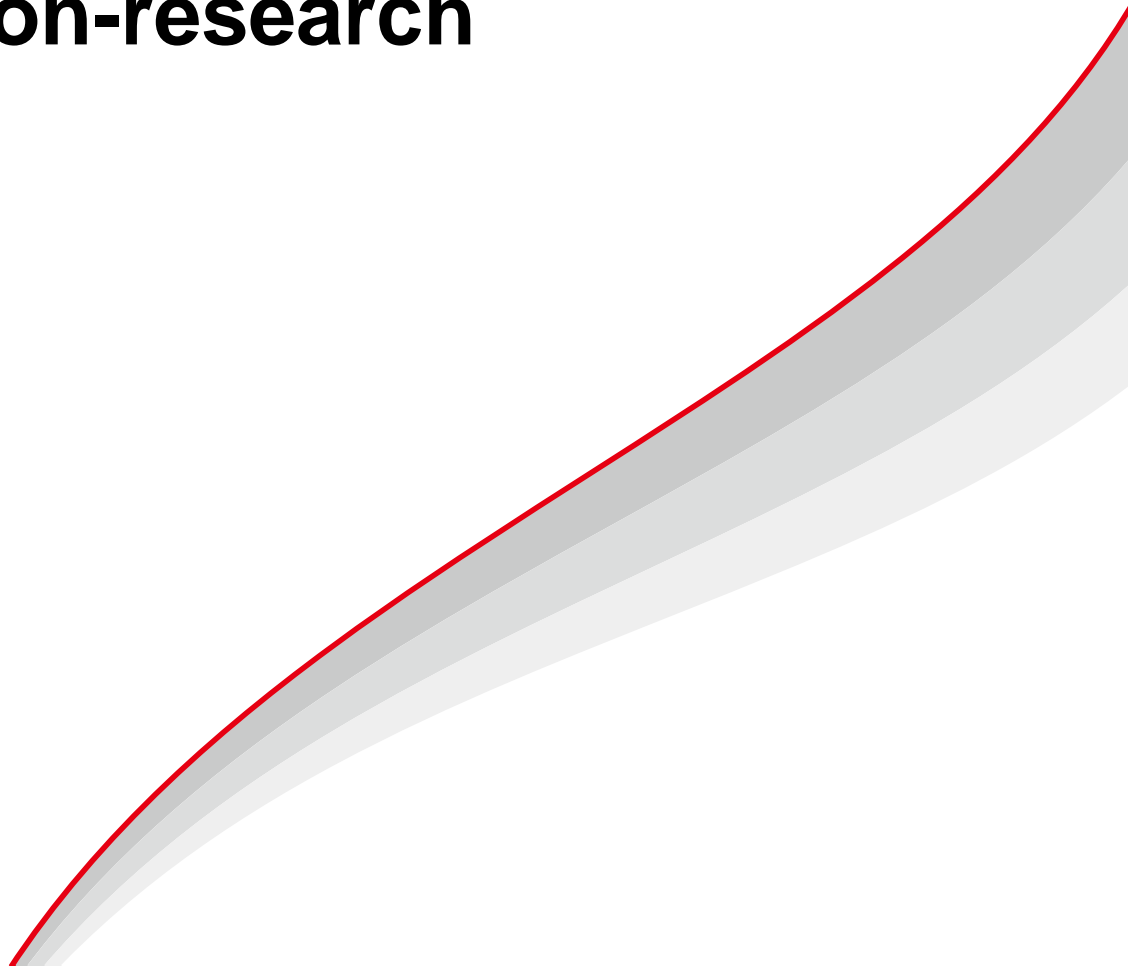


Monitoring of PFAS by LC-QTOF: Streamlined Workflow for the Non-research Environmental Laboratories

Ethan Hain, PhD

Ruth Marfil-Vega, PhD

August 1st , 2023



Today's presentation



1. HRMS for non-R&D labs
2. Experimental plan
3. Results
4. Take-home messages
5. Q&A

HRMS for the non-research labs

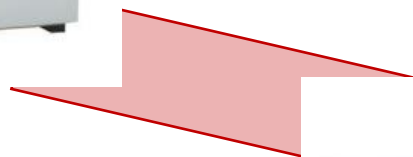
- High resolution mass spectrometry (HRMS) instruments are the best tool for analyzing unknown and suspected PFAS;
- Data processing required for extracting meaningful results has traditionally been the bottleneck of analytical workflows:
Non-research laboratories:
 - quick turn-around times are required
 - HRMS experts may not be present

Easy workflows that enable screening and quantitation are essential

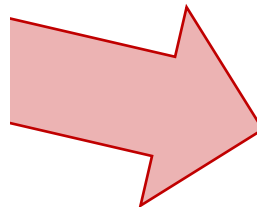
LCMS-9030 and LCMS-9050



LCMS IT-TOF
(2004)



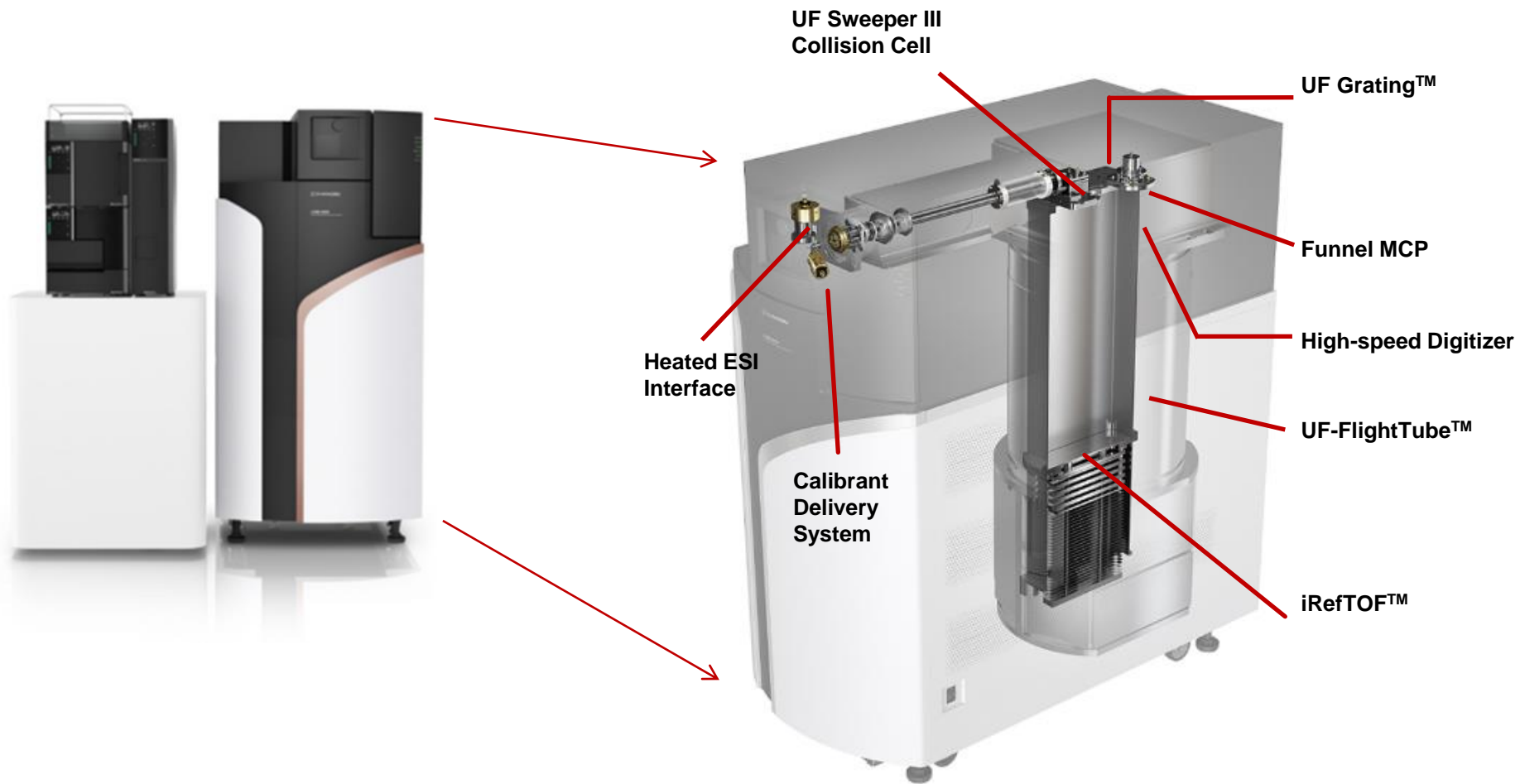
LCMS-9030
(QTOF)



LCMS-9030
(QTOF)

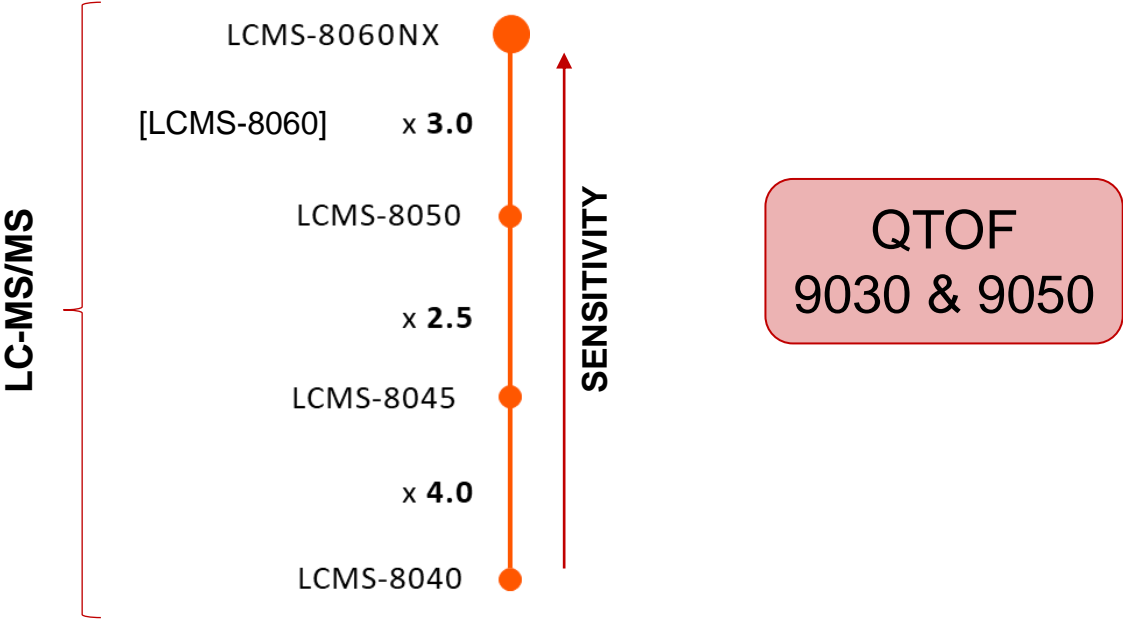


LCMS-9030 and LCMS-9050



PFAS and the quest for sensitivity

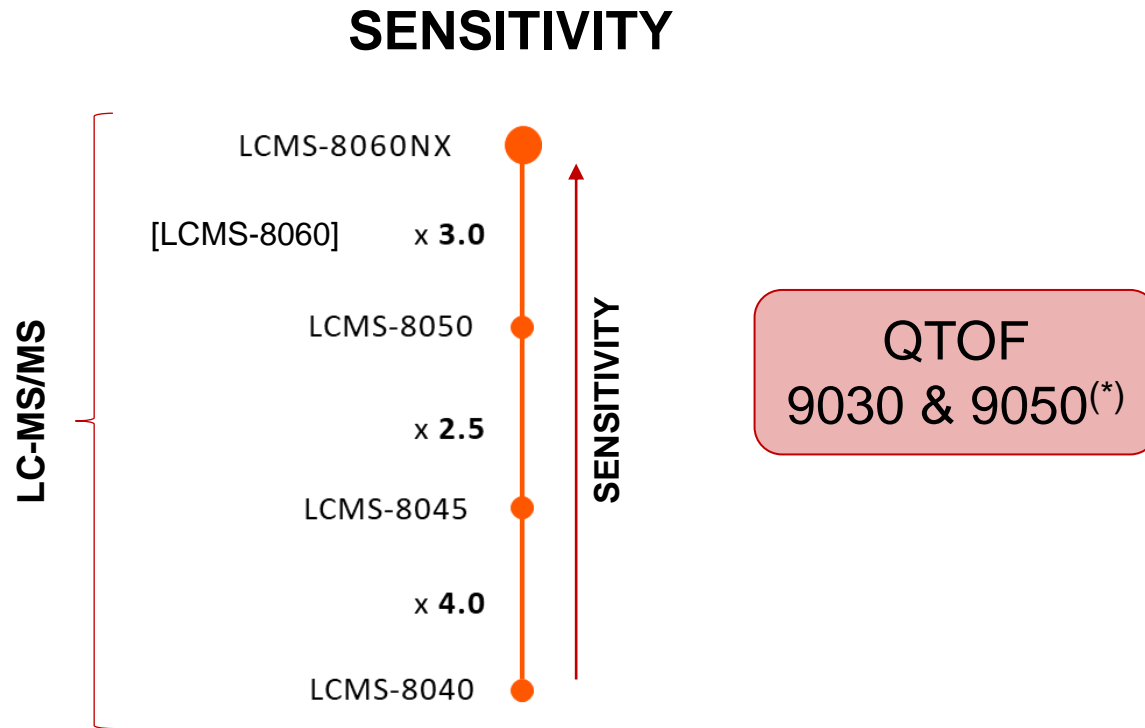
SENSITIVITY



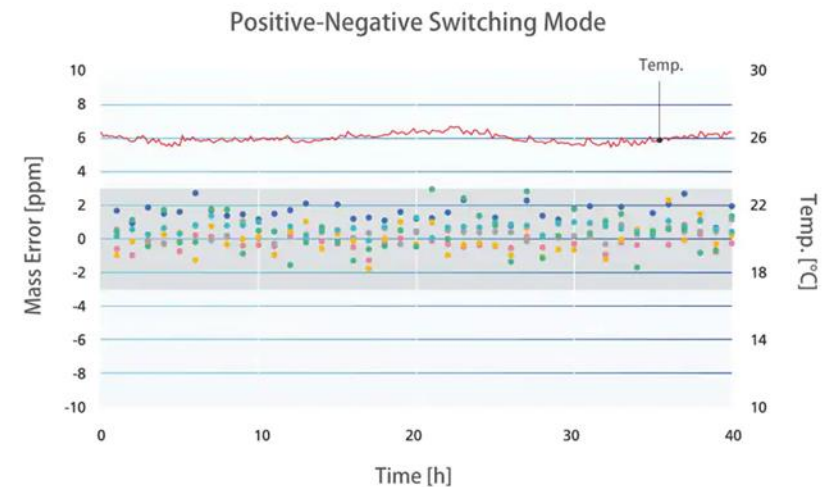
Compound	Proposed MCLG	Proposed MCL (enforceable levels)
PFOA	Zero	4.0 parts per trillion (also expressed as ng/L)
PFOS	Zero	4.0 ppt
PFNA	1.0 (unitless) Hazard Index	1.0 (unitless) Hazard Index
PFHxS		
PFBS		
HFPO-DA (commonly referred to as GenX Chemicals)		

All LCMS manufacturers offer instruments designed to achieve different sensitivity

PFAS and other parameters of interest



- Polarity Switching (fast)
- Acquisition Rate (fast)
- Mass accuracy (stable long term)



(*)LCMS-9050: pol switching 800 ms; 200 Hz (200 MS/MS spectra per second)

All LCMS manufacturers offer instruments designed to achieve different sensitivity

Experimental plan

Goal: To validate an accessible workflow for the analysis of known and suspected PFAS, using targets included in EPA 1633 as reference.

Nexera LC		LCMS-9030	
Flow Rate:	0.25 mL/min	Nebulizing Gas:	3 L/min
Oven Temp.:	45 °C	Drying Gas:	5 L/min
Injection Vol.:	1 µL	Heating Gas:	15 L/min
Mobile Phase:	A: 5 mM Ammonium acetate in water B: Methanol	Desolvation Temp.:	160 °C
Delay Column:	Shim-pack Scepter C18-120 (2.1 × 50 mm; 3 µm)	DL Temp.:	150 °C
Analytical Column:	Shim-pack Scepter C18-120 (2.1 × 100 mm; 3 µm)	Heat Block Temp.:	250 °C
Gradient:	0 min %B = 5; 1 min %B = 40; 8 min %B = 95; 8.1 min %B = 100; 13 min %B = 100; 13.1 min %B = 5; 18 min %B = 5	Interface Temp:	100 °C
		Probe Position:	+1 mm
		Interface:	ESI

Experimental plan

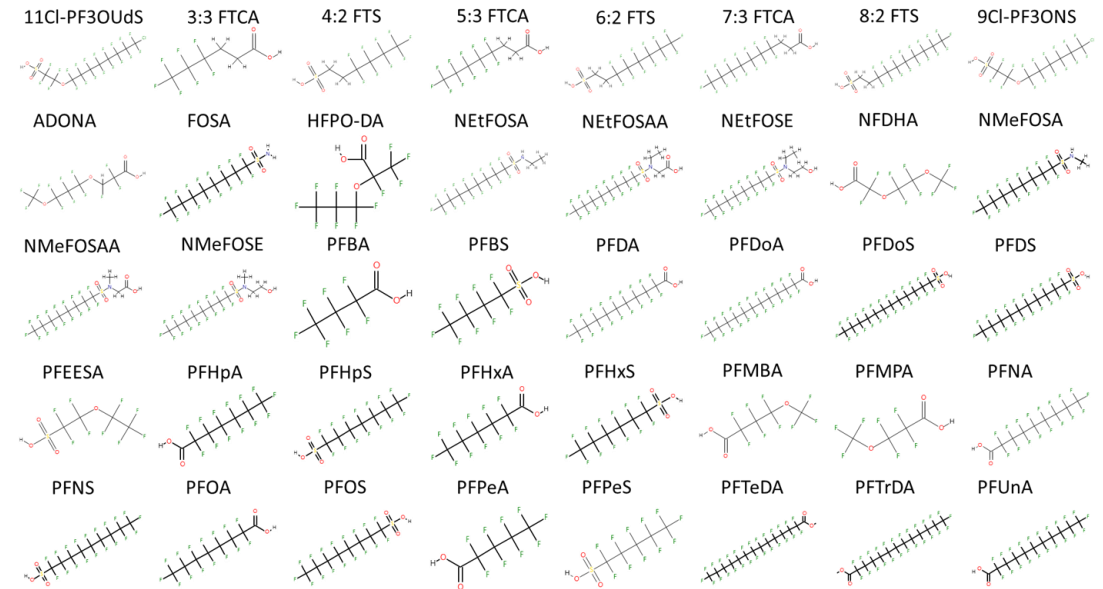
Goal: To validate an accessible workflow for the analysis of known and suspected PFAS, using targets included in EPA 1633 as reference.

Step 1 – Library creation and curation

Step 2 – Suspects screening

Step 3 – Quantitation

LabSolutions and LabSolutions Insight Explore software were used to acquire and process the data



Library Creation and Curation

1. Acquire MS1 scan of EPA 1633 Standard Mix to identify precursors and RT
2. Acquire MS/MS spectra to create library entries:
 - CE 5-55 V
 - CE 15-65 V
3. Library curation:
 - 1) With Assign:
 - confirm all fragments match theoretical fragmentation, and
 - realign fragmentation patternOther open-source resources can be used for fragmentation matching
 - 2) With Explore: confirm m/z of fragments, precursor and RT

Suspects Screening

- EPA 1633 standard mix spiked into ground water, known to be contaminated with several classes of organic compounds
- Data acquired using Data Independent Acquisition (DIA) mode
- Results evaluated against the in-house curated library

Targets Quantification

- Based on EPA 537.1
- Comparison of LCMS-9030 and LCMS-8045

Table 1: LC parameters

LC System	Nexera-X2 UHPLC System
Analytical Column	Shim-pack™ Velox , 150mm x 2.1mm x 2.7µm, Part No. 227-320094-04
Solvent Delay Column	Shim-pack XR-ODS 50mm x 2mm x 2.2µm, Part No. 228-41605-93
Column Temp.	40 °C
Injection Volume	5 µL
Mobile Phase	A: 20 mM Ammonium Acetate B: Methanol
Flow Rate	0.25 mL/min
Run Time	35 minutes

Table 2: LCMS parameters

MS Instrument	LCMS-8045 and LCMS-9030
Interface	Electrospray Ionization (ESI) Negative mode
Interface Temp.	100 °C
Desolvation Line Temp.	100 °C
Heat Block Temp.	200 °C
Heating Gas Flow	15 L/min
Drying Gas Flow	5 L/min
Nebulizing Gas Flow	3 L/min
Total MRMs	48

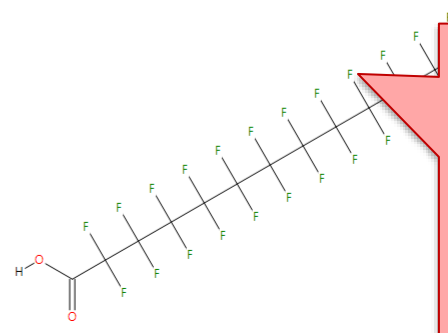
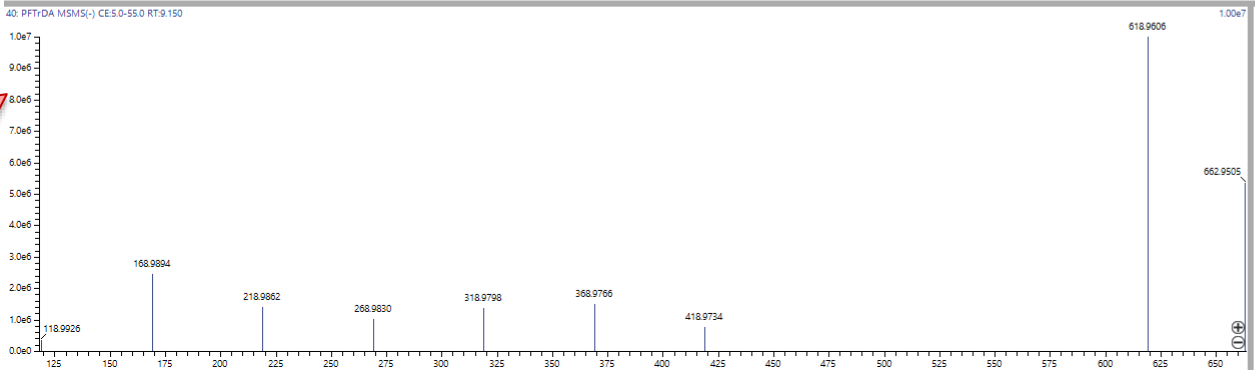
Results – PFAS Library Creation and Curation

- Shortcuts
- File
- View
- Edit
- Edit Method
- Integrate Batch
- Integrate Sample
- Integrate Compound
- Integrate Result
- Edit Tables
- Edit Flags
- Select Compounds
- Edit Library
- Review
- Report
- Explore

Library information: CAS #, formula, RT, and CE spread

#	CAS #	Compound Name	Theory MW	Formula	Structure	Class	Comment	Column	RT	Collision Energy	Precursor Ion	Precursor m/z	MS Stage	Ionization	Mass Range	Instrument	Sample Info	Polarity
14	2991-50-6	NEFOSAA	584.990300	C12H8F17NO4S	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	8.569	5.0 - 55.0	[M-H]-	583.983000	2	ESI	168.989370 - 58...	LCMS-9030	EPAM1633CaI7	negative
15	1691-99-2	NEFOSE	571.011000	C12H10F17NO3...	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	9.513	5.0 - 55.0	[M+CH3COO]-	630.024900	2	ESI	41.383240 - 634...	LCMS-9030	EPAM1633CaI7	negative
16	151772-58-6	NFDHA	295.973100	C5HF9O4	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	5.809	5.0 - 55.0	[M-H]-	294.965800	2	ESI	84.990670 - 294...	LCMS-9030	EPAM1633CaI7	negative
17	31506-32-8	NMeFOSA	512.969100	C9H4F17NO2S	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	9.345	5.0 - 55.0	[M-H]-	511.961900	2	ESI	168.989370 - 51...	LCMS-9030	EPAM1633CaI7	negative
18	2355-31-9	NMeFOSAA	570.974600	C11H6F17NO4S	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	8.388	5.0 - 55.0	[M-H]-	569.967300	2	ESI	168.989370 - 56...	LCMS-9030	EPAM1633CaI7	negative
19	24448-09-7	NMeFOSE	556.995300	C11H8F17NO3S	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	9.312	5.0 - 55.0	[M+CH3COO]-	616.009200	2	ESI	616.007700 - 61...	LCMS-9030	EPAM1633CaI7	negative
20	375-22-4	PFBA	213.986500	C4HF7O2	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	4.005	5.0 - 55.0	[M-H]-	212.979200	2	ESI	168.989370 - 21...	LCMS-9030	EPAM1633CaI7	negative
21	375-73-5	PFBS	299.950300	C4HF9O3S	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	5.200	5.0 - 55.0	[M-H]-	298.943000	2	ESI	79.957360 - 298...	LCMS-9030	EPAM1633CaI7	negative
22	335-76-2	PFDA	513.967300	C10HF19O2	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	8.193	5.0 - 55.0	[M-H]-	512.960000	2	ESI	168.989370 - 51...	LCMS-9030	EPAM1633CaI7	negative
23	307-55-1	PFDaA	613.969900	C12HF23O2	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	8.862	5.0 - 55.0	[M-H]-	612.953700	2	ESI	118.992560 - 61...	LCMS-9030	EPAM1633CaI7	negative
24	79780-39-5	PFDoS	699.924700	C12HF25O3S	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	9.102	5.0 - 55.0	[M-H]-	698.917400	2	ESI	98.955770 - 698...	LCMS-9030	EPAM1633CaI7	negative
25	335-77-3	PFDS	599.931100	C10HF21O3S	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	8.513	5.0 - 55.0	[M-H]-	598.923800	2	ESI	98.955770 - 598...	LCMS-9030	EPAM1633CaI7	negative
26	113507-82-7	PFEESA	315.945200	C4HF9O4S	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	5.563	5.0 - 55.0	[M-H]-	314.937900	2	ESI	82.960850 - 314...	LCMS-9030	EPAM1633CaI7	negative
27	375-85-9	PFHpA	363.976900	C7HF13O2	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	6.642	5.0 - 55.0	[M-H]-	362.969600	2	ESI	118.992560 - 36...	LCMS-9030	EPAM1633CaI7	negative
28	375-92-8	PFHpS	449.940700	C7HF15O3S	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	7.252	5.0 - 55.0	[M-H]-	448.933400	2	ESI	98.955770 - 448...	LCMS-9030	EPAM1633CaI7	negative
29	307-24-4	PFHxA	313.980100	C6HF11O2	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	5.937	5.0 - 55.0	[M-H]-	312.972800	2	ESI	118.992560 - 31...	LCMS-9030	EPAM1633CaI7	negative
30	355-46-4	PFHxS	399.943900	C6HF13O3S	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	6.669	5.0 - 55.0	[M-H]-	398.936600	2	ESI	98.955770 - 398...	LCMS-9030	EPAM1633CaI7	negative
31	863090-89-5	PFMBA	279.978200	C5HF9O3	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	5.353	5.0 - 55.0	[M-H]-	278.970900	2	ESI	84.990670 - 278...	LCMS-9030	EPAM1633CaI7	negative
32	377-73-1	PFMPA	229.981400	C4HF7O3	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	4.425	5.0 - 55.0	[M-H]-	228.974100	2	ESI	84.990670 - 228...	LCMS-9030	EPAM1633CaI7	negative
33	375-95-1	PFNA	463.970500	C9HF17O2	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	7.765	5.0 - 55.0	[M-H]-	462.963200	2	ESI	118.992560 - 46...	LCMS-9030	EPAM1633CaI7	negative
34	68259-12-1	PFNS	549.934300	C9HF19O3S	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	8.152	5.0 - 55.0	[M-H]-	548.927000	2	ESI	98.955770 - 548...	LCMS-9030	EPAM1633CaI7	negative
35	335-67-1	PFOA	413.973700	C8HF15O2	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	7.258	5.0 - 55.0	[M-H]-	412.966400	2	ESI	168.989370 - 41...	LCMS-9030	EPAM1633CaI7	negative
36	1763-23-1	PFOS	499.937500	C8HF17O3S	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	7.730	5.0 - 55.0	[M-H]-	498.930200	2	ESI	98.955770 - 498...	LCMS-9030	EPAM1633CaI7	negative
37	2706-90-3	PFPeA	263.983300	C5HF9O2	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	5.075	5.0 - 55.0	[M-H]-	262.976000	2	ESI	118.986180 - 26...	LCMS-9030	EPAM1633CaI7	negative
38	2706-91-4	PFPeS	349.947100	C5HF11O3S	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	6.004	5.0 - 55.0	[M-H]-	348.939800	2	ESI	79.957360 - 348...	LCMS-9030	EPAM1633CaI7	negative
39	376-06-7	PFTdA	713.954500	C14HF27O2	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	9.392	5.0 - 55.0	[M-H]-	712.947300	2	ESI	118.992560 - 71...	LCMS-9030	EPAM1633CaI7	negative
40	72629-94-8	PFTdA	663.957700	C13HF25O2	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	9.150	5.0 - 55.0	[M-H]-	662.950500	2	ESI	118.992560 - 66...	LCMS-9030	EPAM1633CaI7	negative
41	2058-94-8	PFUnA	563.964100	C11HF21O2	[MOL]	Fluorinated ...	Delay Column: ...	Shim-pack Sce...	8.553	5.0 - 55.0	[M-H]-	562.956800	2	ESI	118.992560 - 56...	LCMS-9030	EPAM1633CaI7	negative

MS/MS spectrum of the highlighted library compound



Structure of the highlighted library compound

Results – Suspects Screening *After running Library Search*

LabSolutions Insight (Admin) - (EPA1633_MS_DIA_CE5-55.damp - EPA1633_MS_DIA_CE5-55.lcm) [Pre-Release]

Sample List

#	Flags	Data Filename	Sample Name	Flag ID	Sample Type	Vial
1		GW 1510_Cal 7_EPA1633_MS_DIA_CE5-55_0...	GW 1:10	ppm	Unknown	7
2		EPA 1633_Cal 7_EPA1633_MS_DIA_CE5-55_...	EPA 1633		Unknown	9
3		GW 151_Cal 7_EPA1633_MS_DIA_CE5-55_004	GW 1:1		Unknown	8
4		GW 151_Cal 7_EPA1633_MS_DIA_CE5-55_003	GW 1:1		Unknown	8
5		Blank_Null_EPA1633_MS_DIA_CE5-55_001	Blank	mDa, ppm	Unknown	-1

Compound Results - GW 151_Cal 7_EPA1633_MS_DIA_CE5-55_003

#	Name	Formula	m/z	Found Mz	Mass Error (ppm)	Found RT	RT Diff	Lib. SI	CAS #	Mass E
1	11CI-PF3OUdS	C10HCIF20O4S	630.8892	630.88761	-2.504	8.726	0.041	100	763051-92-9	
2	3:3 FTCA	C6H5F7O2	241.0105	241.00953	-4.025	5.143	0.024	94	356-02-5	
3	4:2 FTS	C6H5F9O3S	326.9743	326.97322	-3.272	5.909	0.028	94	757124-72-4	
4	5:3 FTCA	C8H5F11O2	341.0041	341.00342	-2.023	6.847	0.033	97	914637-49-3	
5	6:2 FTS	C8H5F13O3S	426.9679	426.96701	-2.084	7.288	0.052	100	27619-97-2	
6	7:3 FTCA	C10H5F15O2	440.9977	440.99665	-2.449	7.966	0.038	98	812-70-4	
7	8:2 FTS	C10H5F17O3S	526.9615	526.96035	-2.220	8.235	0.033	99	39108-34-4	
8	9CI-PF3ONS	C8HCIF16O4S	530.8956	530.89452	-1.997	8.011	0.035	94	756426-58-1	
9	ADONA	C7H2F12O4	376.9689	376.96799	-2.334	6.779	0.032	100	919005-14-4	
10	FOSA	C8H2F17NO2S	497.9462	497.94495	-2.510	8.638	0.015	100	754-91-6	
11	HFPO-DA	C6HF11O3	284.9779	284.97721	-2.421	6.221	0.037	97	13252-13-6	
12	NHFOA	C10H4F17NO2S	525.9775	525.97610	-2.662	9.564	0.112	100	4151-50-2	

Mass Spectrometry Plots

Library Search Results

#	Name	Formula	Mass
1	4,4,5,5,6,6,7,7,8,...	C8H5F11O2	342.01139
2	ethyl 2,2,3,3,4,4,...	C8H5F11O2	342.01139
3	1-(2,2-difluoro...	C8H5F11O2	342.01139
4	1,1,1,2,3,3-hexa...	C8H5F11O2	342.01139
5	1,1,1,2,3,3-hexa...	C8H5F11O2	342.01139
6	5-(1,1-difluoro...	C8H5F11O2	342.01139
7	2-[[1,1,2,3,4,4,...	C8H5F11O2	342.01139
8	4,5,5,5-tetraflu...	C8H5F11O2	342.01139
9	2-ethyl-2,3,5,5-	C8H5F11O2	342.01139

Compound Details

Assign - 11:MSMS(-)[337.5000] CE:5.0-55.0 RT:[6.830-6.860]-[6.755-7.115] - 4,4,5,5,6,6,7,7,8,8-undecafluorooctanoic acid

4,4,5,5,6,6,7,7,8,8-undecafluorooctanoic acid

#	Formulae (M)	m/z	Pred. m/z	Intensity	+/- ppm	+/- mDa	Charge	Depth	Structures
10	C7HO2 C2HO...	116.99773	116.99820	270	-4.017	-0.47	----	----	----
11	C2F5	118.99153	118.99256	116	-8.656	-1.03	-	1	1
12	C4HO2F2	118.99153	118.99501	116	-29.245	-3.48	[-H]-	2	1
13	C4H2OF3	123.00748	123.00632	120	9.430	1.16	[-H]-	2	1

Sample information such as file names, sample names, sample type, and flags are shown in the sample list.

Library Search

Results – Suspects Screening

LabSolutions Insight (Admin) - (EPA1633_MS_DIA_CE5-55.damlp - EPA1633_MS_DIA_CE5-55.lcm) [Pre-Release]

Shortcuts

- File
- View
- Edit
- Edit Method
- Integrate Batch
- Integrate Sample
- Edit Tables
- Edit Flags
- Select Compounds
- Edit Library
- Review
- Report
- Explore

Sample List						Compound Results - GW 151_Cal 7_EPA1633_MS_DIA_CE5-55_003										
#	Flags	Data Filename	Sample Name	Flag ID	Sample Type	#	Name	Formula	m/z	Found Mz	Mass Error (ppm)	Found RT	RT Diff	Lib. SI	CAS #	Mass E
1		GW 1510_Cal 7_EPA1633_MS_DIA_CE5-55_0...	GW 1:10		Unknown	1	11CI-PF3OUdS	C10HCIF20O4S	630.8892	630.88761	-2.504	8.726	0.041	100	763051-92-9	
2		EPA 1633_Cal 7_EPA1633_MS_DIA_CE5-55_...	EPA 1633		Unknown	2	3:3 FTCA	C6H5F7O2	241.0105	241.00953	-4.025	5.143	0.024	94	356-02-5	
3		GW 151_Cal 7_EPA1633_MS_DIA_CE5-55_004	GW 1:1		Unknown	3	4:2 FTS	C6H5F9O3S	326.9743	326.97322	-3.272	5.909	0.028	94	757124-72-4	
4		GW 151_Cal 7_EPA1633_MS_DIA_CE5-55_003	GW 1:1		Unknown	4	5:3 FTCA	C8H5F11O2	341.0041	341.00342	-2.023	6.847	0.033	97	914637-49-3	
5		Blank_Null_EPA1633_MS_DIA_CE5-55_001	Blank		Unknown	5	6:2 FTS	C8H5F13O3S	426.9679	426.96701	-2.084	7.288	0.052	100	27619-97-2	
						6	7:3 FTCA	C10H5F15O2	440.9977	440.99665	-2.449	7.966	0.038	98	812-70-4	
						7	8:2 FTS	C10H5F17O3S	526.9615	526.96035	-2.220	8.235	0.033	99	39108-34-4	
						8	9CI-PF3ONS	C8HCIF16O4S	530.8956	530.89452	-1.997	8.011	0.035	94	756426-58-1	
						9	ADONA	C7H2F12O4	376.9689	376.96799	-2.334	6.779	0.032	100	919005-14-4	
						10	FOSA	C8H2F17NO2S	497.9462	497.94495	-2.510	8.638	0.015	100	754-91-6	
						11	HFPO-DA	C6HF11O3	284.9779	284.97721	-2.421	6.221	0.037	97	13252-13-6	
						12	NHFFOΔ	C10H4F17NO2S	525.9775	525.97610	-2.662	9.564	0.012	100	4151-50-2	

Compound Details - GW 151_Cal 7_EPA1633_MS_DIA_CE5-55_003 - 5:3 FTCA

Assign - 11:MSMS(-)[337.5000] CE:5.0-55.0 RT:[6.830-6.860]-[6.755-7.115] - 4,4,5,5,6,6,7,7,8,8-undecafluorooctanoic acid

#	Name	Formula	Mass
1	4,4,5,5,6,6,7,7,8,...	C8H5F11O2	342.01139
2	ethyl 2,2,3,3,4,4,...	C8H5F11O2	342.01139
3	1-(2,2-difluoro...	C8H5F11O2	342.01139
4	1,1,1,2,3,3-hexa...	C8H5F11O2	342.01139
5	1,1,1,2,3,3-hexa...	C8H5F11O2	342.01139
6	5-(1,1-difluoro...	C8H5F11O2	342.01139
7	2-[[1,1,2,3,4,4,...	C8H5F11O2	342.01139
8	4,5,5,5-tetraflu...	C8H5F11O2	342.01139
9	2-ethyl-2,3,5,5-	C8H5F11O2	342.01139

#	Formulae (M)	m/z	Pred. m/z	Intensity	+/- ppm	+/- mDa	Charge	Depth	Structures
<input checked="" type="checkbox"/>	Available								
<input checked="" type="checkbox"/>	C7HO2 C2HO...	116.99773	116.99820	270	-4.017	-0.47	----	----	----
<input checked="" type="checkbox"/>	C2F5	118.99153	118.99256	116	-8.656	-1.03	-	1	1
<input checked="" type="checkbox"/>	C4HO2F2	118.99153	118.99501	116	-29.245	-3.48	[-H]-	2	1
<input checked="" type="checkbox"/>	C4H2OF3	123.00748	123.00632	120	9.430	1.16	[-H]-	2	1

Compound details of the highlighted compound, including chromatogram, theoretical (top) and actual (bottom) MS and MS/MS spectrum and structure.

SHIMADZU

Why?

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Results – Suspects Screening

The Assign function can be used to search ChemSpider or PubChem based off the acquired mass or a formula.

LabSolutions Insight (Admin) - (EPA1633_MS_DIA_CE5-55.damlp - EPA1633_MS_DIA_CE5-55.lcm) [Pre-Release]

Sample List

#	Flags	Data Filename	Sample Name	Flag ID	Sample Type	Vial
1		GW 1510_Cal 7_EPA1633_MS_DIA_CE5-55_0...	GW 1:10	ppm	Unknown	7
2		EPA 1633_Cal 7_EPA1633_MS_DIA_CE5-55_...	EPA 1633		Unknown	9
3		GW 151_Cal 7_EPA1633_MS_DIA_CE5-55_004	GW 1:1		Unknown	8
4		GW 151_Cal 7_EPA1633_MS_DIA_CE5-55_003	GW 1:1		Unknown	8
5		Blank_Null_EPA1633_MS_DIA_CE5-55_001	Blank	mDa, ppm	Unknown	-1

Compound Results - GW 151_Cal 7_EPA1633_MS_DIA_CE5-55_003

#	Name	Formula	m/z	Found Mz	Mass Error (ppm)	Found RT	RT Diff	Lib. SI	CAS #	Mass E
1	11CI-PF3OUdS	C10HCIF20O4S	630.8892	630.88761	-2.504	8.726	0.041	100	763051-92-9	
2	3:3 FTCA	C6H5F7O2	241.0105	241.00953	-4.025	5.143	0.024	94	356-02-5	
3	4:2 FTS	C6H5F9O3S	326.9743	326.97322	-3.272	5.909	0.028	94	757124-72-4	
4	5:3 FTCA	C8H5F11O2	341.0041	341.00342	-2.023	6.847	0.033	97	914637-49-3	
5	6:2 FTS	C8H5F13O3S	426.9679	426.96701	-2.084	7.288	0.052	100	27619-97-2	
6	7:3 FTCA	C10H5F15O2	440.9977	440.99665	-2.449	7.966	0.038	98	812-70-4	
7	8:2 FTS	C10H5F17O3S	526.9615	526.96035	-2.220	8.235	0.033	99	39108-34-4	
8	9CI-PF3ONS	C8HCIF16O4S	530.8956	530.89452	-1.997	8.011	0.035	94	756426-58-1	
9	ADONA	C7H2F12O4	376.9689	376.96799	-2.334	6.779	0.032	100	919005-14-4	
10	FOSA	C8H2F17NO2S	497.9462	497.94495	-2.510	8.638	0.015	100	754-91-6	
11	HFPO-DA	C6HF11O3	284.9779	284.97721	-2.421	6.221	0.037	97	13252-13-6	
12	NHFOA	C10H4F17NO2S	525.9775	525.97610	-2.662	9.564	0.112	100	4151-50-2	

Compound Details - GW 151_Cal 7_EPA1633_MS_DIA_CE5-55_003 - 5:3 FTCA

Assign - 11:MSMS(-)[337.5000] CE:5.0-55.0 RT:[6.830-6.860]-[6.755-7.115] - 4,4,5,5,6,6,7,7,8,8-undecafluorooctanoic acid

#	Formulae (M)	m/z	Pred. m/z	Intensity	+/- ppm	+/- mDa	Charge	Depth	Structures
10	C7HO2 C2HO...	116.99773	116.99820	270	-4.017	-0.47	----	----	----
11	C2F5	118.99153	118.99256	116	-8.656	-1.03	-	1	1
12	C4HO2F2	118.99153	118.99501	116	-29.245	-3.48	[-H]-	2	1
13	C4H2OF3	123.00748	123.00632	120	9.430	1.16	[-H]-	2	1

Library Hits

#	Name	Formula	Mass
1	4,4,5,5,6,6,7,7,8,...	C8H5F11O2	342.01139
2	ethyl 2,2,3,3,4,4,...	C8H5F11O2	342.01139
3	1-(2,2-difluoro...	C8H5F11O2	342.01139
4	1,1,1,2,3,3-hexa...	C8H5F11O2	342.01139
5	1,1,1,2,3,3-hexa...	C8H5F11O2	342.01139
6	5-(1,1-difluoro...	C8H5F11O2	342.01139
7	2-[[1,1,2,3,4,4,...	C8H5F11O2	342.01139
8	4,5,5,5-tetraflu...	C8H5F11O2	342.01139
9	2-ethyl-2,2,3,3,5...	C8H5F11O2	342.01139

Results – Suspects Screening

LabSolutions Insight (Admin) - (EPA1633_MS_DIA_CE5-55.damlp - EPA1633_MS_DIA_CE5-55.lcm) [Pre-Release]

Shortcuts

- File
- View
- Edit
- Edit Method
- Edit Tables
- Edit Flags
- Select Compounds
- Edit Library
- Review
- Report
- Explore

Sample List					
#	Flags	Data Filename	Sample Name	Flag ID	Sample Type
1		GW 1510_Cal 7_EPA1633_MS_DIA_CE5-55_0...	GW 1:10	ppm	Unknown
2		EPA 1633_Cal 7_EPA1633_MS_DIA_CE5-55_...	EPA 1633		Unknown
3		GW 151_Cal 7_EPA1633_MS_DIA_CE5-55_004	GW 1:1		Unknown
4		GW 151_Cal 7_EPA1633_MS_DIA_CE5-55_003	GW 1:1		Unknown
5		Blank_Null_EPA1633_MS_DIA_CE5-55_001	Blank	mDa, ppm	Unknown

Compound Results - GW 151_Cal 7_EPA1633_MS_DIA_CE5-55_003										
#	Name	Formula	m/z	Found Mz	Mass Error (ppm)	Found RT	RT Diff	Lib. SI	CAS #	Mass E
1	11CI-PF3OUdS	C10HCIF20O4S	630.8892	630.88761	-2.504	8.726	0.041	100	763051-92-9	
2	3:3 FTCA	C6H5F7O2	241.0105	241.00953	-4.025	5.143	0.024	94	356-02-5	
3	4:2 FTS	C6H5F9O3S	326.9743	326.97322	-3.272	5.909	0.028	94	757124-72-4	
4	5:3 FTCA	C8H5F11O2	341.0041	341.00342	-2.023	6.847	0.033	97	914637-49-3	
5	6:2 FTS	C8H5F13O3S	426.9679	426.96701	-2.084	7.288	0.052	100	27619-97-2	
6	7:3 FTCA	C10H5F15O2	440.9977	440.99665	-2.449	7.966	0.038	98	812-70-4	
7	8:2 FTS	C10H5F17O3S	526.9615	526.96035	-2.220	8.235	0.033	99	39108-34-4	
8	9CI-PF3ONS	C8HCIF16O4S	530.8956	530.89452	-1.997	8.011	0.035	94	756426-58-1	
9	ADONA	C7H2F12O4	376.9689	376.96799	-2.334	6.779	0.032	100	919005-14-4	
10	FOSA	C8H2F17NO2S	497.9462	497.94495	-2.510	8.638	0.015	100	754-91-6	
11	HFPO-DA	C6HF11O3	284.9779	284.97721	-2.421	6.221	0.037	97	13252-13-6	
12	NHFFOΔ	C10H4F17NO2S	525.9775	525.97610	-2.662	9.564	0.112	100	4151-50-2	

Compound Details Analyze Library Hits Mass Calculator

Assign - 11:MSMS(-)[337.5000] CE:5.0-55.0 RT:[6.830-6.860]-[6.755-7.115] - 4,4,5,5,6,6,7,7,8,8-undecafluorooctanoic acid

4,4,5,5,6,6,7,7,8,8-undecafluorooctanoic acid

#	Formulae (M)	m/z	Pred. m/z	Intensity	+/- ppm	+/- mDa	Charge	Depth	Structures
10	C7HO2 C2HO...	116.99773	116.99820	270	-4.017	-0.47	----	----	----
11	C2F5	118.99153	118.99256	116	-8.656	-1.03	-	1	1
12	C4HO2F2	118.99153	118.99501	116	-29.245	-3.48	[-H]-	2	1
13	C4H2OF3	123.00748	123.00632	120	9.430	1.16	[-H]-	2	1

#	Name	Formula	Mass
1	4,4,5,5,6,6,7,7,8,...	C8H5F11O2	342.01139
2	ethyl 2,2,3,3,4,4,...	C8H5F11O2	342.01139
3	1-(2,2-difluoro...	C8H5F11O2	342.01139
4	1,1,1,2,3,3-hexa...	C8H5F11O2	342.01139
5	1,1,1,2,3,3-hexa...	C8H5F11O2	342.01139
6	5-(1,1-difluoro...	C8H5F11O2	342.01139
7	2-[[1,1,2,2,3,4,4,...	C8H5F11O2	342.01139
8	4,5,5,5-tetraflu...	C8H5F11O2	342.01139
9	2-ethyl-2,2,3,5,5...	C8H5F11O2	342.01139

The compound table shows the name, formula, theoretical and acquired precursor m/z , mass error, many more compound specific parameters.

Results – Suspects Screening

LabSolutions Insight (Admin) - (EPA1633_MS_DIA_CE5-55.damlp - EPA1633_MS_DIA_CE5-55.lcm) [Pre-Release]

Shortcuts

- File
- View
- Edit
- Edit Method
- Integrate Batch
- Integrate Sample
- Integrate Compound
- Integrate Result
- Edit Tables
- Edit Flags
- Select Compounds
- Edit Library
- Review
- Report
- Explore

Sample List						Compound Results - GW 151_Cal 7_EPA1633_MS_DIA_CE5-55_003										
#	Flags	Data Filename	Sample Name	Flag ID	Sample Type	#	Name	Formula	m/z	Found Mz	Mass Error (ppm)	Found RT	RT Diff	Lib. SI	CAS #	Mass E
1		GW 1510_Cal 7_EPA1633_MS_DIA_CE5-55_003	GW 1:10	ppm	Unknown	1	11CI-PF3OUdS	C10HCIF20O4S	630.8892	630.88761	-2.504	8.726	0.041	100	763051-92-9	
2		EPA 1633_Cal 7_EPA1633_MS_DIA_CE5-55_003	EPA 1633		Unknown	2	3:3 FTCA	C6H5F7O2	241.0105	241.00953	-4.025	5.143	0.024	94	356-02-5	
3		GW 151_Cal 7_EPA1633_MS_DIA_CE5-55_004	GW 1:1		Unknown	3	4:2 FTS	C6H5F9O3S	326.9743	326.97322	-3.272	5.909	0.028	94	757124-72-4	
4		GW 151_Cal 7_EPA1633_MS_DIA_CE5-55_003	GW 1:1		Unknown	4	5:3 FTCA	C8H5F11O2	341.0041	341.00342	-2.023	6.847	0.033	97	914637-49-3	
5		Blank_Null_EPA1633_MS_DIA_CE5-55_001	Blank	mDa, ppm	Unknown	5	6:2 FTS	C8H5F13O3S	426.9679	426.96701	-2.084	7.288	0.052	100	27619-97-2	
						6	7:3 FTCA	C10H5F15O2	440.9977	440.99665	-2.449	7.966	0.038	98	812-70-4	
						7	8:2 FTS	C10H5F17O3S	526.9615	526.96035	-2.220	8.235	0.033	99	39108-34-4	
						8	9CI-PF3ONS	C8HCIF16O4S	530.8956	530.89452	-1.997	8.011	0.035	94	756426-58-1	
						9	ADONA	C7H2F12O4	376.9689	376.96799	-2.334	6.779	0.032	100	919005-14-4	
						10	FOSA	C8H2F17NO2S	497.9462	497.94495	-2.510	8.638	0.015	100	754-91-6	
						11	HFPO-DA	C6HF11O3	284.9779	284.97721	-2.421	6.221	0.037	97	13252-13-6	
						12	NHFOA	C10H4F17NO2S	525.9775	525.97610	-2.662	9.564	0.112	100	4151-50-2	

Compound Details - GW 151_Cal 7_EPA1633_MS_DIA_CE5-55_003 - 5:3 FTCA

Compound Details - Analyze Library Hits Mass Calculator

Assign - 11:MSMS(-)[337.5000] CE:5.0-55.0 RT:[6.830-6.860]-[6.755-7.115] - 4,4,5,5,6,6,7,7,8,8-undecafluorooctanoic acid

#	Formulae (M)	m/z	Pred. m/z	Intensity	+/- ppm	+/- mDa	Charge	Depth	Structures
<input checked="" type="checkbox"/>	Available								
<input checked="" type="checkbox"/>	10 C7HO2 C2HO...	116.99773	116.99820	270	-4.017	-0.47	----	----	----
<input checked="" type="checkbox"/>	11 C2F5	118.99153	118.99256	116	-8.656	-1.03	-	1	1
<input checked="" type="checkbox"/>	12 C4HO2F2	118.99153	118.99501	116	-29.245	-3.48	[-H]-	2	1
<input checked="" type="checkbox"/>	13 C4H2OF3	123.00748	123.00632	120	9.430	1.16	[-H]-	2	1

#1 - C2 F5

Acquired MS/MS spectra are compared, and fragments are assigned with relevant structures and formula.

SHIMADZU

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Results – Suspects Screening

LabSolutions Insight (Admin) - (EPA1633_MS_DIA_CES-55.damlp - EPA1633_MS_DIA_CES-55.lcm) [Pre-Release]

Shortcuts

File

View

Compound

All

Compound Details

Calibration Curve

Library Hits

Survey

QC Chart

Settings

Edit

Review

Report

Explore

#	Flags	Data Filename	Sample Name	Flag ID	Sample Type	Cal Point	Level	Acquired Date
1		GW 1510_Cal 7_...	GW 1:10	eRT, LSI, NAME,...	Unknown		----	5/11/2023 3:27:...
2		EPA 1633_Cal 7_...	EPA 1633		Standard		1	5/11/2023 4:23:...
3		GW 151_Cal 7_E...	GW 1:1		Unknown		----	5/11/2023 4:04:...
4		GW 151_Cal 7_E...	GW 1:1		Unknown		----	5/11/2023 3:46:...
5		Blank_Null_EPA...	Blank	eRT, LSI, mDa, ...	Unknown		----	5/11/2023 3:09:...

#	Flags	Flag ID	Flag Result	Name	Found RT	Conc.	Unit	m/z
1			Pass	11CI-PF3OUdS	8.718	0.03097	ppm	630.8892
2			Pass	3:3 FTCA	5.184	0.07907	ppm	241.0105
3			Pass	4:2 FTS	5.912	0.08483	ppm	326.9743
4			Pass	5:3 FTCA	6.838	0.1142	ppm	341.0041
5			Pass	6:2 FTS	7.272	0.07830	ppm	426.9679
6			Pass	7:3 FTCA	7.966	0.1141	ppm	440.9977
7			Pass	8:2 FTS	8.234	0.07648	ppm	526.9615
8			Pass	9CI-PF3ONS	8.009	0.07674	ppm	530.8956
9			Pass	ADONA	6.779	0.08539	ppm	376.9689
10			Pass	FOSA	8.637	0.07086	ppm	497.9462
11			Pass	HFPO-DA	6.207	0.07096	ppm	284.9779
12			Pass	NETFOSA	9.553	0.05224	ppm	525.9775
13		LSI	Pass	NETFOSAA	8.599	0.04970	ppm	583.9830
14			Pass	NETFOSE	9.527	0.01581	ppm	630.0249
15		ppm	Pass	NFDHA	5.850	0.07915	ppm	294.9658
16			Pass	NMeFOSA	9.349	0.06077	ppm	511.9619
17			Pass	NMeFOSAA	8.420	0.05947	ppm	569.9673
18			Pass	NMeFOSE	9.317	0.02051	ppm	616.0092
19			Pass	PFBA	4.037	0.09259	ppm	212.9792
20			Pass	PFBS	5.238	0.08993	ppm	298.9430
21			Pass	PFDA	8.225	0.06996	ppm	512.9600

Survey

Zoom Show selected compound only

QC Chart

Show selected compound only Found RT

Results – Suspects Screening

LabSolutions Insight (Admin) - (EPA1633_MS_DIA_CES-55.damlp - EPA1633_MS_DIA_CES-55.lcm) [Pre-Release]

Sample List										Compound Results - GW 1510_Cal 7_EPA1633_MS_DIA_CES-55_002									
#	Flags	Data Filename	Sample Name	Flag ID	Sample Type	Cal Point	Level	Acquired Date		#	Flags	Flag ID	Flag Result	Name	Found RT	Conc.	Unit	m/z	
1		GW 1510_Cal 7_...	GW 1:10	eRT, LSI, NAME,...	Unknown			5/11/2023 3:27:...		1			Pass	11CI-PF3OUdS	8.718	0.03097	ppm	630.8892	6
2		EPA 1633_Cal 7_...	EPA 1633		Standard			5/11/2023 4:23:...		2			Pass	3:3 FTCA	5.184	0.07907	ppm	241.0105	2
3		GW 151_Cal 7_E...	GW 1:1		Unknown			5/11/2023 4:04:...		3			Pass	4:2 FTCS	5.912	0.08483	ppm	326.9743	2
4		GW 151_Cal 7_E...	GW 1:1		Unknown			5/11/2023 3:46:...		4			Pass	5:3 FTCA	6.838	0.1142	ppm	341.0041	23
5		Blank_Null_EPA...	Blank	eRT, LSI, mDa, ...	Unknown			5/11/2023 3:09:...		5			Pass	6:2 FTCS	7.272	0.07830	ppm	426.9679	3

NETFOSAA
 The library similarity index of 66 is below the minimum threshold of 75.

NFDHA
 The mass error of -5.018 ppm is outside the maximum threshold of 5.000 ppm.

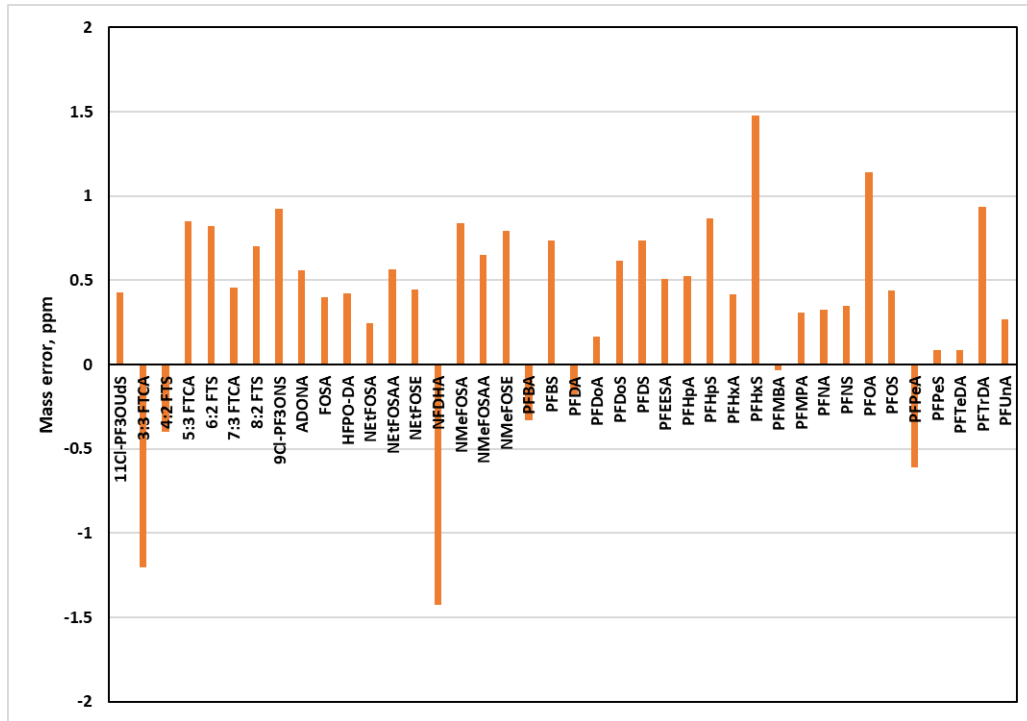
PFTeDA
 The library similarity index of 62 is below the minimum threshold of 75.

PFTrDA
 The library compound name does not match the result compound name.
 The library error RT of the identified peak is above the library error RT limit.
 The library similarity index of the identified peak spectrum is below the minimum threshold.

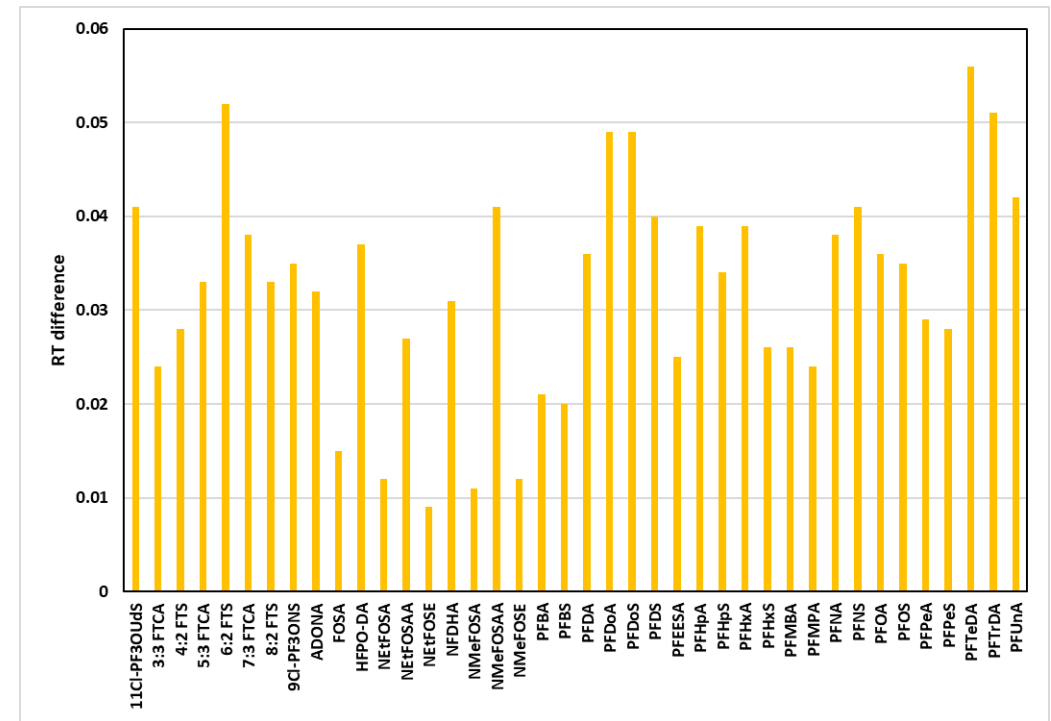
Chromatograms showing peaks at RT 8.726. The peak at 8.726 is highlighted in red in the bottom right plot.

Results – Suspects Screening *After reviewing Library hits*

All PFAS listed in EPA 1633 were identified by comparing results against the in-house developed library, and when reprocessing samples in an untargeted analysis workflow



Mass accuracy ± 2 ppm



RT diff < 0.06

Library Similarity Index: >75% (88-100%), and alignment of fragmentation pattern

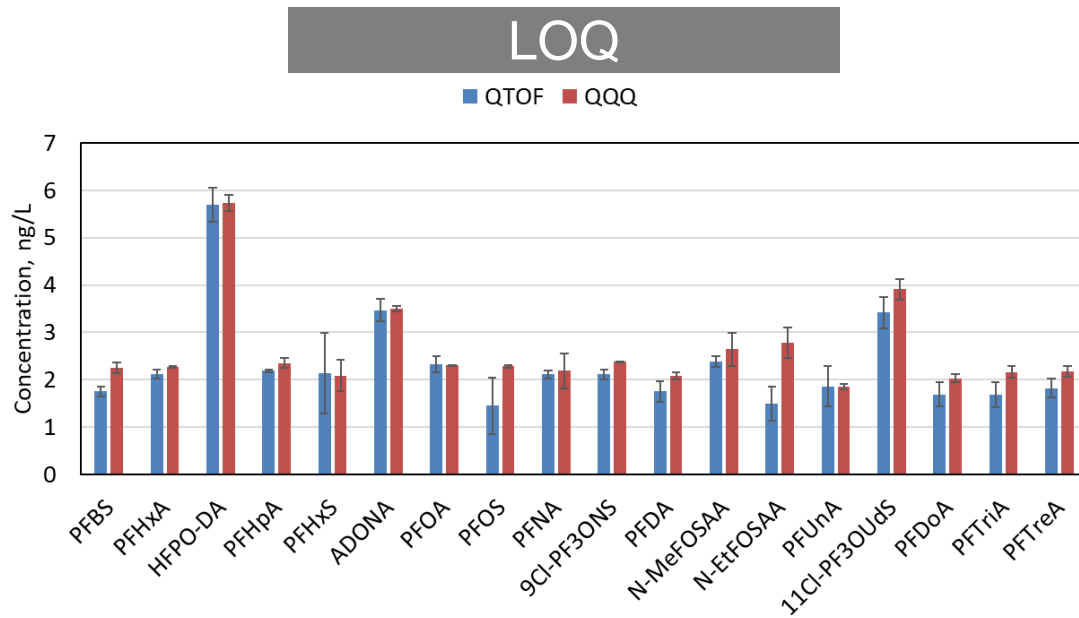
Results - Quantitation

The screenshot displays the LabSolutions Insight interface with the following components:

- Sample List:** A table listing samples with columns for #, Flags, Data Filename, Sample Name, Flag ID, Sample Type, Cal Point, Level, and Acquired Date.
- Compound Results - GW 1510_Cal 7_EPA1633_MS_DIA_CES-55_002:** A table listing compounds with columns for #, Flags, Flag ID, Flag Result, Name, Found RT, Conc., Unit, and m/z. A red arrow points to the top of this table.
- Survey:** A section containing five chromatograms for different samples, showing peaks for 11CI-PF30Uds. The peak at RT=8.718 is highlighted in blue, and the peak at RT=8.726 is highlighted in red.
- QC Chart:** A chart showing Found RT for various compounds across five samples. A red dot is visible at sample 2, RT=8.718.
- Navigation Panel:** A vertical sidebar on the left with icons for Compound, All, Compound Details, Calibration Curve (highlighted with a red arrow), Library Hits, Survey, QC Chart, and Settings.

Results - Quantitation

Comparable LOQs and Precision and Accuracy between QTOF and Triple Quadrupole



LCMS-9030

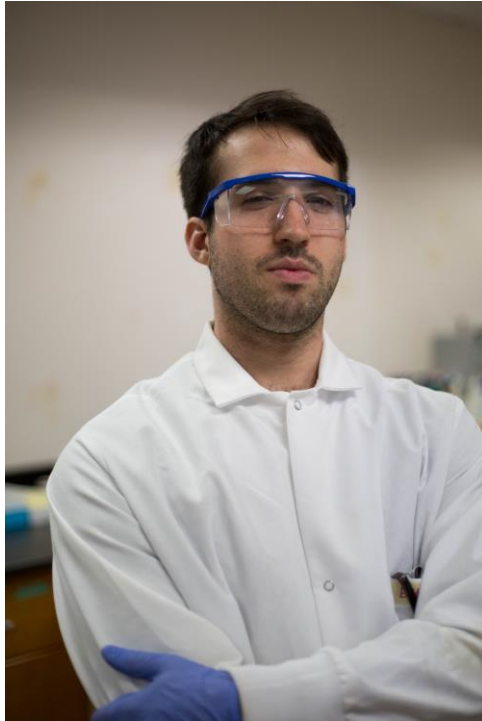
LCMS-8045

25 ppt P&A	Avg	Std Dev	%REC	%RSD	Avg	Std Dev	%REC	%RSD
PFBS	23.7	1.80416	107	7.6	25.6	0.62923	115	2.5
PFHxA	29	1.38519	116	4.8	25.6	1.009	103	3.9
HFPO-DA	80.4	5.89465	129	7.3	67.2	2.45917	108	3.7
PFHpA	27.6	2.16336	110	7.8	26.4	0.95392	106	3.6
PFHxS	25.9	3.72591	113	14.4	26.4	1.09444	80	4.1
ADONA	46.2	2.80041	185	6.1	39.7	1.28787	159	3.2
PFOA	28.7	1.65379	115	5.8	25.1	0.61854	100	2.5
PFOS	21.7	4.99594	94	23.1	25.8	0.7969	112	3.1
PFNA	28.2	1.78411	113	6.3	25.7	0.93568	103	3.6
9Cl-PF3ONS	28	1.68409	121	6	27.7	0.80942	119	2.9
PFDA	25	1.56208	100	6.3	23.4	0.68554	94	2.9
N-MeFOSAA	29.2	3.31232	117	11.3	31.4	1.74514	126	5.6
N-EtFOSAA	25.9	6.11145	104	23.6	34.8	1.90454	139	5.5
PFUnA	24.4	1.88443	98	7.7	24.1	0.87477	96	3.6
11Cl-PF3OUdS	44.5	2.93683	189	6.6	46	1.8548	196	4
PFDoA	22.1	1.43221	88	6.5	23.5	0.66533	94	2.8
PFTriA	22.8	1.44296	91	6.3	24.4	0.91626	98	3.8
PFTreA	22.8	1.54577	91	6.8	25.2	0.72595	101	2.9

Take-Home Messages

- Highly reliable workflow for the targeted and suspect screening of 40 common PFAS was demonstrated in clean and environmental matrices, including:
 - High mass accuracy and
 - Low retention time differences
- The presented workflow for suspect and target screening workflows, including quantitation, is easily manageable and transferrable between platforms (LC-MS/MS and LC-QTOF) in non-R&D laboratories.

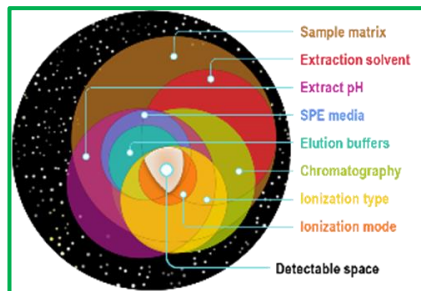
Acknowledgments



Ethan Hain, PhD
Product Specialist, LCMS



Christopher Gilles
General Manager – Product Managers




REFERENCE CONTENT
 Study design
 Data acquisition
 Data processing and analysis
 Data outputs
 QA/QC metrics
 Glossary

ADDITIONAL RESOURCES
 Literature library
 NTA Software Tools
 Online Databases
 and Libraries
 Events and organizations
 Mass Spec vendors

Section	Category	Sub-Category	Example Information to Report	Score	Rationale	
NTA Study Chronology	Study Design	Objectives & Scope				
		Sample Info & Preparation				
		QC Spikes & Samples				
	Methods	Data Acquisition	Analytical Sequence	<ul style="list-style-type: none"> 3-4 examples of representative information to report for each of the 13 sub-categories. 	NA	Space for reviewer to explain assigned score (i.e., typical peer review rationale)
			Chromatography			
		Mass Spectrometry	0			
		Data Processing	1			
	Data Processing & Analysis	Statistical & Chemometric Analysis	Annotation & Identification	<ul style="list-style-type: none"> Not an exhaustive list – intended to guide researcher/reviewer and relies on expertise/discretion. 	2	
			Statistical & Chemometric Outputs	3		
		Data Outputs	ID & Confidence Levels		1	
	Results	QA/QC Metrics	Data Acquisition QA/QC		1	
			Data Processing & Analysis QA/QC		3	

Study Planning Tool – in preparation

Forum	Topics	Posts	Last Post
<p>NTA Forum</p> <p>This is the BP4NTA forum to discuss all topics related to non-targeted analysis including instruments, software, workflows, and more!</p> <p>Instruments (3, 5), Software (3, 6), Workflows (1, 7), General (2, 3)</p>	9	30	1 week ago  gblack



Access the website
of the Best Practices and
Publications for Non-Targeted
Analysis (BP4NTA)
workgroup



Become a member
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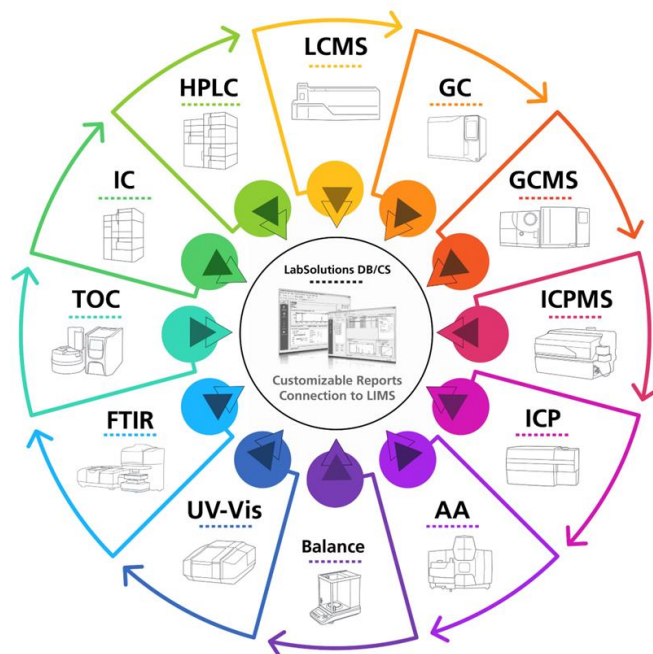
And get access to the **monthly meetings and webinar series**
(3rd Tuesday of the month, 12 pm EST)

For more information, reach out to the current BP4NTA Chairs at:



Ruth Marfil-Vega: rmmarfilvega@shimadzu.com
Christine Fisher (O'Donnell): christine.odonnell@fda.hhs.gov





For any questions, contact:
Ruth Marfil-Vega, PhD
rmmarfilvega@shimadzu.com

For more information, visit:
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