,3,7,8,9-HxCDD
$^{13}C_{12}$-1,2,3,6,7,8-HxCDD	401.9 → 337.9	25	403.9 → 339.9	25	$^{13}C_{12}$-1,2,3,7,8,9-HxCDD
$^{13}C_{12}$-1,2,3,4,6,7,8-HpCDD	435.8 → 371.9	25	437.8 → 373.9	25	$^{13}C_{12}$-1,2,3,7,8,9-HxCDD
$^{13}C_{12}$-OCDD	469.8 → 405.8	26	471.8 → 407.8	26	$^{13}C_{12}$-1,2,3,7,8,9-HxCDD
$^{13}C_{12}$-2,3,7,8 -TCDF	315.9 → 252.0	33	317.9 → 254.0	33	$^{13}C_{12}$-1,2,3,4-TCDD
$^{13}C_{12}$-1,2,3,7,8-PeCDF	351.9 → 287.9	35	349.9 → 285.9	35	$^{13}C_{12}$-1,2,3,4-TCDD
$^{13}C_{12}$-2,3,4,7,8-PeCDF	351.9 → 287.9	35	349.9 → 285.9	35	$^{13}C_{12}$-1,2,3,4-TCDD
$^{13}C_{12}$-1,2,3,4,7,8-HxCDF	385.9 → 321.9	35	387.9 → 323.9	35	$^{13}C_{12}$-1,2,3,7,8,9-HxCDD
$^{13}C_{12}$-1,2,3,6,7,8-HxCDF	385.9 → 321.9	35	387.9 → 323.9	35	$^{13}C_{12}$-1,2,3,7,8,9-HxCDD
$^{13}C_{12}$-1,2,3,7,8,9-HxCDF	385.9 → 321.9	35	387.9> 323.9	35	$^{13}C_{12}$-1,2,3,7,8,9-HxCDD
$^{13}C_{12}$-2,3,4,6,7,8-HxCDF	385.9 → 321.9	35	387.9 → 323.9	35	$^{13}C_{12}$-1,2,3,7,8,9-HxCDD
$^{13}C_{12}$-1,2,3,4,6,7,8-HpCDF	419.8 → 355.9	36	421.8 → 357.9	36	$^{13}C_{12}$-1,2,3,7,8,9-HxCDD
$^{13}C_{12}$-1,2,3,4,7,8,9-HpCDF	419.8 → 355.9	36	421.8 → 357.9	36	$^{13}C_{12}$-1,2,3,7,8,9-HxCDD
Recovery Standards					
$^{13}C_{12}$-1,2,3,4-TCDD	331.9 → 268.0	24	333.9 → 270.0	24	
$^{13}C_{12}$-1,2,3,7,8,9-HxCDD	401.9 → 337.9	25	403.9 → 339.9	25	
CI-DPE Transitions					
Descriptor			**Type**	**Substance**	
1	375.8 → 305.9	30	M+2	HxCDPE	
2	409.8 → 339.9	25	M+2	HpCDPE	
3	445.8 → 373.8	30	M+4	OCDPE	
4	479.7 → 407.8	30	M+4	NCDPE	
5	513.7 → 443.7	30	M+4	DCDPE	

Species Monitored	MRM Transition Precursor *m/z* (Primary/Secondary)	MRM Transition Product* Ion Theoretical Ratio†	QC Limit**	
			Lower	Upper
$Cl_4CDD^{\dagger}$	(M+2)/M	0.96	0.82	1.10
Cl_4CDF	(M+2)/M	0.96	0.82	1.10
Cl_5CDD	M/(M+2)	0.78	0.66	0.90
Cl_5CDF	M/(M+2)	0.78	0.66	0.90
Cl_6CDD	(M+4)/(M+2)	0.64	0.54	0.74
Cl_6CDF	(M+4)/(M+2)	0.64	0.54	0.74
Cl_7CDD	(M+4)/(M+2)	0.80	0.68	0.92
Cl_7CDF	(M+4)/(M+2)	0.80	0.68	0.92
Cl_8CDD	(M+4)/(M+2)	0.96	0.82	1.10
Cl_8CDF	(M+4)/(M+2)	0.96	0.82	1.10

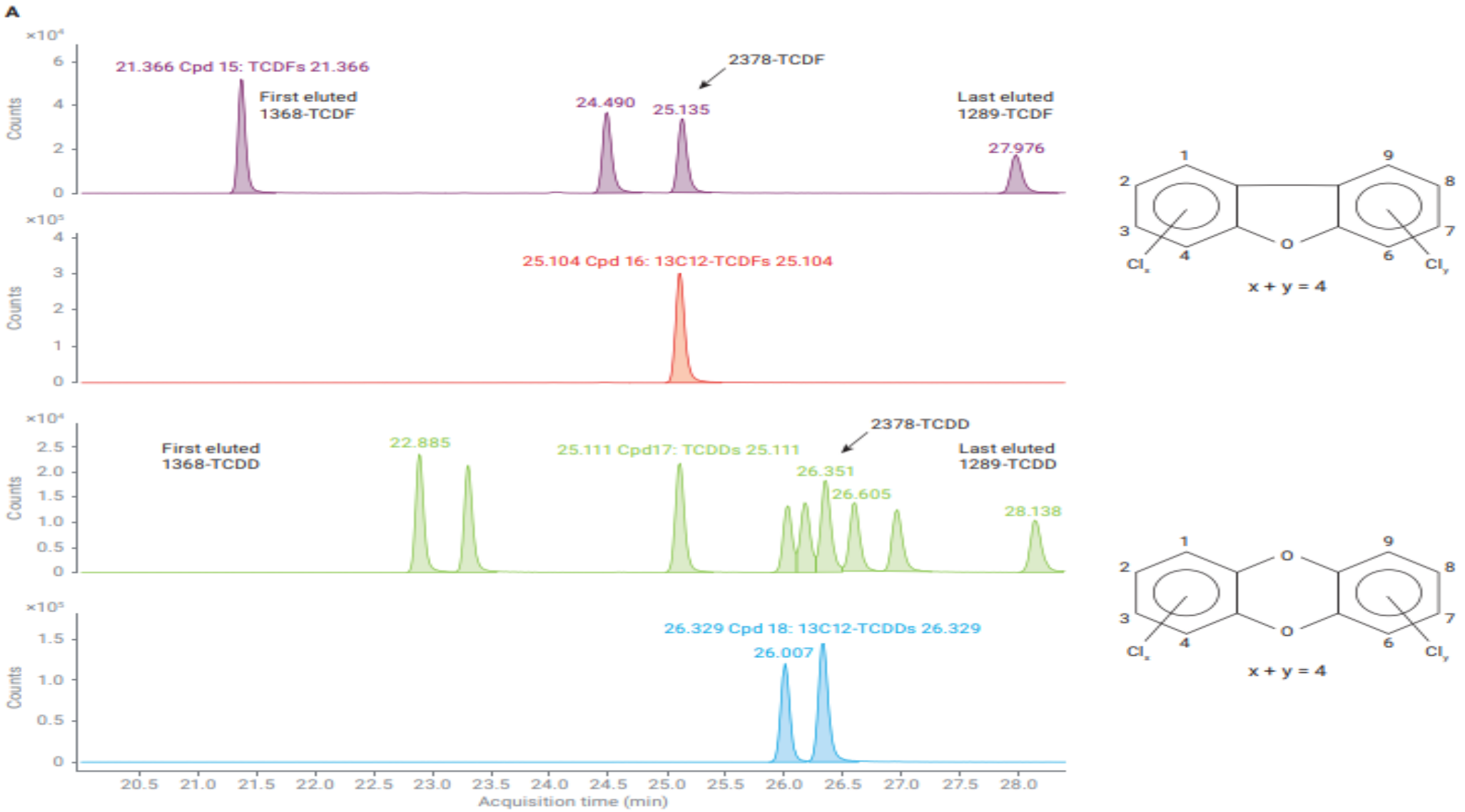


Figure 1A. MRM chromatograms for tetrachlorinated dibenzofurans (TCDFs), labeled TCDF ISTD, tetrachlorinated dibenzodioxins (TCDDs), and labeled TCDD ISTD.

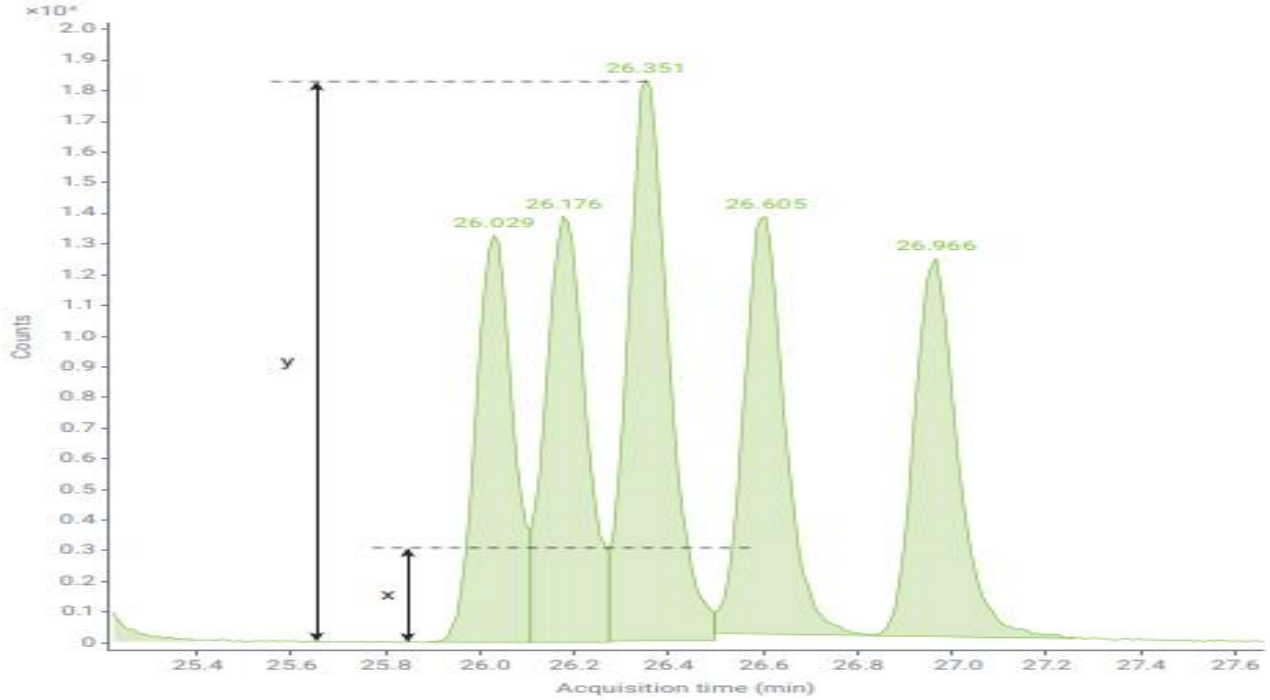


Figure 2. 2,3,7,8-TCDD and its close eluters.

A

Method Table

Time Segment: <All> Compound: 2,3,7,8-TCDD Reset Table View

Quantifier

Name	TS	Transition	Scan	Type	Resolution Calculation Type	Resolution Limit
2,3,7,8-TCDD	1	319.9 -> 256.9...	MRM	Target	Valley Height/Peak Heig...	25.0

B

Sample							2,3,7,8-TCDD Results						
Name	Data File	Type	Level	Vial	Acq. Date-Time	Acq. Method File	RT	Resp.	MI	Calc. Conc.	Accuracy	S/N	Resolution F.	Resolution R.
DX041D-CAL_/01-73	DX9Z0444.D	Cal	CS3	7	8/22/2019 2:31 AM	TQEI_DB5_DX_11	26.351	221151		9.0183	90.2	2339.21	20.4	7.8

Figure 3. (A) Method setup for resolution check in MassHunter Quantitative Analysis; (B) front and rear valley height/peak height resolution calculated for 2,3,7,8-TCDD and its closest eluting isomers.

Calibration and linear range

EPA 1613b CS1

Cal. Sample Name	Level
200 ppt Cal Std.	L1
500 ppt Cal Std.	L2
1000 pptCal Std.	L3
4000 ppt Cal Std.	L4
10000 ppt Cal Std.	L5
50000 ppt Cal Std.	L6
250000 ppt Cal Std.	L7
1000000 ppt Cal Std.	L8
2500000 ppt Cal Std.	L9

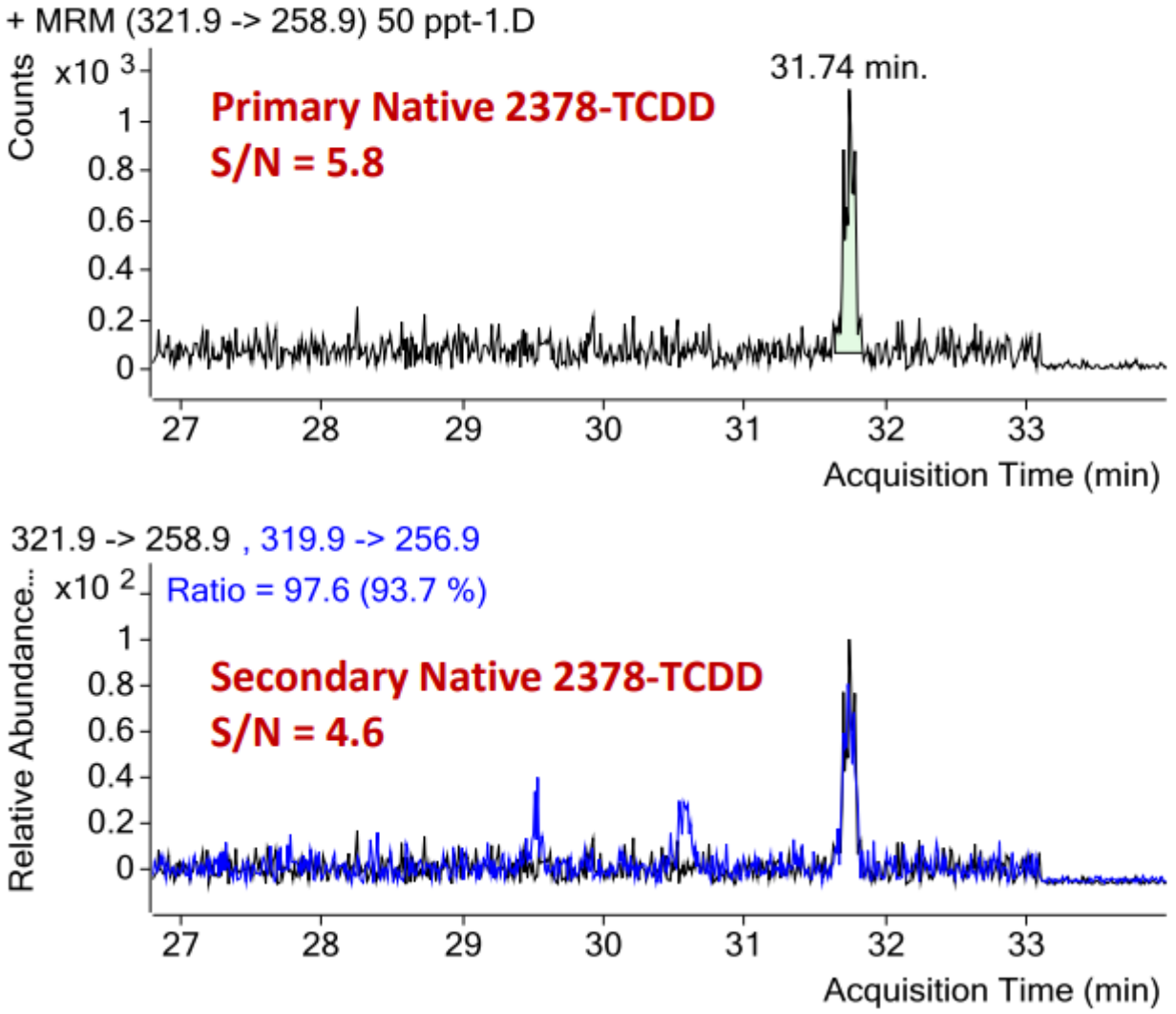
Name	Avg. RF	Avg. RF RSD	CS1 RF	Difference	CS1 S/N	CS1 RRT	1613b RRT criteria	Pass/Fail
2378-TCDD	1.123	6	1.004	-11%	*25*	1.002	0.999-1.002	*Pass*
2378-TCDF	0.97	2.9	0.943	-3%	*50*	1.001	0.999-1.003	*Pass*
12378-PeCDD	0.985	3.5	0.994	1%	*42*	1.001	0.999-1.002	*Pass*
12378-PeCDF	0.991	2.8	1.025	3%	*54*	1.001	0.999-1.002	*Pass*
23478-PeCDF	1.007	2.1	0.997	-1%	*63*	1.000	0.999-1.002	*Pass*
123478-HxCDD	0.991	4.2	0.999	1%	*21*	1.001	0.999-1.001	*Pass*
123478-HxCDF	0.924	4.4	0.921	0%	*33*	1.001	0.998-1.004	*Pass*
123678-HxCDD	0.929	3.6	0.917	-1%	*25*	1.000	1.000-1.019	*Pass*
123678-HxCDF	0.908	4.5	0.877	-3%	*43*	1.000	0.999-1.001	*Pass*
123789-HxCDD	1.027	5.3	1.000	-3%	*42*	1.000	0.997-1.005	*Pass*
123789-HxCDF	0.912	5.2	0.902	-1%	*38*	1.000	0.999-1.001	*Pass*
234678-HxCDF	0.983	4.1	0.999	2%	*48*	1.000	0.999-1.001	*Pass*
1234678-HpCDD	1.008	4	1.033	2%	*83*	1.000	0.999-1.001	*Pass*
1234678-HpCDF	0.912	3.5	0.943	3%	*92*	1.000	0.999-1.001	*Pass*
1234789-HpCDF	0.902	4.2	0.948	5%	*90*	1.000	0.999-1.001	*Pass*
OCDD	1.056	2.4	1.040	-1%	*150*	1.000	0.999-1.001	*Pass*
OCDF	0.913	3.5	0.940	3%	*148*	1.000	0.999-1.008	*Pass*

$$\mathbf{RF} = \frac{A_{2,3,7,8-\mathrm{TCDD},Std}}{A_{13C,Std}} \times \frac{M_{13C,Std}\ (ng)}{M_{2,3,7,8-\mathrm{TCDD},Std}\ (ng)}$$

Verification Standards recoveries

Comp. Name	Chemstation Amt (ng)	Theoretical Amt (ng)	% Recovery	1613b criteria	Pass/Fail
2378-TCDF	1.815	2	91%	84-120%	Pass
2378-TCDD	1.833	2	92%	78-129%	Pass
12378-PCDF	4.790	5	96%	82-120%	Pass
23478-PCDF	4.705	5	94%	82-122%	Pass
12378-PCDD	4.742	5	95%	78-130%	Pass
123478-HxCDF	4.642	5	93%	90-112%	Pass
123678-HxCDF	4.629	5	93%	88-114%	Pass
234678-HxCDF	4.600	5	92%	88-114%	Pass
123789-HxCDF	4.701	5	94%	90-112%	Pass
123478-HxCDD	4.342	5	87%	78-128%	Pass
123678-HxCDD	4.385	5	88%	78-128%	Pass
123789-HxCDD	4.422	5	88%	82-122%	Pass
1234678-HpCDF	4.823	5	96%	90-110%	Pass
1234789-HpCDF	5.097	5	102%	86-116%	Pass
1234678-HpCDD	4.840	5	97%	86-116%	Pass
OCDF	9.221	10	92%	63-159%	Pass
OCDD	9.175	10	92%	79-126%	Pass

Low working range and sensitivity



50 femtogram of 2378-TCDD on the column

- > 2.5 signal to noise ratio
- The relative ion intensities is within 15% difference to the calibration average.

Continuing Calibration Report

Batch Name E:\Pace 1613\03292018\QuantResults\05082018-4.batch.bin
Method File E:\Pace 1613\03292018\1613 proccessing.m
Daily CC E:\Pace 1613\03292018032918_08.D

Level name	Injection Time	Calibration Files
0.5	3/29/2018 3:06:11 PM	E:\Pace 1613\03292018\032918_03.D
2	3/29/2018 3:55:41 PM	E:\Pace 1613\03292018\032918_04.D
10	3/29/2018 4:45:12 PM	E:\Pace 1613\03292018\032918_05.D
40	3/29/2018 5:34:38 PM	E:\Pace 1613\03292018\032918_06.D
200	3/29/2018 6:24:08 PM	E:\Pace 1613\03292018\032918_07.D
CC	3/29/2018 7:13:32 PM	E:\Pace 1613\03292018\032918_08.D <=======

ISTD Compound:	Avg Resp	Mid Resp	CC Resp	Area%	RT	Diff	A/M
1 I 13C 2,3,7,8 TCDD	1200841	1128653	1007947	83.94	28.361	0.001	A
2 I 13C 2,3,7,8 TCDF	1855290	1720447	1752220	94.44	26.922	0.013	A
4 I 13C- 2,3,4,7,8 PCDF	1626873	1511377	1342970	82.55	35.182	0.000	A
3 I 13C-1,2,3,6,8 PCDD	1053816	994703	909185	86.28	35.451	0.010	A
5 I 13C- 1,2,3,4,7,8 HxCDD	803043	726069	540764	67.34	37.903	0.010	A
4 I 13C 1,2,3,7,8 PCDF	1465718	1494749	1310795	89.43	34.269	0.010	A
6 13C- 1,2,3,4,7,8 HxCDF	946111	849834	261104	27.60	37.756	0.020	A
6 I13C 1.2.3.6.7.8 HxCDF	992349	899279	788085	79.42	37.380	0.010	A
6 I 13C 1,2,3,7,8,9 HxCDF	992349	899279	788085	79.42	37.380	0.010	A
6 13C 2,3,4,6,7,8 HxCDF	891790	814024	586423	65.76	38.273	0.010	A
5 I 13C- 1,2,3,4,6,7,8 HpCDD	524813	475001	375918	71.63	39.939	0.020	A
6 I 13C- 1,2,3,4,6,7,8 HpCDF	591050	539042	354137	59.92	40.299	0.010	A
5 I 13C- 1,2,3,6,7,8 HxCDD	803043	726069	540764	67.34	37.903	0.010	A
R 13C- 1,2,3,7,8,9 HxCDD	753263	703170	557521	74.01	38.065	0.010	A
7 I 13C-OCDD	698557	647464	389167	55.71	42.067	0.020	A
R 13C 1,2,3,4 TCDD	1200841	1128653	1007947	83.94	28.361	0.001	A
6 I 13C- 1,2,3,4,7,8,9 HpCDF	591050	539042	354137	59.92	40.299	0.010	A

Target Compound	AvgRF/R2	CC RF	Exp. Conc	Calc. Conc	%Dev	Area%	Curve Fit
1 I 13C 2,3,7,8 TCDD	----------ISTD----------						
W2 1,2,8,9 TCDD	1.2589	1.2335	10.10	9.90	2.02	19.92	Avg RF
T 2,3,7,8 TCDD	1.1474	1.2327	10.00	10.74	-7.44	19.75	Avg RF
W2 1,3,6,8 TCDD	1.1666	1.2230	10.10	10.59	-4.83	19.75	Avg RF
2 I 13C 2,3,7,8 TCDF	----------ISTD----------						
W1 1,2,8,9 TCDF	1.3158	1.1754	10.10	9.02	10.67	20.69	Avg RF
T 2,3,7,8 TCDF	1.2287	1.1870	10.00	9.66	3.40	20.60	Avg RF
W1 1,3,6,8 TCDF	1.2575	1.1754	10.10	9.44	6.53	20.60	Avg RF
4 I 13C- 2,3,4,7,8 PCDF	----------ISTD----------						
T 2,3,4,7,8 PCDF	5.9087	5.5221	10.00	9.35	6.54	17.01	Avg RF
3 I 13C-1,2,3,6,8 PCDD	----------ISTD----------						
W4 1,2,3,8,,9 PCDD	5.8731	5.3763	10.10	9.25	8.46	19.03	Avg RF
T 1,2,3,7,8 PCDD	5.5231	5.4312	10.00	9.83	1.66	19.03	Avg RF
W4 1,2,4,7,9 PCDD	5.6973	5.3766	10.10	9.53	5.63	19.03	Avg RF
5 I 13C- 1,2,3,4,7,8 HxCDD	----------ISTD----------						
W6 1,2,3,7,8,9 HxCDD	4.6378	4.9515	10.10	10.78	-6.76	16.35	Avg RF
T 1,2,3,4,7,8 HxCDD	4.8254	4.6984	10.00	9.74	2.63	14.60	Avg RF
T 1,2,3,7,8,9 HxCDD	4.7834	5.2586	10.00	10.99	-9.94	16.99	Avg RF
W6 1,2,4,6,7,9 HxCDD	4.7835	5.2126	10.10	11.01	-8.97	17.08	Avg RF
4 I 13C 1,2,3,7,8 PCDF	----------ISTD----------						
W3 1,3,4,6,8 PCDF	5.1718	5.4831	10.10	10.71	-6.02	22.06	Avg RF
T 1,2,3,7,8 PCDF	5.2581	5.5347	10.00	10.53	-5.26	22.04	Avg RF
W3 1,2,3,8,9 PCDF	6.5307	5.6033	10.10	8.67	14.20	17.01	Avg RF
6 13C- 1,2,3,4,7,8 HxCDF	----------ISTD----------						
T 1,2,3,4,7,8 HxCDF	5.1578	6.2640	10.00	12.14	-21.45	7.41	Avg RF

Generated at 2:56 PM on 5/8/2018

ISTD and Surrogate Recovery% Report

Batch Name E:\Pace 1613\03292018\QuantResults\05082018-4.batch.bin
Data File E:\Pace 1613\03292018\032918_03.D
Sample Name CS1

Level name		Calibration Files	
0.5	3/29/2018 3:06:11 PM	E:\Pace 1613\03292018\032918_03.D	
2	3/29/2018 3:55:41 PM	E:\Pace 1613\03292018\032918_04.D	
10	3/29/2018 4:45:12 PM	E:\Pace 1613\03292018\032918_05.D	
40	3/29/2018 5:34:38 PM	E:\Pace 1613\03292018\032918_06.D	
200	3/29/2018 6:24:08 PM	E:\Pace 1613\03292018\032918_07.D	
CC	3/29/2018 7:13:32 PM	E:\Pace 1613\03292018\032918_08.D	<======

ISTD Recovery%

Compound Name	Avg Resp	CC Resp	Resp In Sample	Area%_Avg
2 I 13C 2,3,7,8 TCDF	1855290	1752220	1631619	87.94%
1 I 13C 2,3,7,8 TCDD	1200841	1007947	1026550	85.49%
R 13C 1,2,3,4 TCDD	1200841	1007947	1026550	85.49%
4 I 13C 1,2,3,7,8 PCDF	1465718	1310795	1344326	91.72%
4 I 13C- 2,3,4,7,8 PCDF	1626873	1342970	1495444	91.92%
3 I 13C-1,2,3,6,8 PCDD	1053816	909185	967401	91.80%
6 I13C 1.2.3.6.7.8 HxCDF	992349	788085	898442	90.54%
6 I 13C 1,2,3,7,8,9 HxCDF	992349	788085	898442	90.54%
6 13C- 1,2,3,4,7,8 HxCDF	946111	261104	841051	88.90%
5 I 13C- 1,2,3,4,7,8 HxCDD	803043	540764	727809	90.63%
5 I 13C- 1,2,3,6,7,8 HxCDD	803043	540764	727809	90.63%
R 13C- 1,2,3,7,8,9 HxCDD	753263	557521	645606	85.71%
6 13C 2,3,4,6,7,8 HxCDF	891790	586423	782431	87.74%
5 I 13C- 1,2,3,4,6,7,8 HpCDD	524813	375918	457367	87.15%
6 I 13C- 1,2,3,4,6,7,8 HpCDF	591050	354137	514475	87.04%
6 I 13C- 1,2,3,4,7,8,9 HpCDF	591050	354137	514475	87.04%
7 I 13C-OCDD	698557	389167	597064	85.47%

Surrogate Recovery%

Compound Name	Avg Resp	CC Resp	Resp In Sample	Area%_Avg
S 13C 2,3,7,8 TCDF	1861034	1756952	1625727	87.36%
S 13C 2,3,7,8 TCDD	1208120	1006233	1049221	86.85%
S CL37 TCDD	3710290	0	38874	1.05%
S 13C 1,2,3,7,8 PCDF	1466601	1311308	1345672	91.75%
S 13C- 2,3,4,7,8 PCDF	1626494	1342928	1496542	92.01%
S 13C-PCDD	1053753	909170	967340	91.80%
S 13C 1.2.3.6.7.8 HxCDF	992349	788085	898442	90.54%
S 13C- 1,2,3,4,7,8 HxCDF	946111	261104	841051	88.90%
S 13C 1,2,3,7,8,9 HxCDF	946111	261104	841051	88.90%
S 13C- 1,2,3,4,7,8 HxCDD	802999	541024	720445	89.72%
S 13C- 1,2,3,6,7,8 HxCDD	802999	541024	720445	89.72%
S 13C 2,3,4,6,7,8 HxCDF	891873	584463	784236	87.93%
S 13C- 1,2,3,4.6,7,8 HpCDF	680423	493330	598076	87.90%
S 13C- 1,2,3,4,6,7,8 HpCDD	523734	375085	455413	86.96%
S 13C- 1,2,3,4,7,8,9 HpCDF	591050	354137	514475	87.04%
S 13C-OCDD	698583	389286	597219	85.49%

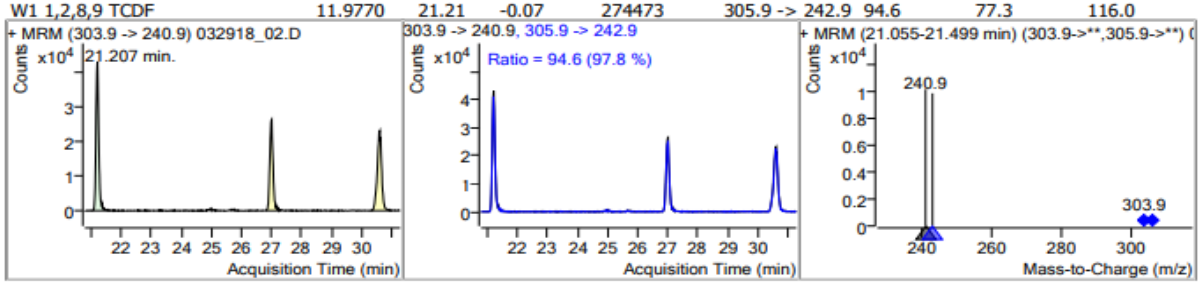
Quantitation Results Report (Not Reviewed)

Compound	RT	QIon	Resp.	Conc.	Units	Dev(Min)
Spiked Amount: 100.000	Range: 50.0 - 150.0%			Recovery = 101.99%		
S 13C- 1,2,3,4,7,8,9 HpCDF	40.309	419.8 -> 355.8	502627	99.9654	ng/ml	0.020
Spiked Amount: 100.000	Range: 50.0 - 150.0%			Recovery = 99.97%		
S 13C-OCDD	42.067	469.8 -> 405.8	601784	101.2227	ng/ml	0.020
Spiked Amount: 100.000	Range: 50.0 - 150.0%			Recovery = 101.22%		
Target Compounds						**QValue**
W1 1,2,8,9 TCDF	21.207	303.9 -> 240.9	274473	11.9770	ng/ml	98
W2 1,2,8,9 TCDD	23.243	319.9 -> 256.9	160866	11.4129	ng/ml	100
T 2,3,7,8 TCDF	27.006	303.9 -> 240.9	197215	9.2156	ng/ml	100
T 2,3,7,8 TCDD	28.436	319.9 -> 256.9	111063	8.6455	ng/ml	82
W2 1,3,6,8 TCDD	30.274	319.9 -> 256.9	120426	9.2194	ng/ml	99
W3 1,3,4,6,8 PCDF	30.483	339.9 -> 276.9	729064	9.9419	ng/ml	99
W1 1,3,6,8 TCDF	30.602	303.9 -> 240.9	235594	10.7572	ng/ml	99
W4 1,2,3,8,,9 PCDD	33.281	355.9 -> 292.9	754312	12.2040	ng/ml	100
T 1,2,3,7,8 PCDF	34.301	339.9 -> 276.9	749416	10.0518	ng/ml	98
T 2,3,4,7,8 PCDF	35.214	339.9 -> 276.9	968414	10.0022	ng/ml	98
T 1,2,3,7,8 PCDD	35.473	355.9 -> 292.9	574486	9.8836	ng/ml	99
W4 1,2,4,7,9 PCDD	35.778	355.9 -> 292.9	705544	11.7673	ng/ml	97
W3 1,2,3,8,9 PCDF	35.965	339.9 -> 276.9	984104	10.6274	ng/ml	99
W6 1,2,3,7,8,9 HxCDD	36.891	389.8 -> 326.8	342825	10.9340	ng/ml	100
T 1,2,3,6,7,8 HxCDF	37.322	373.8 -> 310.8	426620	10.3811	ng/ml	99
W5 1,2,3,4,6,8 HxCDF	37.322	373.8 -> 310.8	426593	10.5183	ng/ml	98
T 1,2,3,7,8,9 HxCDF	37.393	373.8 -> 310.8	413457	10.0337	ng/ml	100
T 1,2,3,4,7,8 HxCDF	37.758	373.8 -> 310.8	440085	10.0977	ng/ml	100
T 1,2,3,4,7,8 HxCDD	37.916	389.8 -> 326.8	326338	10.0037	ng/ml	98
T 1,2,3,6,7,8 HxCDD	37.916	389.8 -> 326.8	326338	10.0037	ng/ml	98
T 1,2,3,7,8,9 HxCDD	38.078	389.8 -> 326.8	324729	10.0418	ng/ml	99
W6 1,2,4,6,7,9 HxCDD	38.078	389.8 -> 326.8	324924	10.0475	ng/ml	99
T 2,3,4,6,7,8 HxCDF	38.286	373.8 -> 310.8	361005	9.7968	ng/ml	100
W5 1,2,3,4,8,9 HxCDF	38.286	373.8 -> 310.8	360794	9.5603	ng/ml	100
T 1,2,3,4,7,8,9 HpCDF	39.226	407.8 -> 344.8	319863	10.5757	ng/ml	99
W7 1,2,3,4,6,7,8 HpCDF	39.226	407.8 -> 344.8	319801	10.5726	ng/ml	99
W8 1,2,3,4,6,7,9 HpCDD	39.424	423.8 -> 360.8	282634	10.7411	ng/ml	100
T 1,2,3,4,6,7,8 HpCDD	39.941	423.8 -> 360.8	262512	10.2756	ng/ml	100
W8 1,2,3,4,6,7,8 HpCDD	39.941	423.8 -> 360.8	262732	10.2663	ng/ml	100
T 1,2,3,4,6,7,8 HpCDF	40.312	407.8 -> 344.8	274884	10.1779	ng/ml	99
W7 1,2,3,4,7,8,9 HpCDF	40.312	407.8 -> 344.8	275806	10.2046	ng/ml	99
T OCDD	42.080	457.7 -> 394.7	281234	9.7196	ng/ml	100
T OCDF	42.237	441.7 -> 378.7	304751	9.7216	ng/ml	98

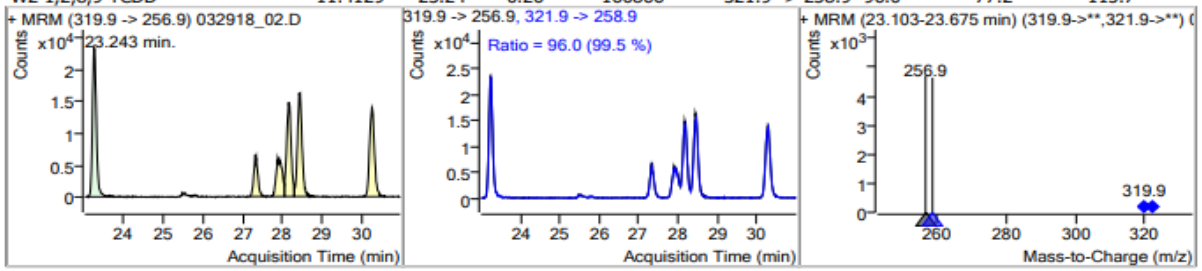
(#) = Qualifier Out of Range; (m) = Manual Integration; (+) = Area Summed; (*) = Surrogate Percent Recovery Out of Range; (d): Zeroed Peak

Quantitation Results Report (Not Reviewed)

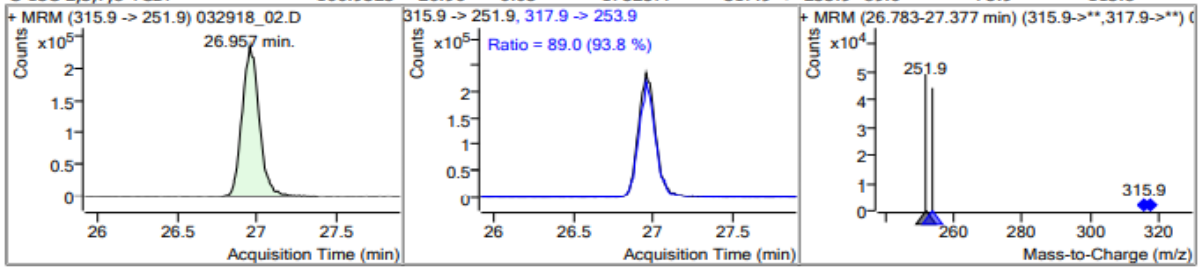
Compound	Conc.	RT	Dev(Min)	Resp.	QIon	QRatio	Lower	Upper
W1 1,2,8,9 TCDF	11.9770	21.21	-0.07	274473	305.9 -> 242.9	94.6	77.3	116.0



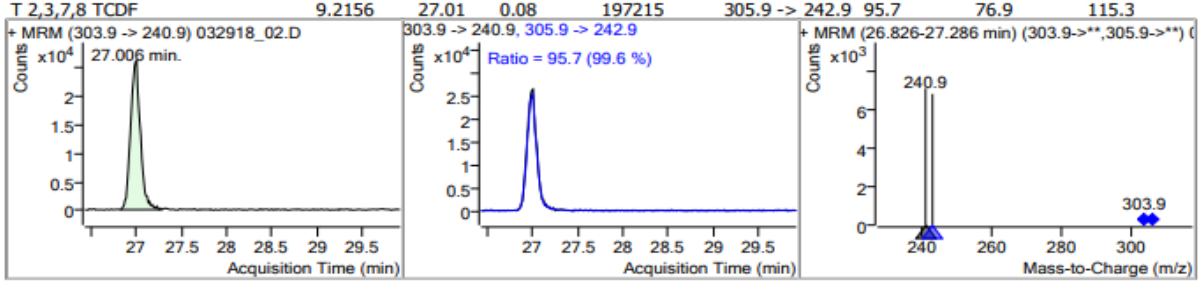
Compound	Conc.	RT	Dev(Min)	Resp.	QIon	QRatio	Lower	Upper
W2 1,2,8,9 TCDD	11.4129	23.24	-0.26	160866	321.9 -> 258.9	96.0	77.2	115.7



Compound	Conc.	RT	Dev(Min)	Resp.	QIon	QRatio	Lower	Upper
S 13C 2,3,7,8 TCDF	100.9325	26.96	0.05	1752577	317.9 -> 253.9	89.0	75.9	113.8



Compound	Conc.	RT	Dev(Min)	Resp.	QIon	QRatio	Lower	Upper
T 2,3,7,8 TCDF	9.2156	27.01	0.08	197215	305.9 -> 242.9	95.7	76.9	115.3



The 7010B triple quadrupole GC/MS is equipped with a high-efficiency EI source that produces up to 20 times more ions and maximizes ion transfer into the quadrupole mass analyzer, allowing significantly more sensitivity while still maintaining robustness.

Linearity, MDLs, total PCDD/PCDF

The GC/TQ system showed good linearity over the Method 1613B calibration range and met Method 1613B specifications. Linearity values expressed in terms of % RSDs of response factors for the target analytes across the calibration range were less than 20% and ranged from 2.2 to 15.4%. The 20% RSD limit does not apply to the labeled compounds, which are quantified by internal standard, not by isotope dilution. The %RSD of the PCDD/PCDF response factors for the five sets (days) of initial calibrations for the GC/TQ system are shown in Table 4. The results underscored the excellent dynamic range of the 7010B triple quadrupole GC/MS system.

The GC/TQ MDL results for the aqueous (1 L), solid (10 g), and tissue (10 g) samples are shown in Table 5. The results obtained using the 7010B triple quadrupole GC/MS system far surpassed Method 1613B MRLs.

Total PCDD and PCDF concentrations from the real-world sample extracts were reported by MassHunter software for each level of chlorination by summing the concentration of the individual peaks meeting quantification criteria (peak shape, S/N, and product ion ratio) in the appropriate retention time window. Figure 4 shows the comparison of the total PCDD and PCDF concentrations determined using GC/HRMS and GC/TQ. The results for the two technologies were comparable.

Table 4. %RSDs of the PCDD/PCDF response factors for the five days of initial calibrations.

Date Acquired	19-AUG-19	21-AUG-19	06-JAN-20	07-JAN-20	08-JAN-20
Data File ID	DX9Z0415-A1	DX9Z0444-A1	DX9Z0830-A1	DX9Z0837-A1	DX9Z0853-A1
Name	RRF %RSD	RRF %RSD	RRF %RSD	RRF %RSD	RRF %RSD
2,3,7,8-TCDF	4.0	3.0	4.4	2.7	2.4
1,2,3,7,8-PeCDF	3.7	2.8	3.4	2.9	2.7
2,3,4,7,8-PeCDF	3.8	3.5	4.1	3.9	4.3
1,2,3,4,7,8-HxCDF	3.1	4.5	4.4	2.3	5.6
1,2,3,6,7,8-HxCDF	3.0	3.5	5.3	3.6	8.1
2,3,4,6,7,8-HxCDF	3.0	3.9	6.2	4.5	1.3
1,2,3,7,8,9-HxCDF	4.6	5.4	6.7	2.7	6.0
1,2,3,4,6,7,8-HpCDF	3.2	4.3	3.7	4.8	4.3
1,2,3,4,7,8,9-HpCDF	4.6	4.7	4.6	5.8	4.0
OCDF	7.1	10.2	9.0	7.0	6.3
2,3,7,8-TCDD	2.9	4.8	6.3	5.6	7.3
1,2,3,7,8-PeCDD	4.6	4.6	2.2	2.3	3.9
1,2,3,4,7,8-HxCDD	4.3	4.0	2.3	2.3	3.1
1,2,3,6,7,8-HxCDD	5.4	5.3	5.2	2.6	5.3
1,2,3,7,8,9-HxCDD	5.3	3.4	6.8	3.6	4.7
1,2,3,4,6,7,8-HpCDD	2.6	3.9	8.4	4.3	4.9
OCDD	3.6	3.6	5.7	4.5	4.8

Table 6. Fortified concentration, mean percent recovery (n = 4), and percent RSD for spiked clean matrix.

	Aqueous			Solids			Tissues		
	Total Conc. (pg/L)	Mean % Recovery	RSD (%)	Total Conc. (pg/L)	Mean % Recovery	RSD (%)	Total Conc. (pg/g)	Mean % Recovery	RSD (%)
2,3,7,8-TCDD	200	99	2	20	102	2	20	102	1
1,2,3,7,8-PECDD	1,000	98	2	100	99	2	100	100	1
1,2,3,4,7,8-HXCDD	1,000	97	2	100	99	1	100	99	1
1,2,3,6,7,8-HXCDD	1,000	96	3	100	98	3	100	98	2
1,2,3,7,8,9-HXCDD	1,000	103	4	100	109	3	100	118	12
1,2,3,4,6,7,8-HPCDD	1,000	98	2	100	100	2	100	98	1
OCDD	2,000	98	2	200	100	2	200	99	1
2,3,7,8-TCDF	200	99	2	20	101	2	20	101	1
1,2,3,7,8-PECDF	1,000	97	2	100	100	2	100	100	1
2,3,4,7,8-PECDF	1,000	97	2	100	99	2	100	99	1
1,2,3,4,7,8-HXCDF	1,000	95	2	100	98	1	100	97	1
1,2,3,6,7,8-HXCDF	1,000	98	4	100	102	2	100	98	2
1,2,3,7,8,9-HXCDF	1,000	102	3	100	103	2	100	102	1
2,3,4,6,7,8-HXCDF	1,000	97	3	100	99	2	100	98	1
1,2,3,4,6,7,8-HPCDF	1,000	107	3	100	108	2	100	109	6
1,2,3,4,7,8,9-HPCDF	1,000	98	3	100	100	2	100	100	1
OCDF	2,000	92	2	200	97	2	200	94	3

Table 5. GC/TQ MDL results with comparison to Method 1613B MRLs.

Compound	Aqueous	Solid	Tissue
	MDL and (MRL) in pg/L	MDL and (MRL) in pg/g	MDL and (MRL) in pg/g
2,3,7,8-TCDD	1.1 (10)	0.029 (1)	0.057 (0.5)
1,2,3,7,8-PeCDD	1.39 (50)	0.037 (5)	0.051 (2.5)
1,2,3,4,7,8-HxCDD	1.05 (50)	0.042 (5)	0.061 (2.5)
1,2,3,6,7,8-HxCDD	1.08 (50)	0.045 (5)	0.033 (2.5)
1,2,3,7,8,9-HxCDD	1.78 (50)	0.064 (5)	0.067 (2.5)
1,2,3,4,6,7,8-HpCDD	1.19 (50)	0.070 (5)	0.032 (2.5)
OCDD	9.4 (100)	0.311 (10)	0.085 (5)
2,3,7,8-TCDF	0.56 (10)	0.60 (1)	0.056 (0.5)
1,2,3,7,8-PeCDF	1.0 (50)	0.037 (5)	0.046 (2.5)
2,3,4,7,8-PeCDF	1.25 (50)	0.039 (5)	0.033 (2.5)
1,2,3,4,7,8-HxCDF	0.89 (50)	0.032 (5)	0.029 (2.5)
1,2,3,6,7,8-HxCDF	1.11 (50)	0.031 (5)	0.046 (2.5)
1,2,3,7,8,9-HxCDF	1.22 (50)	0.048 (5)	0.084 (2.5)
2,3,4,6,7,8-HxCDF	1.26 (50)	0.026 (5)	0.034 (2.5)
1,2,3,4,6,7,8-HpCDF	0.92 (50)	0.255 (5)	0.064 (2.5)
1,2,3,4,7,8,9-HpCDF	1.35 (50)	0.028 (5)	0.043 (2.5)
OCDF	2.81 (100)	0.365 (10)	0.113 (5)

Recoveries

Three sets of spiked clean matrix one each of aqueous (1 L), solids (10 g) and tissues (10 g) were run and the mean percent recovery (n = 4) and percent RSD calculated (Figure 6). Results were compared and determined to conform to Method 1613B IPR specifications.

Proficiency, SRM, and CRM results

The evaluation report from Sigma-Aldrich RTC, Inc. concluded that both GC/HRMS and GC/TQ results obtained from the proficiency tests were acceptable and met study criteria and with an overall score of 100%. These results indicate the accuracy of PCDD/PCDF data from the 7010B Triple Quadrupole GC/MS analysis of the environmental matrices. The results of the GC/TQ analysis of the solids SRM (NIST 1944) and tissue CRM (EDF 2525) also demonstrated the accuracy of the GC/TQ method.

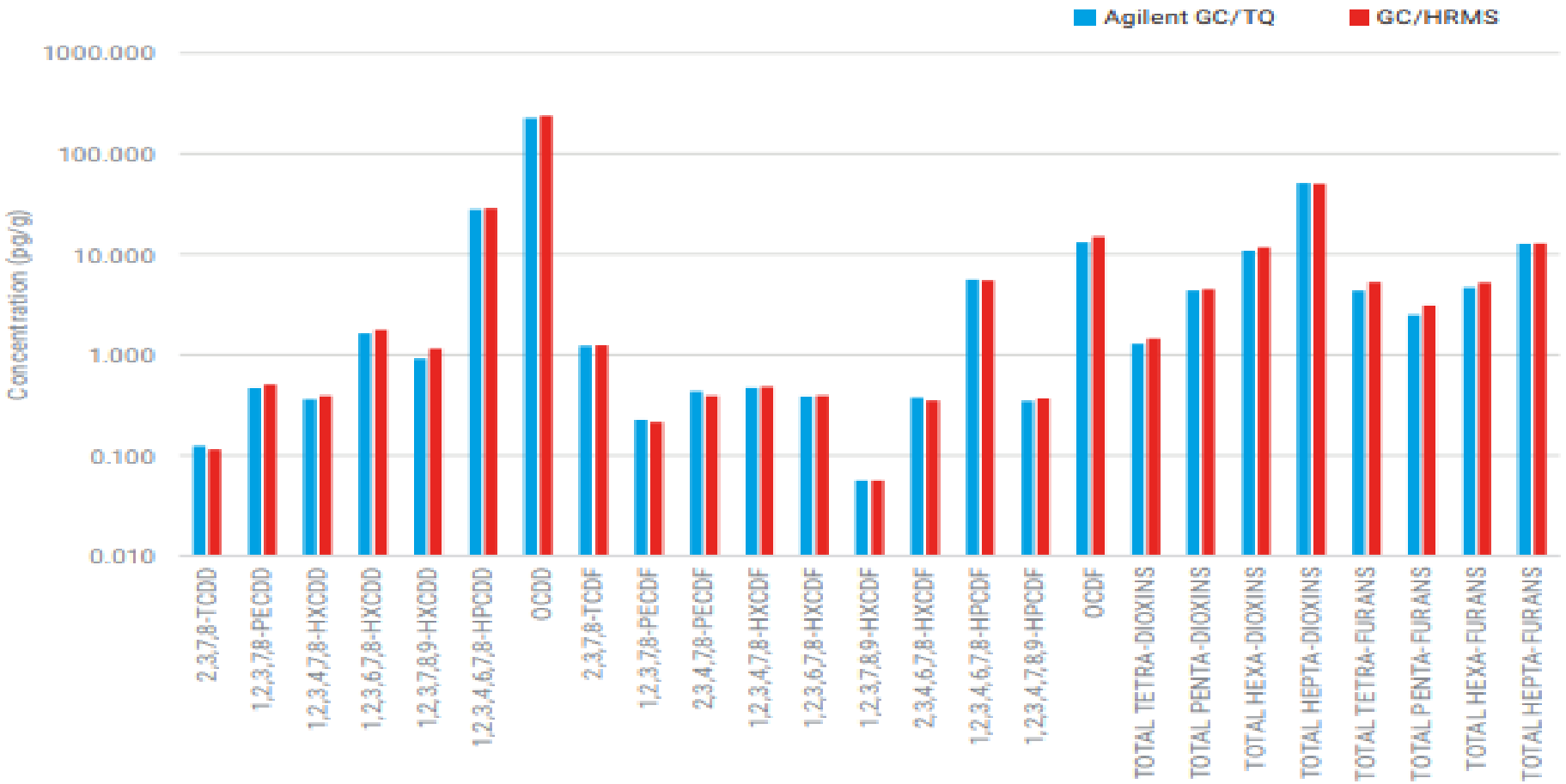
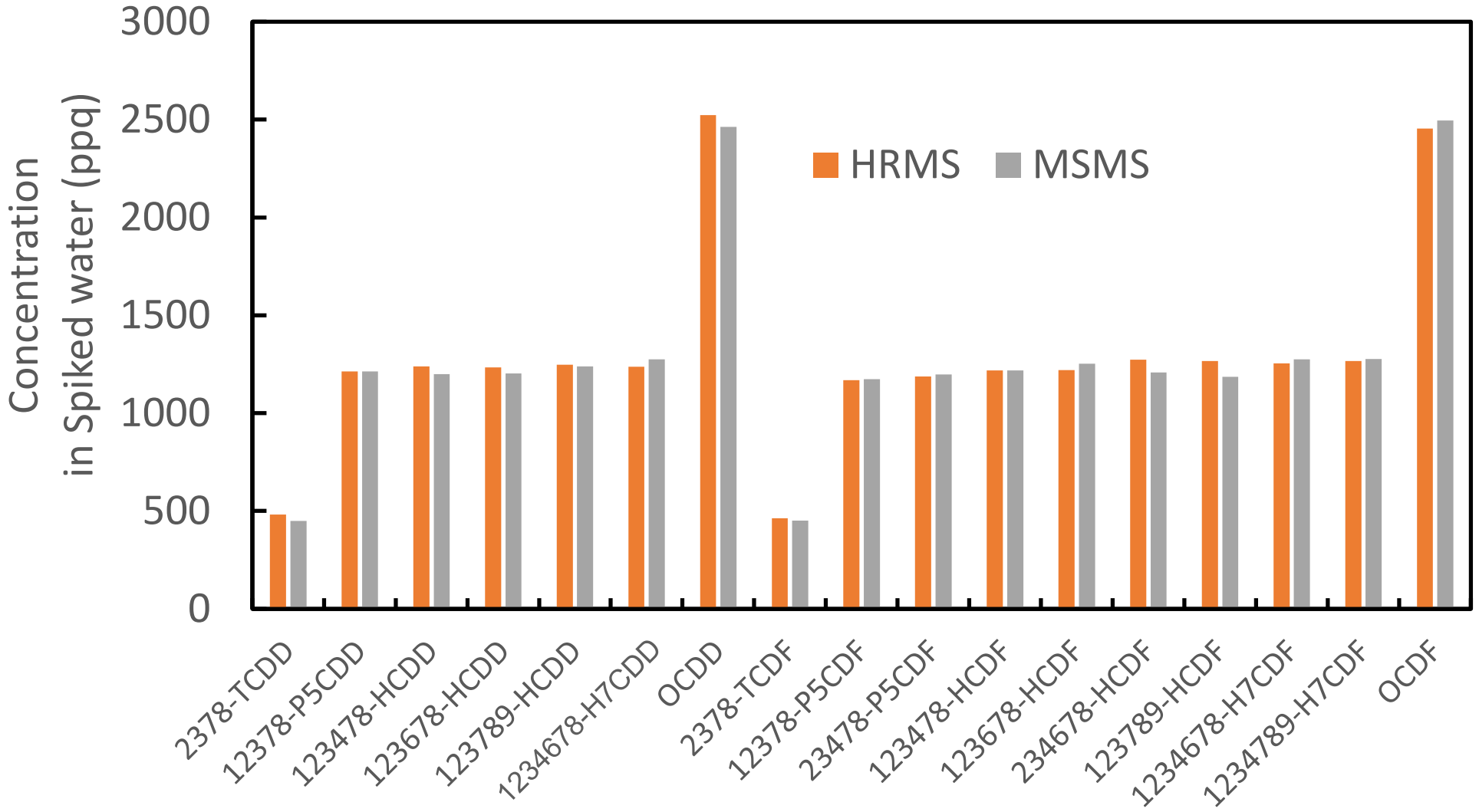


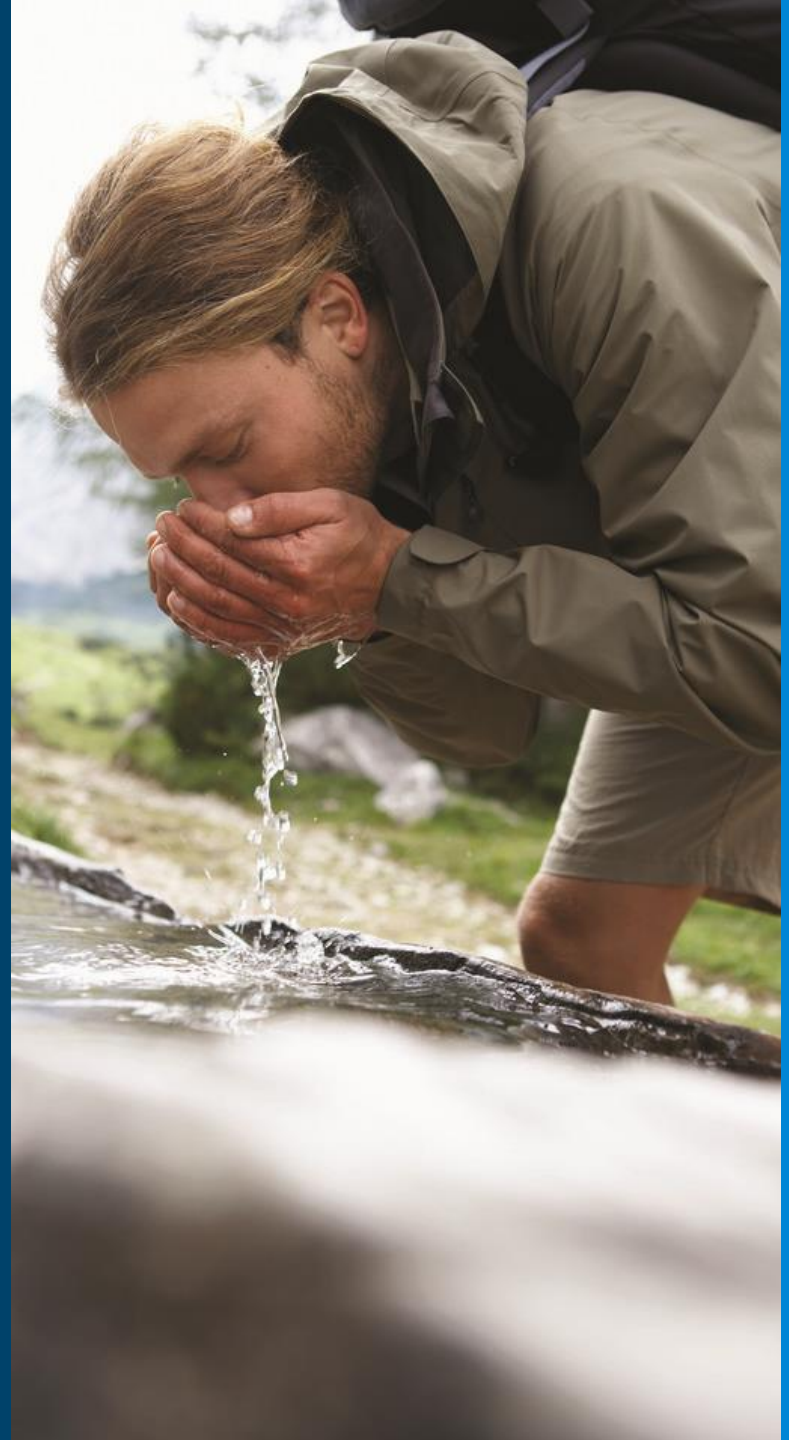
Figure 4. Comparison of total PCDD/PCDF for a real-world biosolids sample determined by GC/TQ (blue bars) and GC/HRMS (red bars).

Spiked Water Sample (GC/HRMS vs GC/MS/MS)

Similar concentration detected in water sample on both systems!



How did we get here from there



Agilent
Trusted Answers

How did we get here from there

Three years of a combined effort has gone into this process. I would like to thank the following

SGS AXYS Sidney B.C Canada

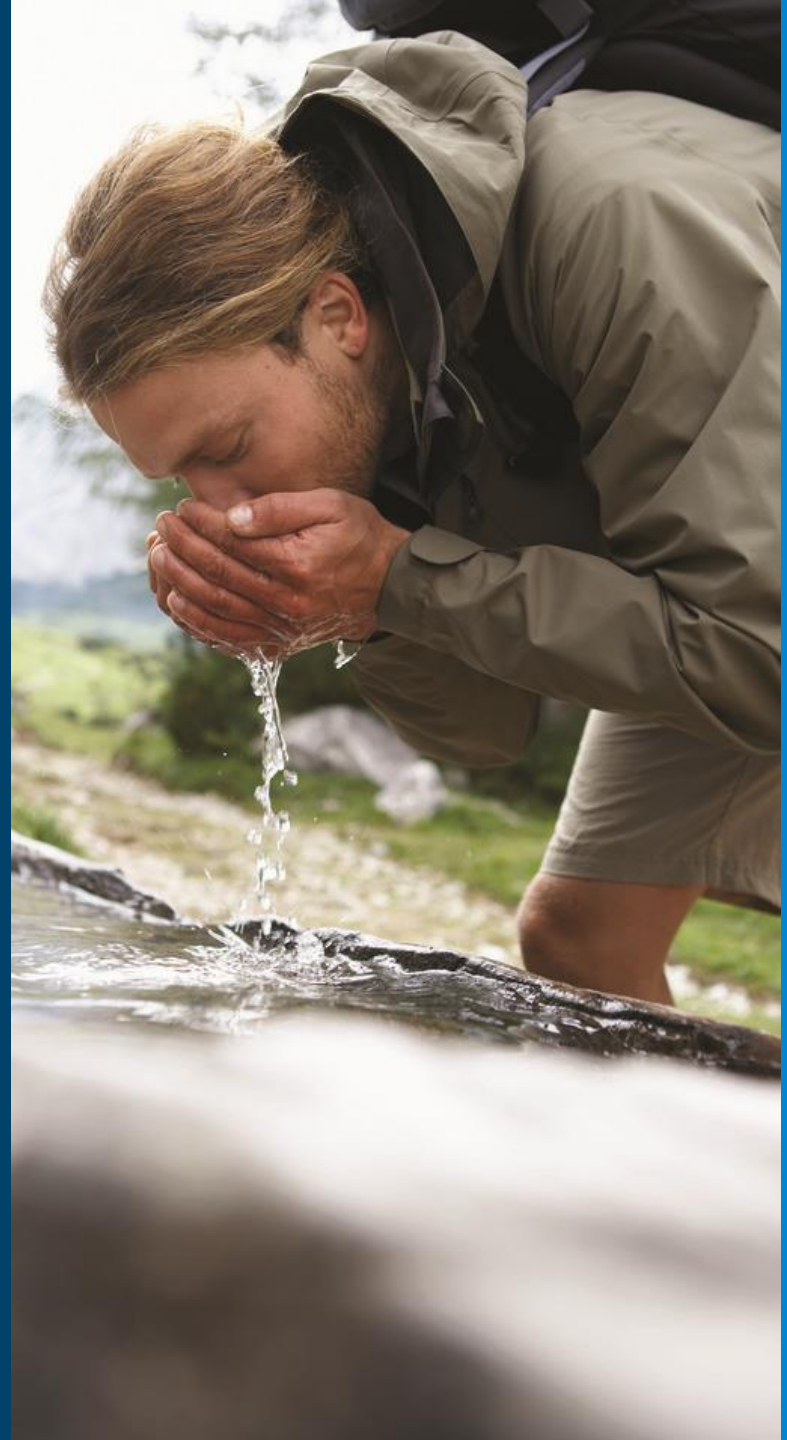
Pace Analytical Minneapolis Minnesota, U.S.A

The Dow Chemical Company, Michigan, USA

U.S EPA Region 4 Athens Georgia U.S.A.

U.S EPA Office Of Water Washington D.C U.S.A.

Without their help and support we would not have been able to establish the protocol for meeting method 16130 by GC/MS/MS



Agilent

Trusted Answers

How did we get here from there

Dale R. Walker
GC/MS/MS Application Scientist

Fred Feyerherm
GC/MS/MS Application Scientist

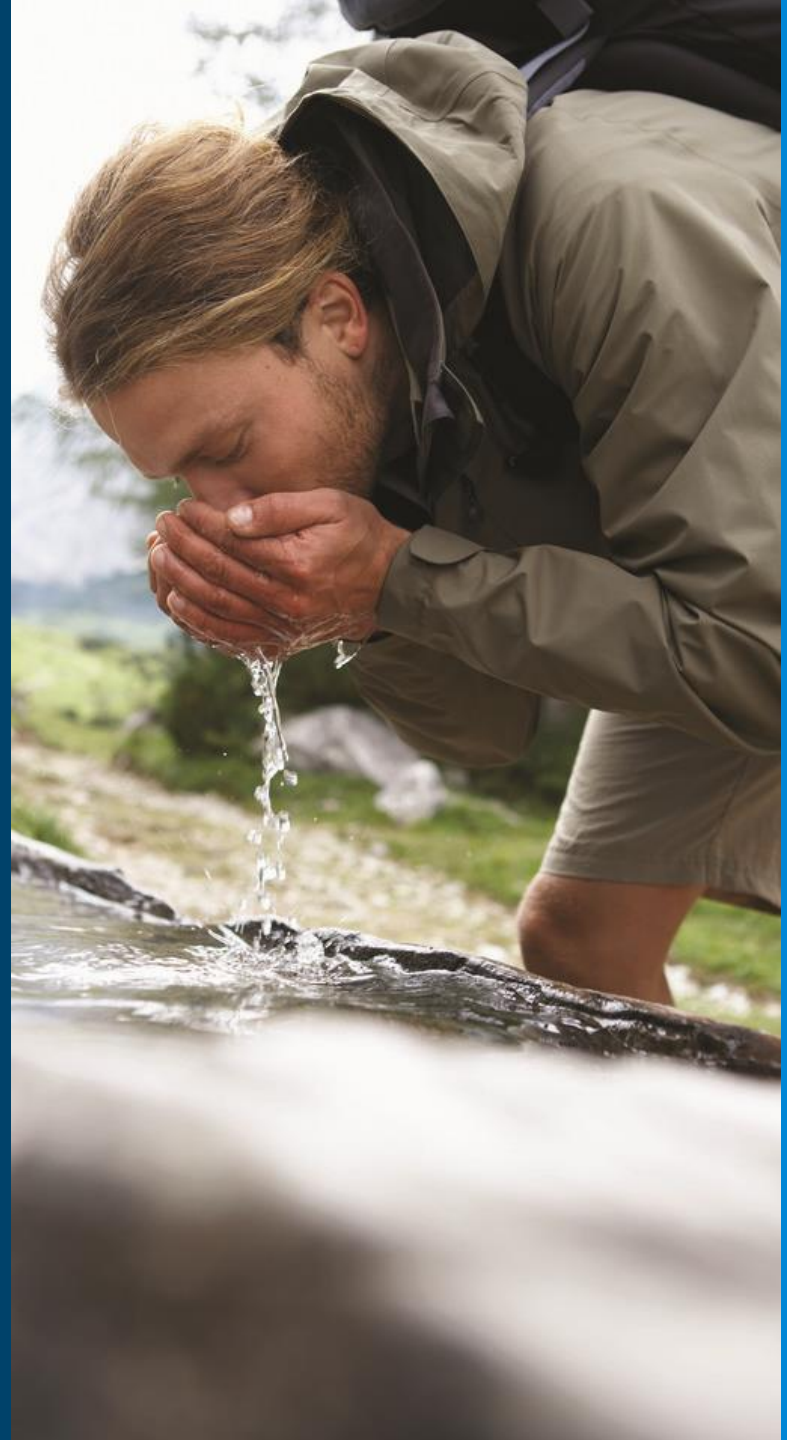
Anastasia Andrianova
GCMS Application Scientist

Harry Prest
GC/MS R&D Chemist

Diana Wong
GC/MS Application Scientist

Tarun Anumol, Ph.D.
Director, Global Environment & Food Markets

Craig Marven
Director, Global Environment & Food Markets



Agilent
Trusted Answers

SGS AXYS Method 16130

Determination of 2,3,7,8-Substituted Tetra- through Octa-Chlorinated Dibenzo-*p*-Dioxins and Dibenzofurans (CDDs/CDFs) Using Waters and Agilent Gas Chromatography Mass Spectrometry (GC-MS/MS)

Revision 1.0

1.0 Scope and Application

1.1 This method is for determination of tetra- through octa-chlorinated dibenzo-*p*-dioxins (CDDs) and dibenzofurans (CDFs) in water, soil, sediment, sludge, tissue, and other sample matrices by gas chromatography coupled with a tandem quadrupole mass spectrometry system (GC-MS/MS). The method is for use in EPA's data gathering and monitoring programs associated with the Clean Water Act, the Resource Conservation and Recovery Act, the Comprehensive Environmental Response, Compensation and Liability Act, and the Safe Drinking Water Act. The method is based on a compilation of EPA, industry, commercial laboratory, and academic methods (References 1-7).

1.2 The seventeen 2,3,7,8-substituted CDDs/CDFs listed in Table 1 may be determined by this method. Specifications are also provided for separate determination of 2,3,7,8-tetrachloro-dibenzo-*p*-dioxin (2,3,7,8-TCDD) and 2,3,7,8-tetrachloro-dibenzofuran (2,3,7,8-TCDF).

1.3 The detection limits and quantitation levels in this method are usually dependent on the level of interferences rather than instrumental limitations. The minimum levels (MLs) in Table 2 are the levels at which the CDDs/CDFs can be determined with no interferences present. The Method Detection Limit (MDL) for 2,3,7,8-TCDD has been determined as 0.9 pg/L (parts-per-quadrillion) using this method and the Waters APGC-MS/MS system and as 1.1 pg/L using this method and the Agilent GC-MS/MS system.

1.4 The GC-MS/MS portions of this method are for use only by analysts experienced with tandem quadrupole mass spectrometry systems or under the close supervision of such qualified persons. Each laboratory that uses this method must demonstrate the ability to generate acceptable results using the procedure in Section 9.2.

1.5 This method is "performance-based". The analyst is permitted to modify the method to overcome interferences or lower the cost of measurements, provided that all performance criteria in this method are met. The requirements for establishing method equivalency are given in Section 9.1.2.

1.6 Any modification of this method, beyond those expressly permitted, shall be considered a major modification subject to application and approval of alternate test procedures under 40 CFR 136.4 and 136.5.

UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

OFFICE OF WATER

Coreen Hamilton, Ph.D.
Senior Scientific Specialist
SGS AXYS Analytical Services, Ltd.
2045 Mills Road West
Sidney, BC V8L 5X2
Canada

SUBJECT: Review of SGS AXYS Method 16130, Determination of 2,3,7,8-Substituted Tetra- through Octa-Chlorinated Dibenzo-*p*-Dioxins and Dibenzofurans (CDDs/CDFs) Using Waters and Agilent Gas Chromatography Mass Spectrometry (GC-MS/MS)
(ATP Case No. N18-0003)

DATE: September 17, 2020

I have reviewed SGS AXYS Method 16130 (ATP Case No. N18-0003), "Determination of 2,3,7,8-Substituted Tetra- through Octa-Chlorinated Dibenzo-*p*-Dioxins and Dibenzofurans (CDDs/CDFs) Using Waters and Agilent Gas Chromatography Mass Spectrometry (GC-MS/MS)," and the supporting validation data in ATP Case No. N18-0003. I determined that this method meets all requirements for measurement of 2,3,7,8-substituted tetra- through octa-chlorinated dibenzo-*p*-dioxins and dibenzofurans (PCDDs/PCDFs) in wastewater. That is, the performance of this method is substantially similar to methods listed at 40 CFR Part 136 for measurement of PCDDs/PCDFs in wastewater.

Based on the attached justification and the performance of SGS AXYS Method 16130, I will recommend that this method be included in future regulatory actions in which EPA adds to the list of approved methods at 40 CFR Part 136. However, this ATP review does not replace the normal notice-and-comment rulemaking process. In the interim, a user may, on a facility-by-facility basis, seek approval from their regional authority for use of this method in measuring PCDDs/PCDFs in wastewater in Clean Water Act (CWA) programs.

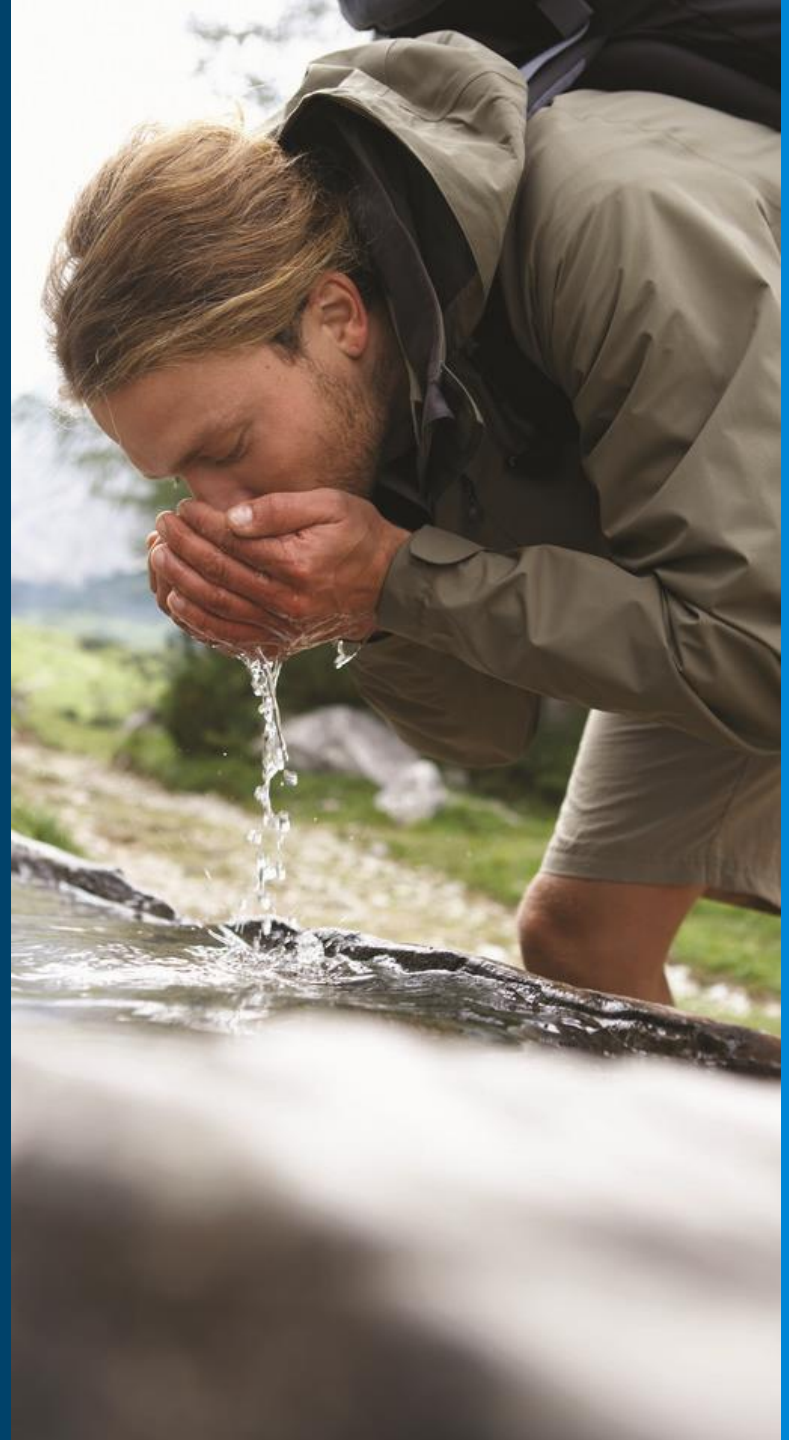
If I can be of any additional assistance on this matter or others, please contact me at walker.lemuel@epa.gov .

Sincerely,

Lemuel Walker
CWA ATP Coordinator
Technology and Analytical Support Branch
Engineering and Analysis Division
Office of Science and Technology
Office of Water

cc: Tarun Anumol – Agilent Technologies
Frank Dorman – Waters Corporation
Quality Assurance Managers (all Regions)
ATP Coordinators (all Regions)

Questions ?



Agilent
Trusted Answers