# Analysis of Persistent Organic Pollutants (POPs) by high-resolution GC/MS

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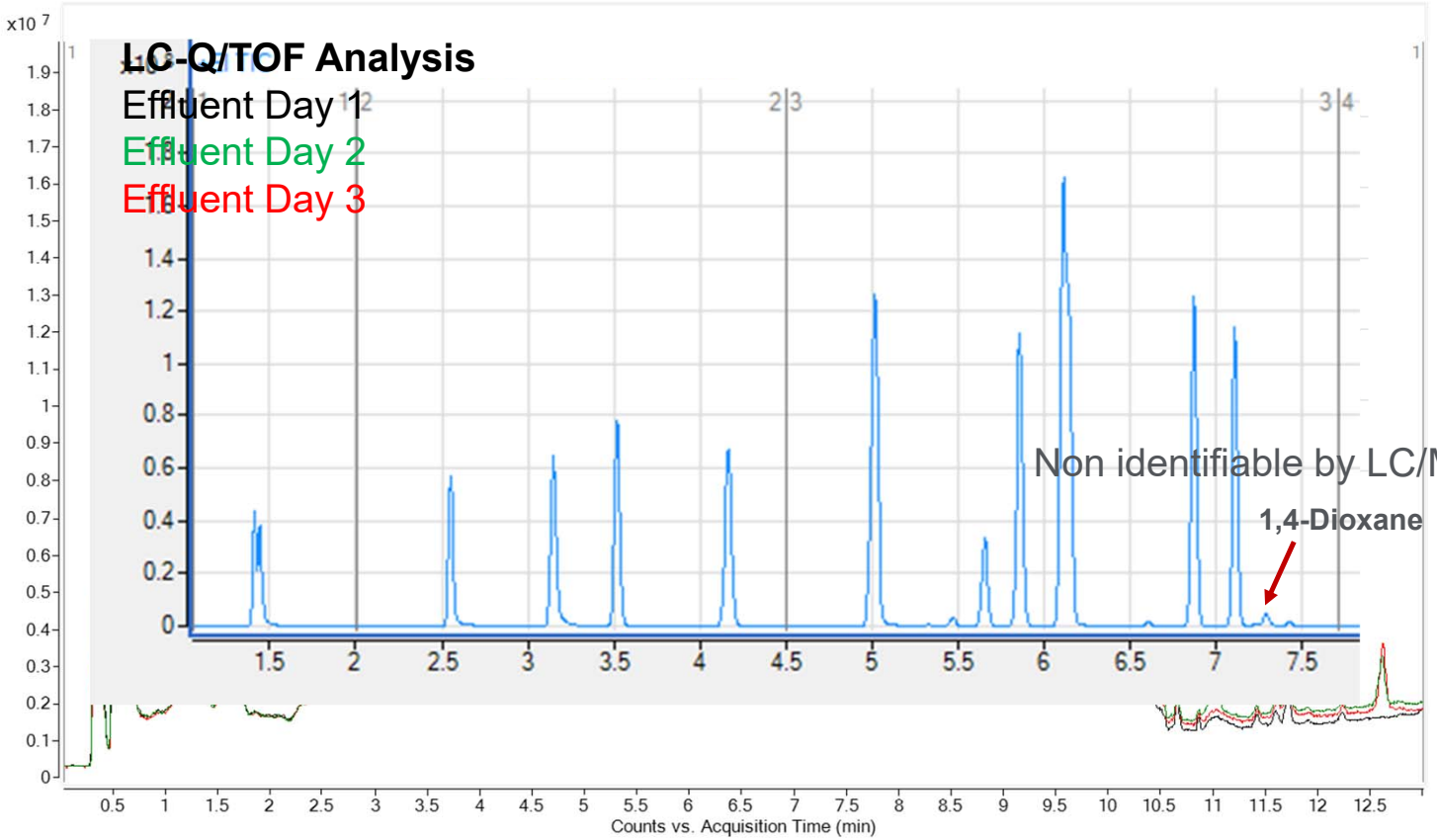
Agilent Technologies Inc.

# Why do you need a GC/Q-TOF?
# Water Treatment plant samples in NY



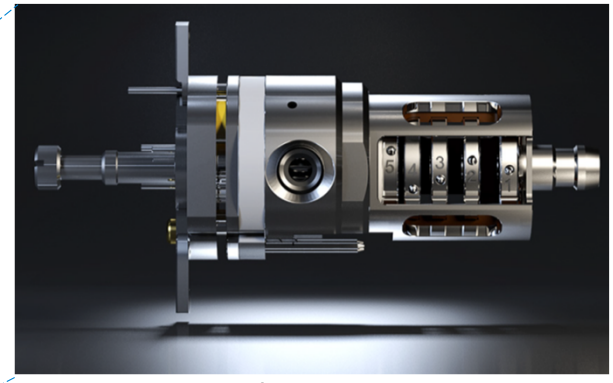

Good reproducibility

Statistically no relevant features to account for spike in toxicity on day 2

Samples re-run on GC/MS with suitable extraction

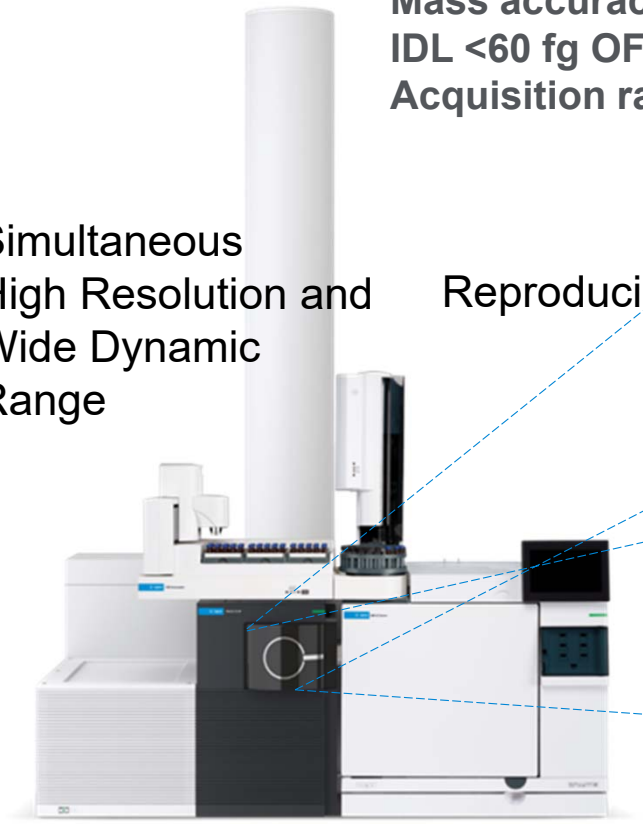
# 7250 GC/Q-TOF

**Resolving power: >25,000 at m/z 272**
**Mass accuracy: <2 ppm**
**IDL <60 fg OFN (EI)**
**Acquisition rate: 1-50 Hz**



Standard/Low Energy EI Source

Simultaneous High Resolution and Wide Dynamic Range



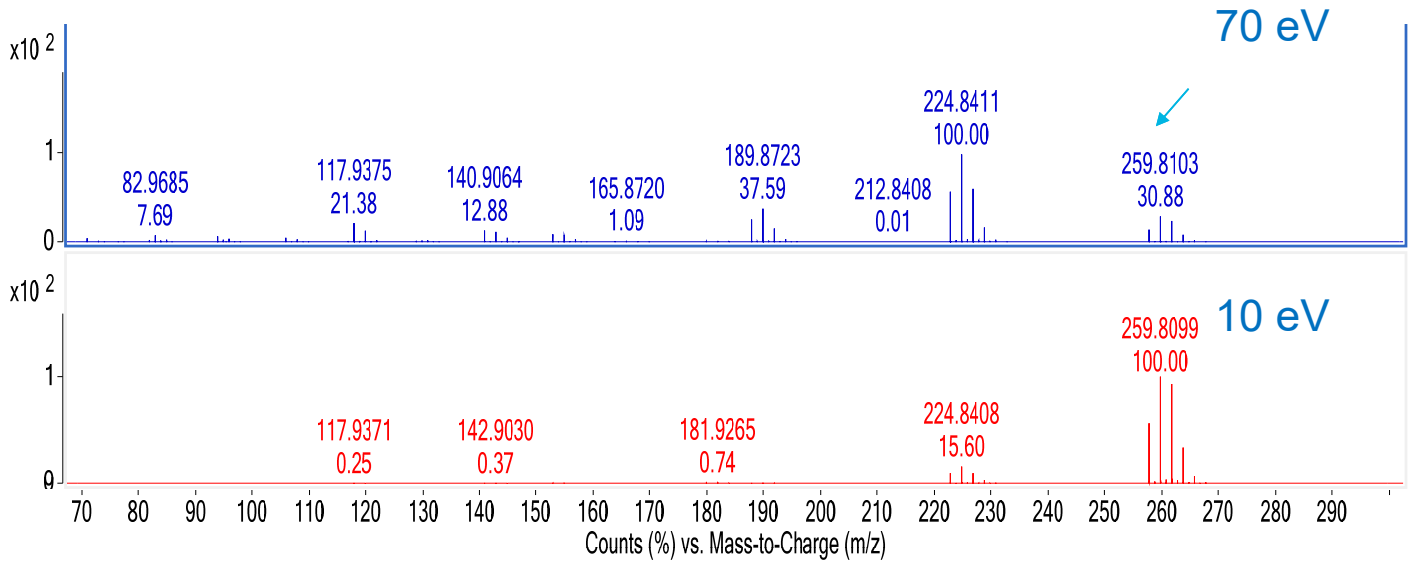
Reproducible Spectral Performance

High Resolution and Mass Accuracy

CI Source

# Traditional EI vs Low Energy EI

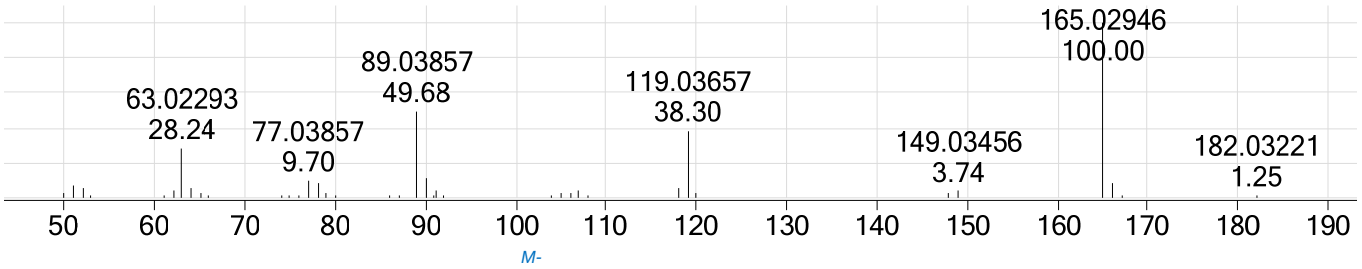
***hexachlorobutadiene***



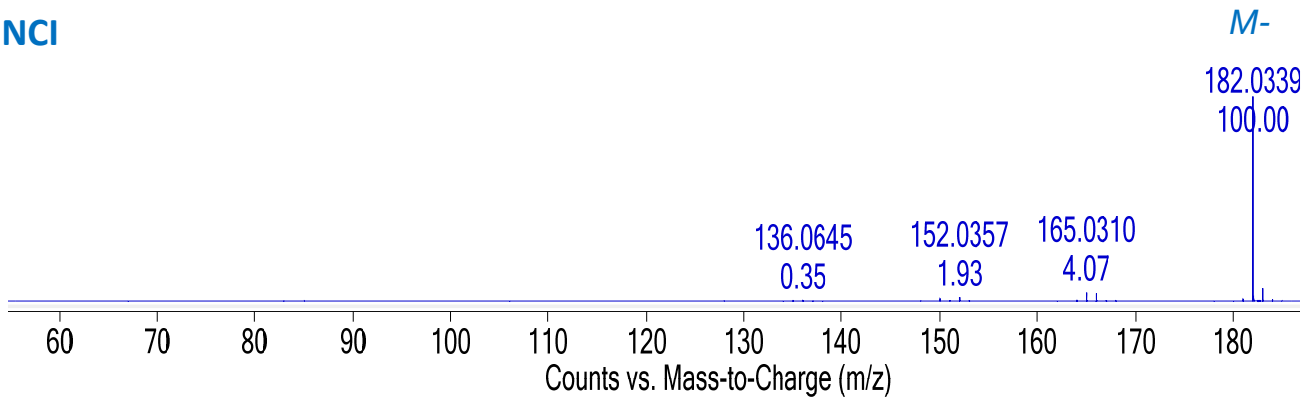
# Chemical Ionization vs Low Energy EI

***2,4-Dinitrotoluene***

**EI**




**NCI**



# Workflow Strategy

**Acquire full-spectrum data**

Targeted Method

Yes

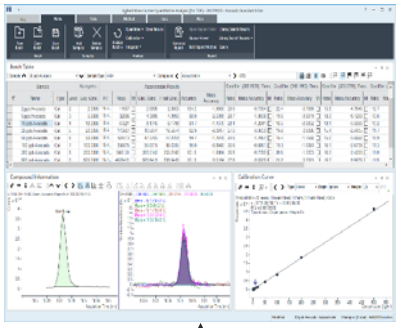
Calibrate?

No

**Targeted Quantitation**

**Suspect Screening**

**Non-targeted Screening**




NIST

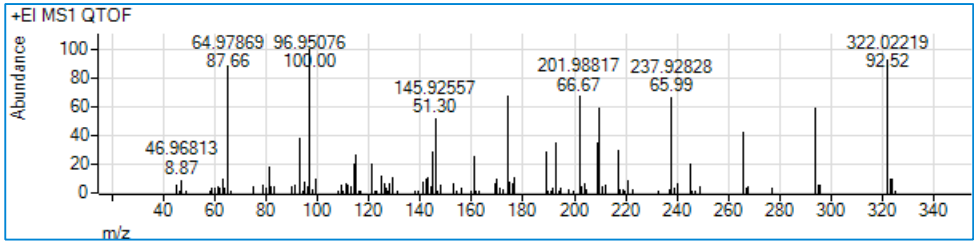
Deconvolution followed by matching public libraries

Suspect added to target

# GC/Q-TOF Accurate Mass Library

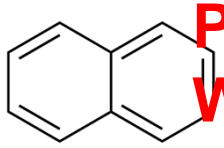
GC/Q-TOF Pesticides & Environmental Pollutants PCDL – now with ***1000+ compounds***:

- High Resolution Spectra
- Expert curation
- Better compound alignment with EPA SVOC targets and Agilent GC/TQ MRM database

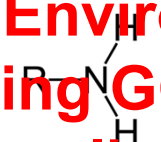



**Talk on Screening & Identification by GC-QTOF**

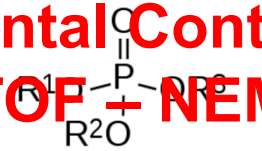
**Profiling Environmental Contaminants in Water Using GC-Q/TOF – NEMC 2019, available online**



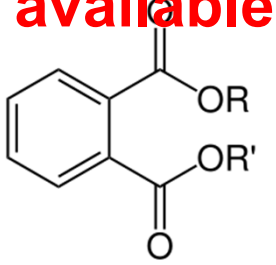
PAHs



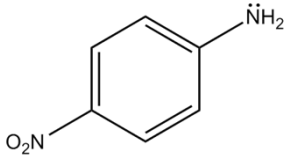
Amines



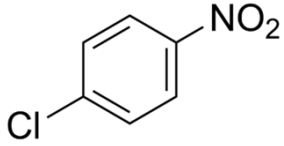
Organophosphates



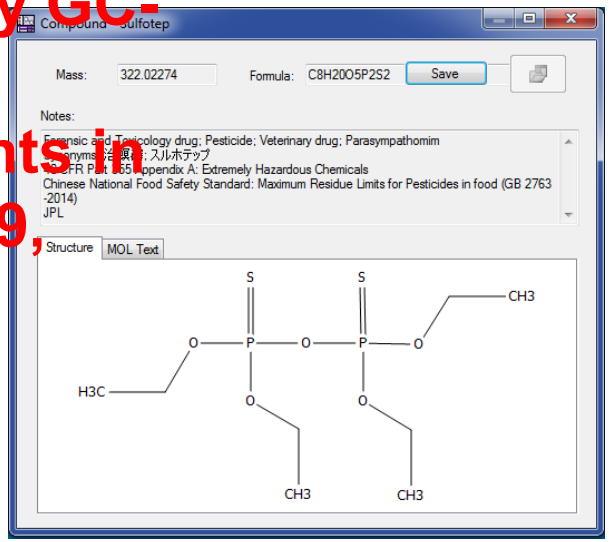
Phthalates



Nitroanilines



Chloronitrobenzenes



# Persistent Organic Pollutants (POPs) in Stockholm Convention

- The Stockholm Convention on POPs (2001) is a global treaty to protect **human health** from chemicals that remain in the environment and are **persistent**, **bioaccumulative** and transportable across the globe.

- Include:
  - Industrial chemicals ex. PCBs, hexachlorobenzene
  - Pesticides ex. Aldrin, DDT, endrin, toxaphene etc.
  - Pharmaceuticals
  - Solvents
  - By-products ex. **Dioxins & Furans**

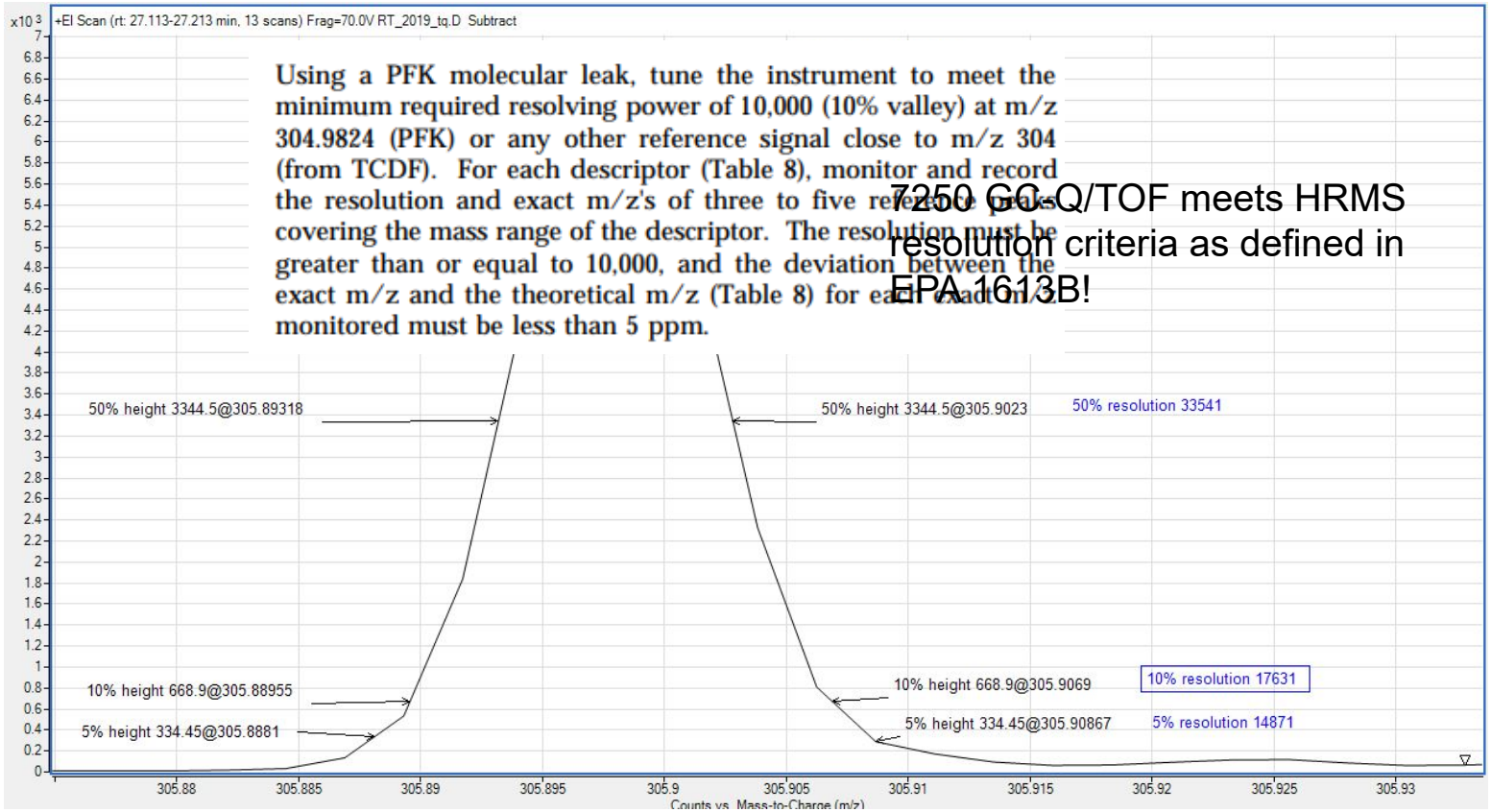

- Initially ‘dirty dozen’ but new POPs include Perfluoro alkyl substances (**PFAS**), short chain chlorinated parafins (**SCCPs**), pesticides, Deca-BDE etc.

- Typically hydrophobic, therefore GC/MS analysis is a superior choice

# Analysis of Dioxins by GC/Q-TOF

# Analysis of Dioxins in US done mainly by EPA 1613B

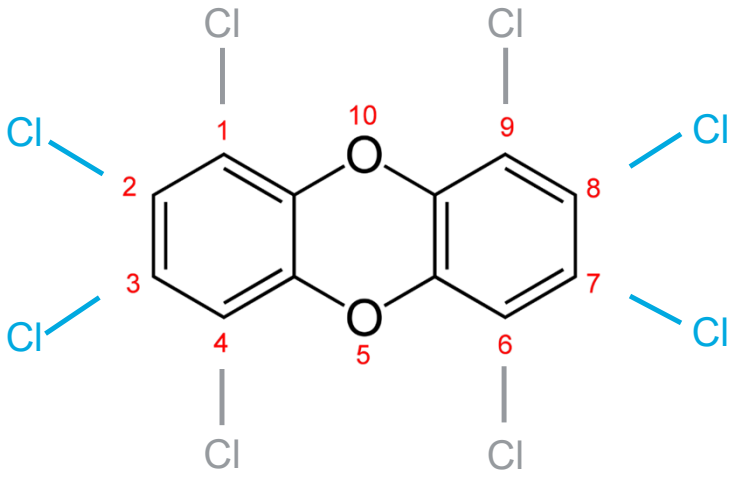
## Tetra- through Octo-Chlorinated Dioxins & Furans by HRGC/HRMS




7250 GC-Q/TOF meets HRMS resolution criteria as defined in EPA 1613B!

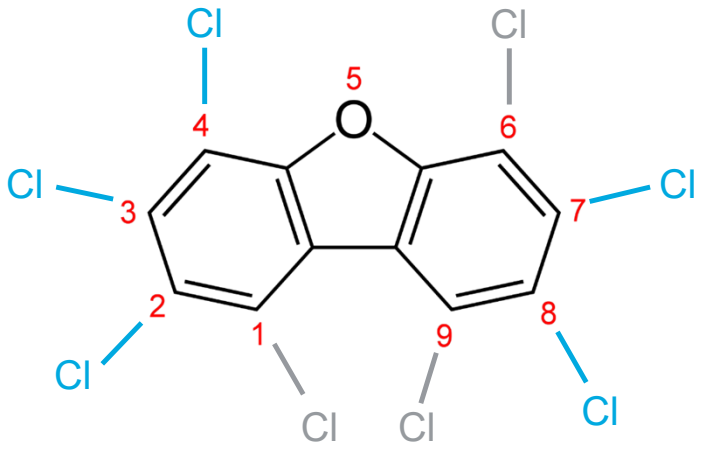
# PCDD and PCDF Nomenclature and Isomers

**Dioxins**
Polychlorinated dibenzodioxin (PCDD)




2,3,7,8 – TCDD
(2,3,7,8-Tetrachlorinated dibenzodioxin)

**Furans**
Polychlorinated dibenzofurans (PCDF)



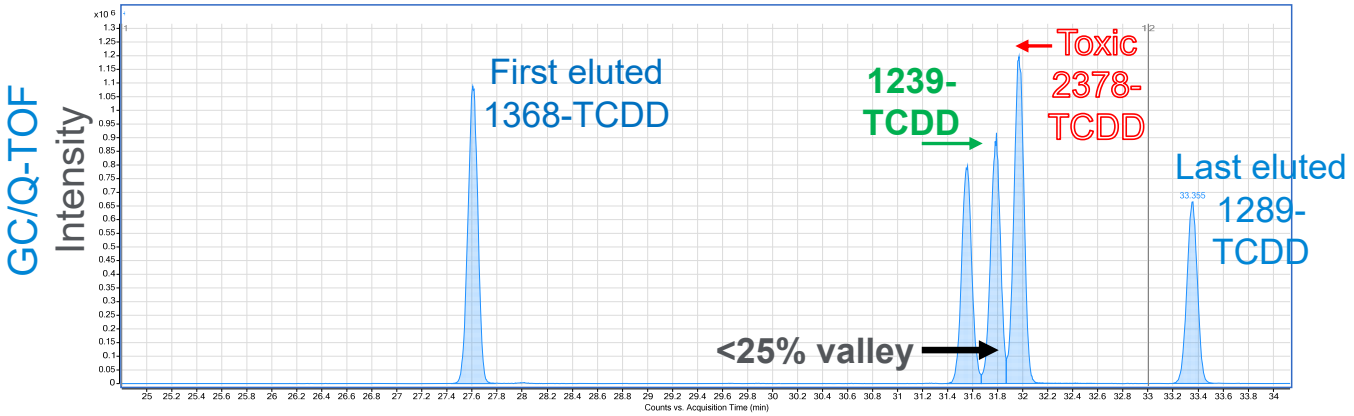

2,3,4,7,8 – PCDF
(2,3,4,7,8-Pentachlorinated dibenzofurans)

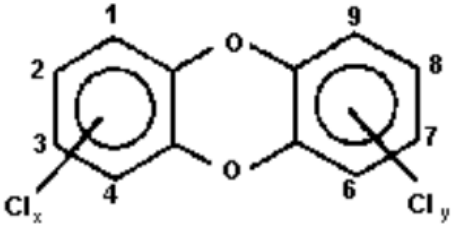
| Chlorine Atoms | PCDD Isomers | PCDF Isomers |
|---|---|---|
| **1** | 2 | 4 |
| **2** | 10 | 16 |
| **3** | 14 | 28 |
| **4** | 22 | 38 |
| **5** | 14 | 28 |
| **6** | 10 | 16 |
| **7** | 2 | 4 |
| **8** | 1 | 1 |
| **Total** | **75** | **135** |

75 total PCDD isomers | 135 total PCDF isomers | **17 toxic isomer**

# Tetrachlorinated dibenzodioxins (TCDD)

## Separation of Isomers

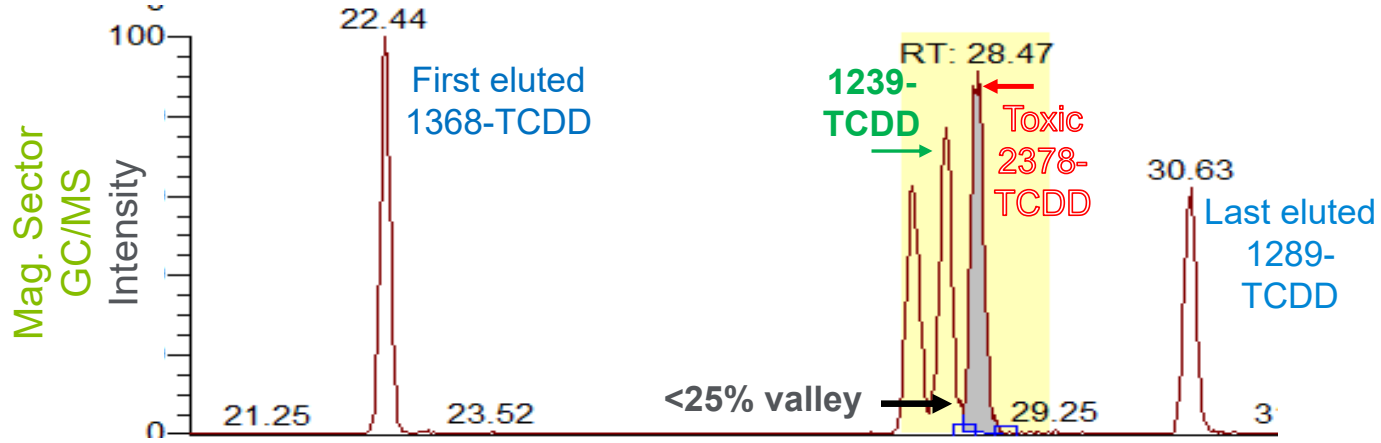



$x + y = 4$

*22 isomers*

***1 toxic***



# Method Detection Limit (MDL) for 2,3,7,8-TCDD

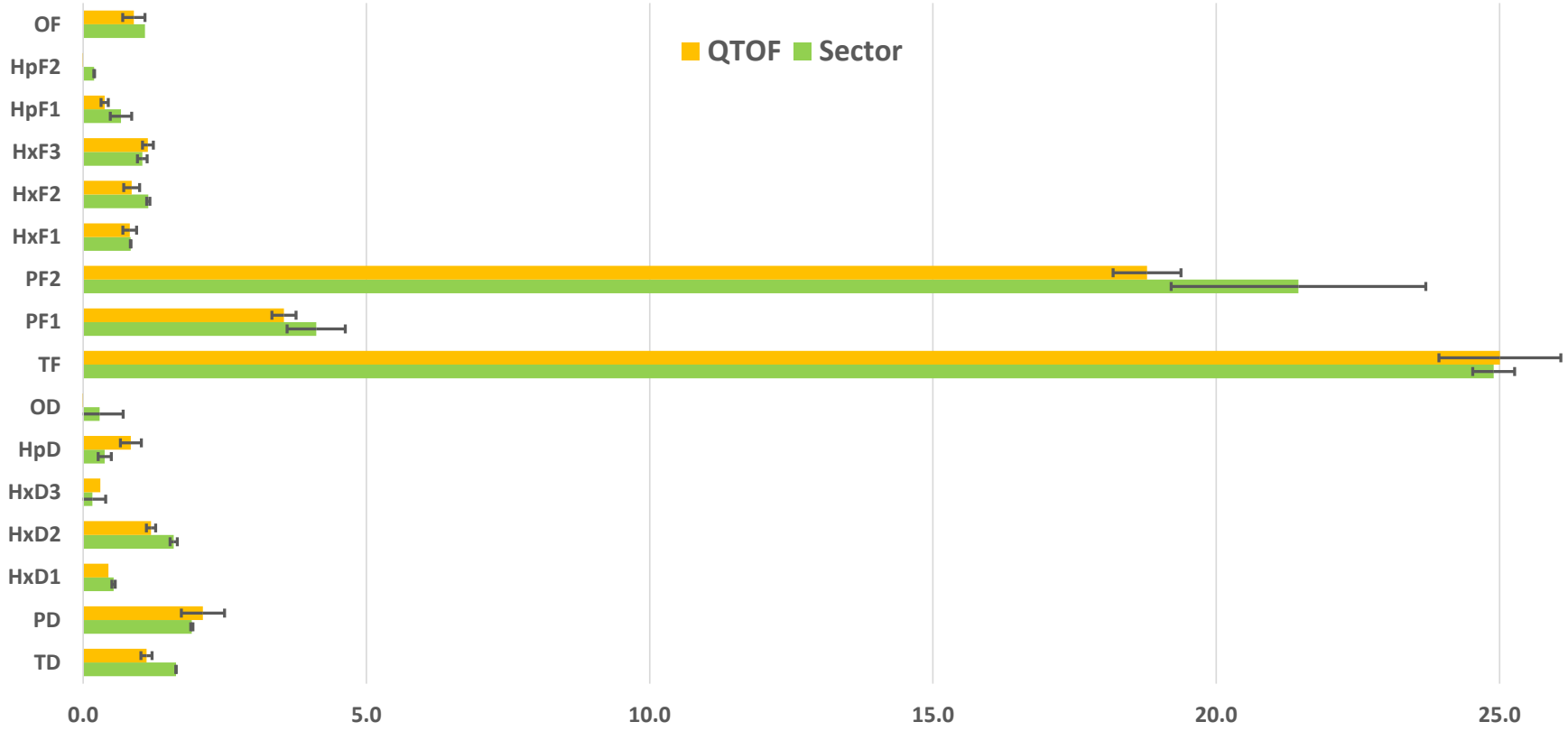
## Samples concentration of 10,000X (500 mL to 50 µL)

MDL for 2,3,7,8-TCDD is 4.4 pg/L in EPA 1613B

| Sample Concentration | Injected amount | 15 eV, 1 µA | | | 70 eV, 1 µA | | | 70 eV, 10 µA | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Spiked equivalent | | RSD % | MDL (pg/L) | IDL (fg) | RSD % | MDL (pg/L) | IDL | RSD % | MDL (pg/L) | IDL |
| 1 pg/L | 10 fg | 43% | **1.6** | 16 | 84% | 3.1 | 31 | **15%** | **0.6** | **6** |
| 2.5 pg/L | 25 fg | **17%** | **1.6** | **16** | 24% | **2.3** | 23 | 9% | 0.8 | 8 |
| 10 pg/L | 100 fg | 7% | 63 | 63 | | | | 5% | 1.8 | 18 |

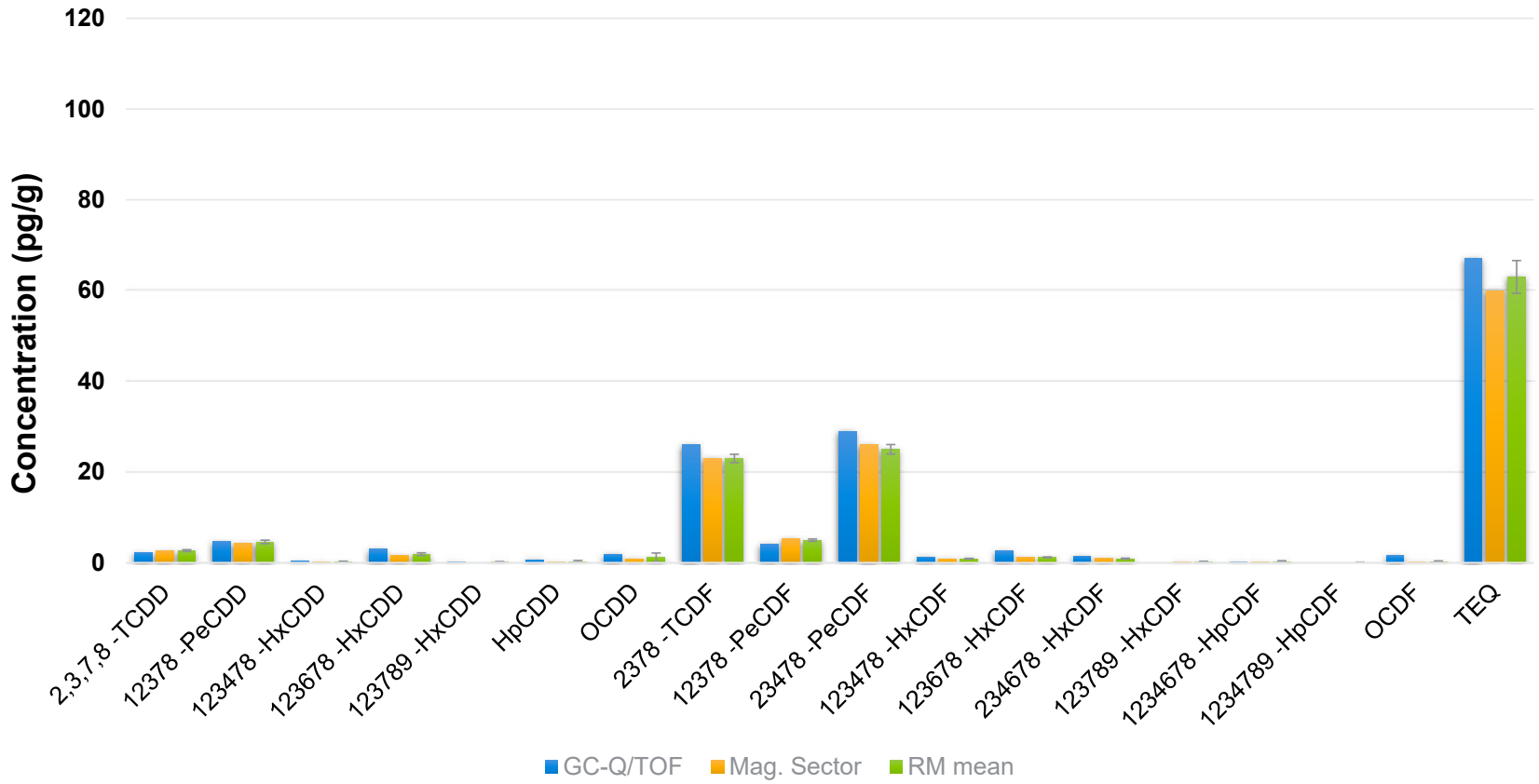
# Dioxins concentration in Herring (6.4% fat)

## Comparison between GC/Q-TOF & Magnetic Sector



# Dioxins in Sediment reference

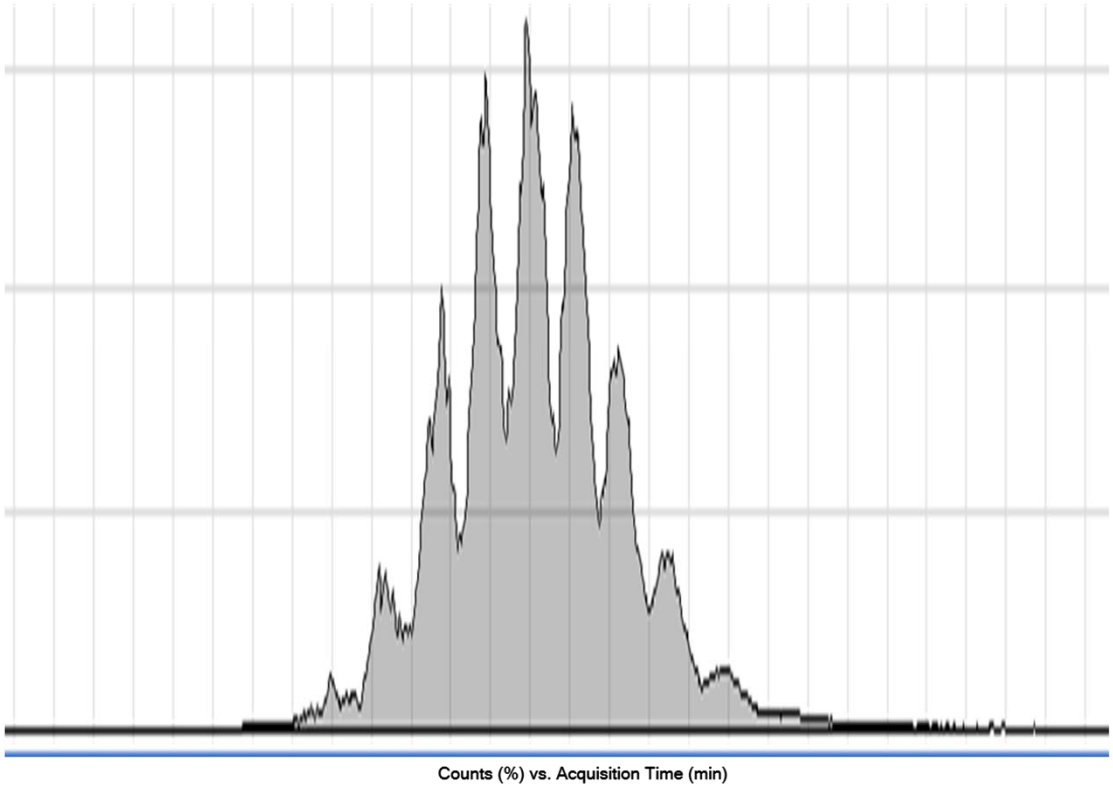
## Comparison between GC/Q-TOF & Magnetic Sector

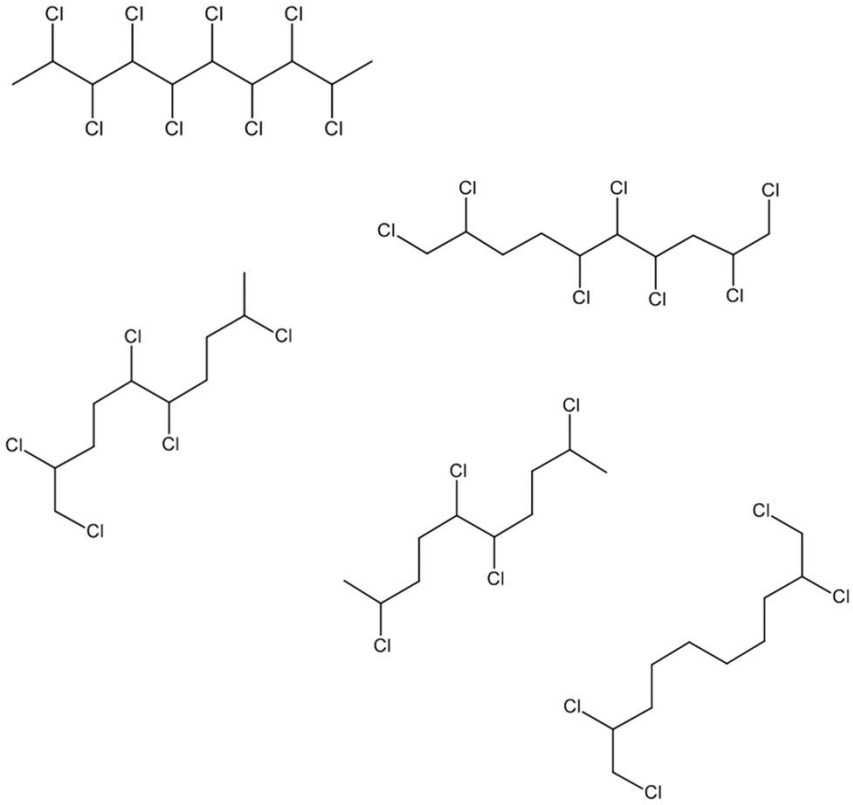


# Short Chain Chlorinated Paraffins (SCCPs)

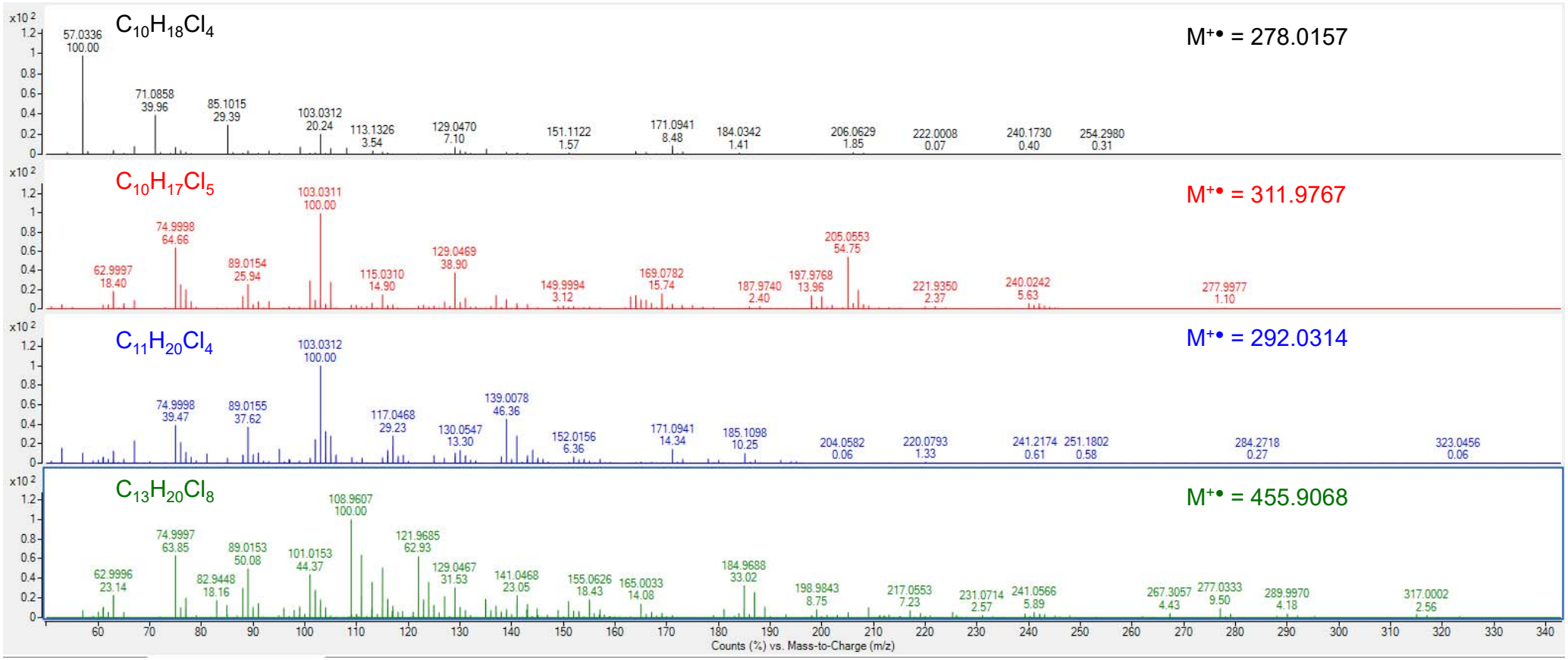
- SCCPs are emerging persistent organic pollutants, bioaccumulative, toxic and persistent in the environment
- It is a complex mixture of n-alkanes with different degree of chlorination
- Used as flame retardants in plastics and other materials, as well as in few other applications such as metal processing
- Represent a substantial challenge due to their self-interference as well as interference with other components of complex industrial matrices

# Challenges for the Analysis of the SCCPs: Chromatographic Separation



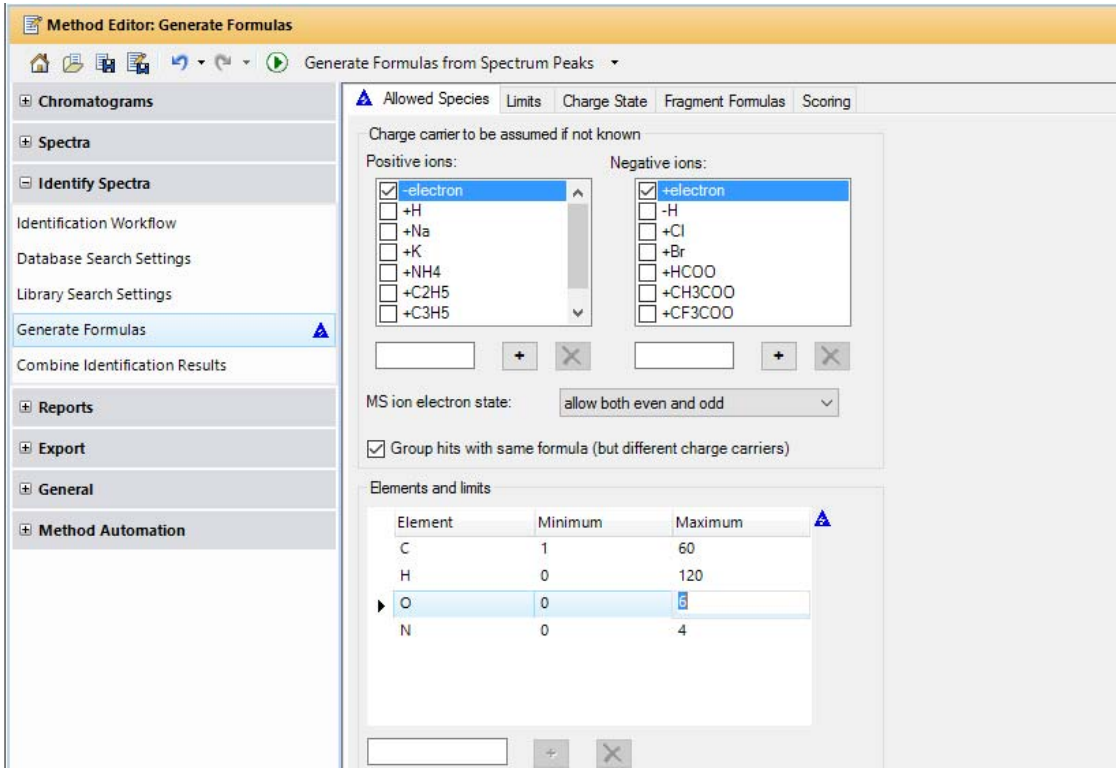



# Challenges for the Analysis of the SCCPs: Excessive Fragmentation



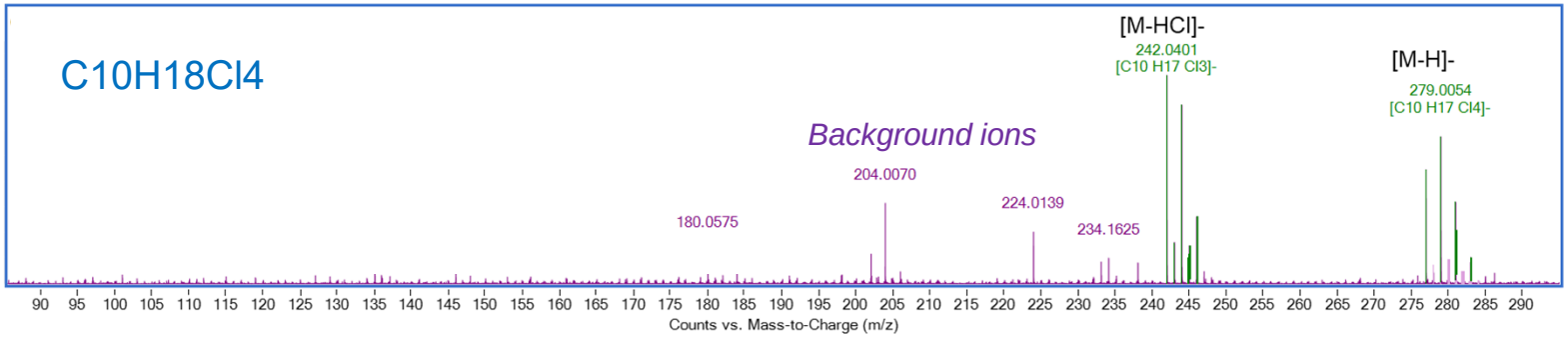

- Spectral fragmentation under normal conditions for 70 eV acquisitions
- The number below the *m/z* is the relative intensity

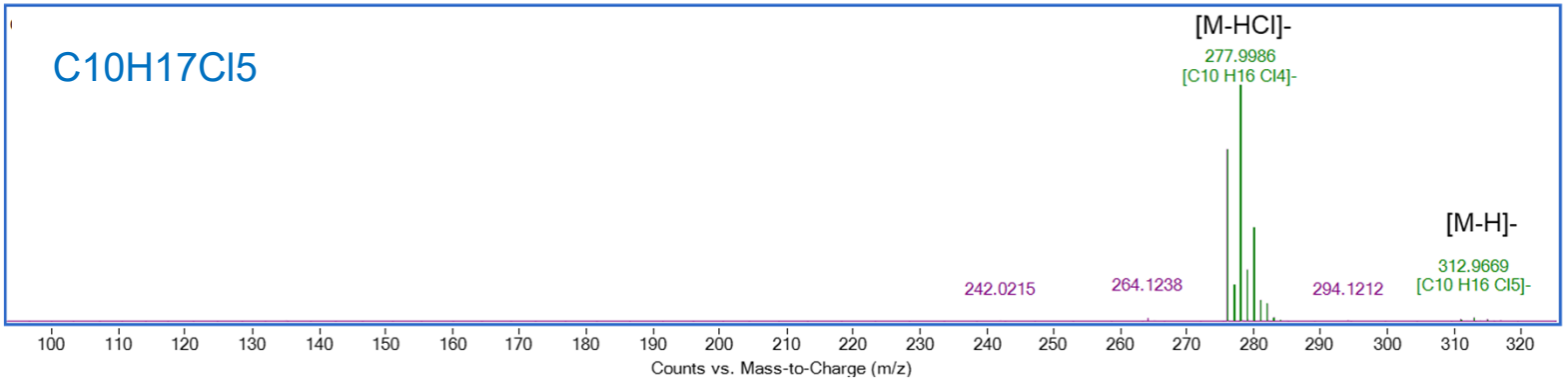
# Automatic Molecular Formula Generation



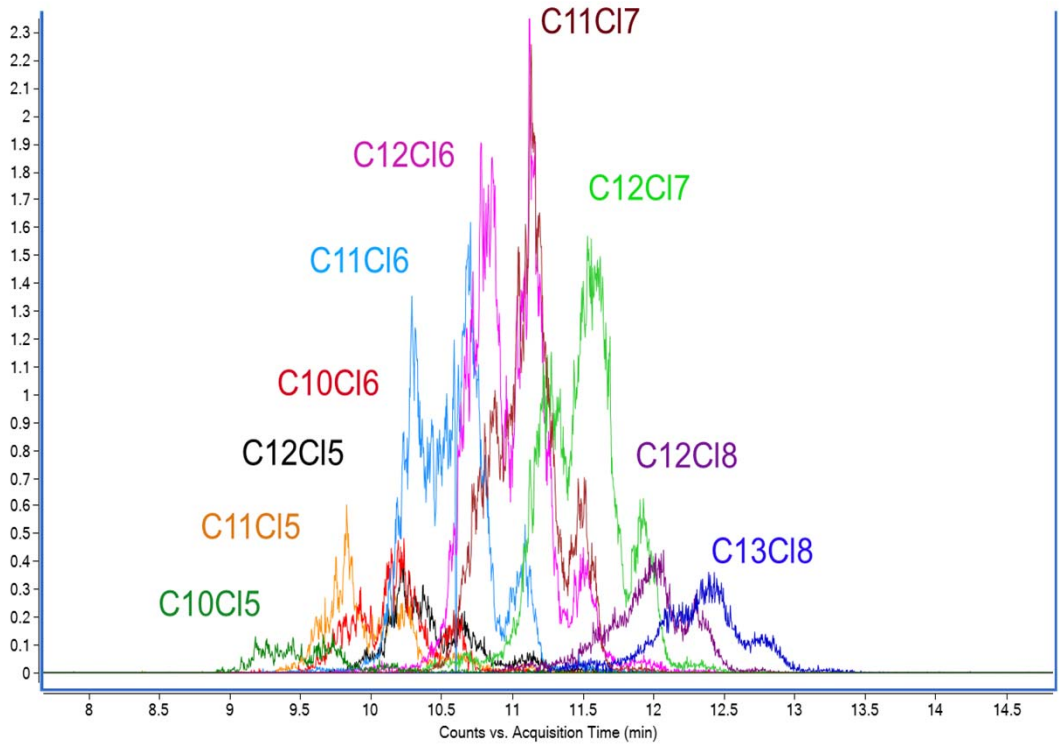

*Molecular Formula Generation uses accurate m/z of the molecular ion, accurate m/z of the isotopes, isotope spacing and isotope intensity for confirmation.*

# Examples of SCCP Spectra in NCI



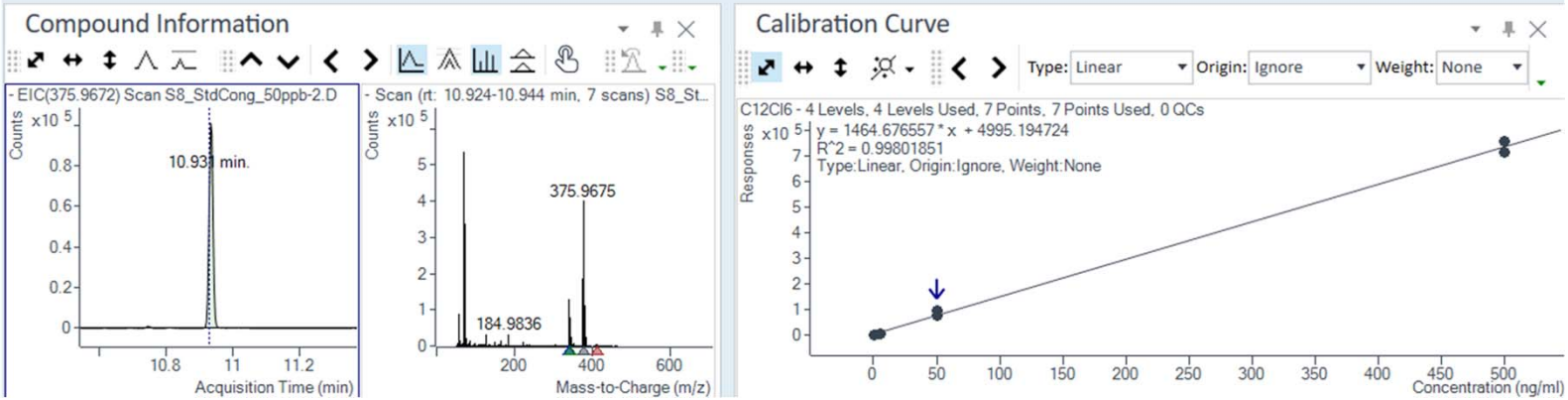



# EIC Overlay of SCCP mixture

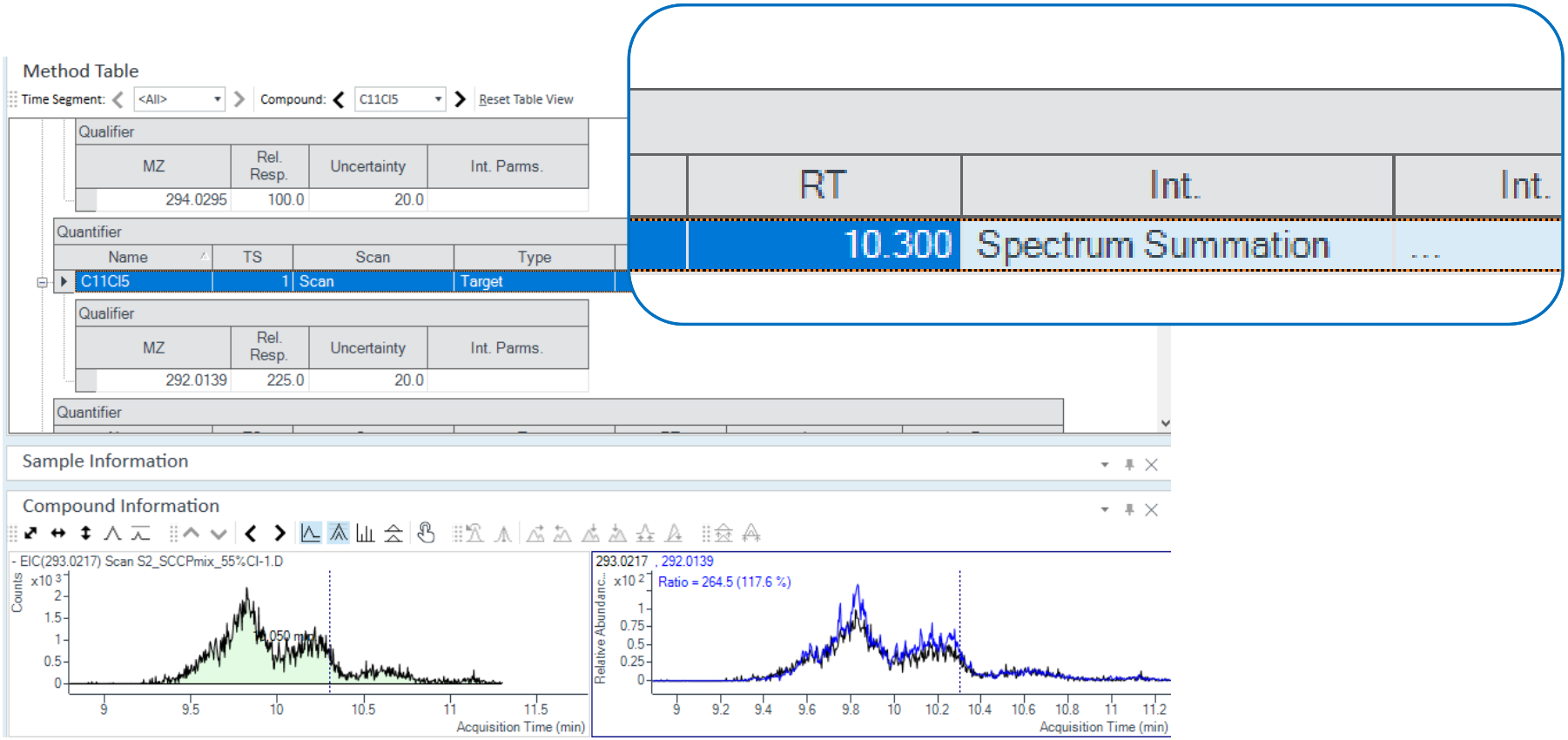


# Quantitation Based on Pure Congeners

| Sample | | | | C12Cl6... | C12Cl6 Results | | | | | Qualifier (339.9905) Resu | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Name | Type | Level | Exp. Conc. | RT | Resp. | Final Conc. | Mass Accuracy | S/N | MZ | Mass Accuracy | S/N |
| | S8_StdCong_50ppb-1 | Cal | 3 | 50.0000 | 10.9... | 78333 | 50.0713 | 0.9431 | 8669.78 | 339.9912 | 2.0387 | 1424 |
| | S8_StdCong_50ppb-2 | Cal | 3 | 50.0000 | 10.9... | 96498 | 62.4729 | 0.6964 | 8225.01 | 339.9908 | 0.7971 | 3467 |
| | S9_StdCong_500ppb-1 | Cal | 4 | 500.0000 | 10.9... | 758521 | 514.4659 | -4.3322 | 20306.... | 339.9892 | -3.9533 | 5015 |
| | S9_StdCong_500ppb-2 | Cal | 4 | 500.0000 | 10.9... | 714430 | 484.3629 | -1.6121 | 20703.... | 339.9902 | -0.8431 | 4520 |
| | S1_SCCPmix_51p5%C... | Sample | | | 10.9... | 298472 | 200.3700 | -1.1223 | 8.29 | 339.9906 | 0.3375 | 12 |
| | S1_SCCPmix_51p5%C... | Sample | | | 10.9... | 314725 | 211.4666 | -1.3633 | 9.60 | 339.9906 | 0.3773 | 13 |



# Challenges of SCCP Quantitation



# Quant Results: Comparison of 55.5% and 63% SCCP Mixtures

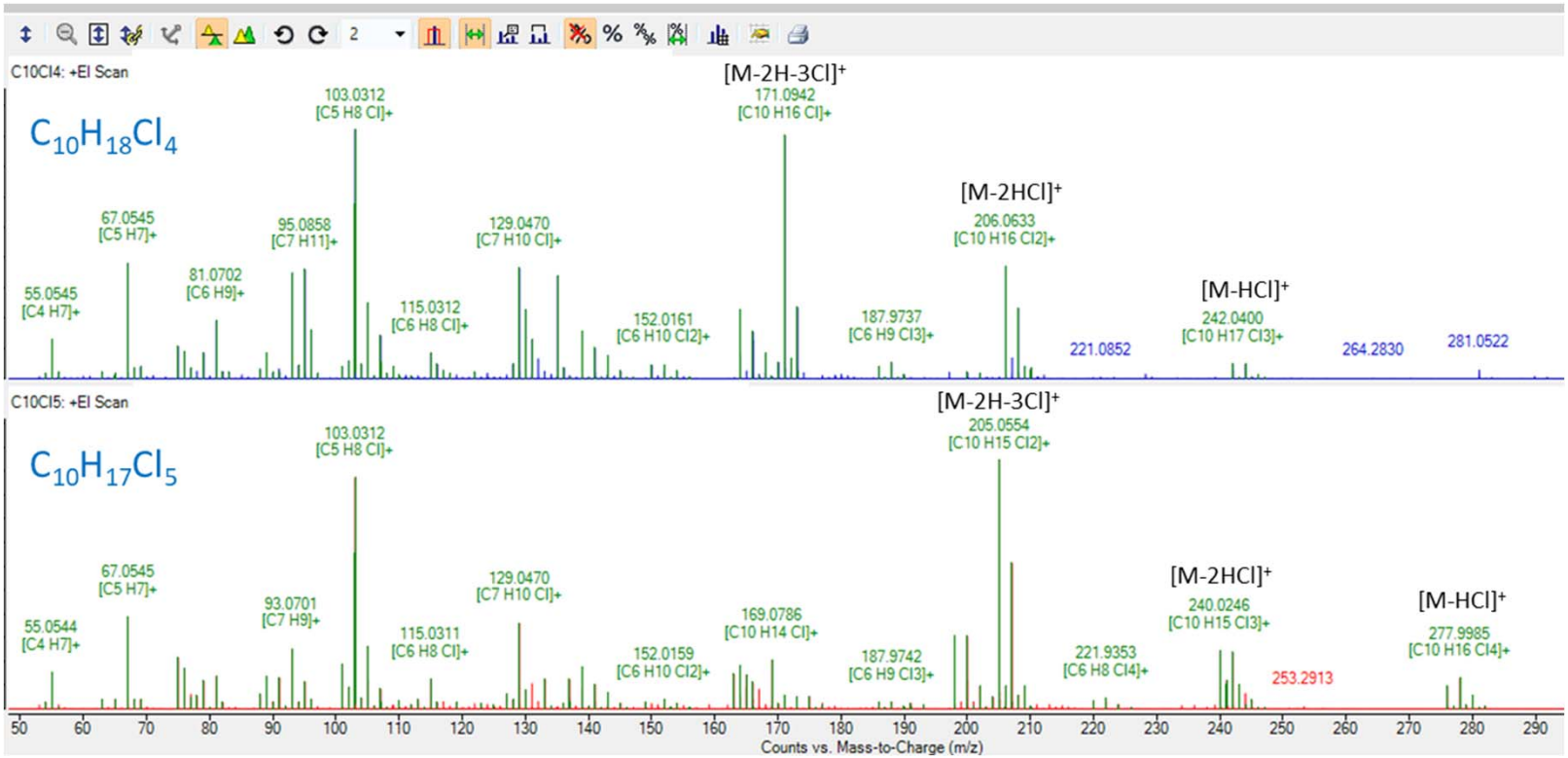
| Congener | RT range, min | Concentration, ppb | | | % | | |
|---|---|---|---|---|---|---|---|
| | | 51.50% | 55.50% | 63% | 51.50% | 55.50% | 63% |
| $C_{10}Cl_4$ | 8.8-9.1 | 115.5 | 193.7 | 23 | 2.3 | 3.9 | 0.5 |
| $C_{10}Cl_5$ | 9-10.3 | 106.1 | 135.3 | 84.3 | 2.1 | 2.7 | 1.7 |
| $C_{10}Cl_6$ | 9.6-10.8 | 5.9 | 15.3 | 41.7 | 0.1 | 0.3 | 0.8 |
| $C_{10}Cl_7$ | 10.1-11.2 | 0.9 | 6.7 | 51.6 | 0.02 | 0.1 | 1 |
| $C_{10}Cl_8$ | 10-11.3 | 2.5 | 4.4 | 38 | 0.05 | 0.1 | 0.8 |
| $C_{11}Cl_4$ | 9.2-10 | 189.2 | 96.2 | 36.6 | 3.8 | 1.9 | 0.7 |
| $C_{11}Cl_5$ | 9.5-10.5 | 364.6 | 340.7 | 89.4 | 7.3 | 6.8 | 1.8 |
| $C_{11}Cl_6$ | 10-10.8 | 342 | 614.5 | 330.3 | 6.8 | 12.3 | 6.6 |
| $C_{11}Cl_7$ | 10.5-11.7 | 70.4 | 353.2 | 825.9 | 1.4 | 7.1 | 16.5 |
| $C_{11}Cl_8$ | 11-12.5 | 3.3 | 25.4 | 210.6 | 0.1 | 0.5 | 4.2 |
| $C_{12}Cl_4$ | 9.4-10.5 | 290.7 | 129.8 | 11.1 | 5.8 | 2.6 | 0.2 |
| $C_{12}Cl_5$ | 10-11.2 | 351.3 | 253.7 | 31.3 | 7.0 | 5.1 | 0.6 |
| $C_{12}Cl_6$ | 10.3-11.5 | 205.9 | 240.2 | 46.8 | 4.1 | 4.8 | 0.9 |
| $C_{12}Cl_7$ | 10.9-12.1 | 331.9 | 733.3 | 763.7 | 6.6 | 14.7 | 15.3 |
| $C_{12}Cl_8$ | 11.4-12.6 | 9.5 | 49.3 | 167.3 | 0.2 | 1 | 3.3 |
| $C_{13}Cl_5$ | 10.1-11.3 | 218.8 | 126.5 | 12.3 | 4.4 | 2.5 | 0.2 |
| $C_{13}Cl_6$ | 10.8-11.8 | 200.9 | 161.9 | 26.1 | 4 | 3.2 | 0.5 |
| $C_{13}Cl_7$ | 11.4-12.5 | 642.3 | 865.9 | 497.4 | 12.8 | 17.3 | 9.9 |
| $C_{13}Cl_8$ | 11.9-13 | 84.9 | 287.8 | 628.2 | 1.7 | 5.8 | 12.6 |
| **Total** | | | | | **70.7** | **92.7** | **78.2** |

# Low Energy EI Analysis of SCCP

- To improve sensitivity of detection and accuracy of quantitation for SCCP congeners with low chlorine content, the low energy EI approach was used.
- Traditional 70 eV EI results in a high degree of fragmentation of SCCP molecules, and does not provide enough unique ion clusters for individual identification.
- Multiple low electron energy settings were evaluated to determine the optimal value.
- The optimum combination of spectral tilt and signal response was achieved with an electron energy set at 22 eV.

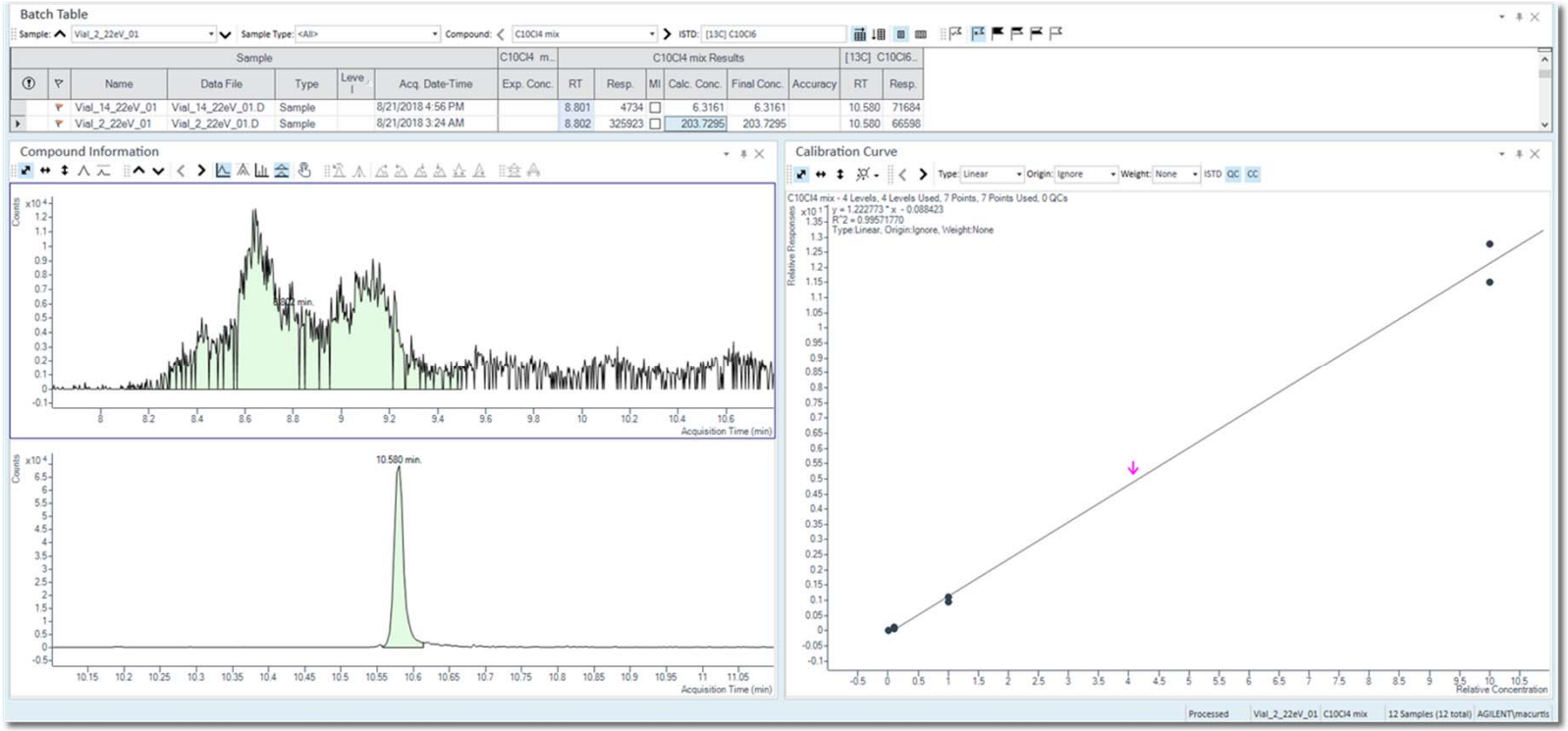
# Low Energy EI Analysis of SCCP

- Low energy EI data indicated a higher degree of fragmentation of the SCCP molecules compared to negative CI



# Low Energy EI Analysis of SCCP

- However, this technique allowed more sensitive detection of the SCCP species with low chlorine atom number (such as C10Cl4)



# Perfluoroalkyl Substances (PFAS)

## Analysis of Fluorinated Alkyl Alcohols in Biosolids

# Standards & Samples

Standards at 100 ng/µL in MTBE

Biosolid extracts prepared in EtOAc

Positive CI, 20% Methanol

MMI: 2 µL cold, splitless injection

65 ºC (0.01 min), 300 ºC/min to 250 ºC

Two min post-column backflush

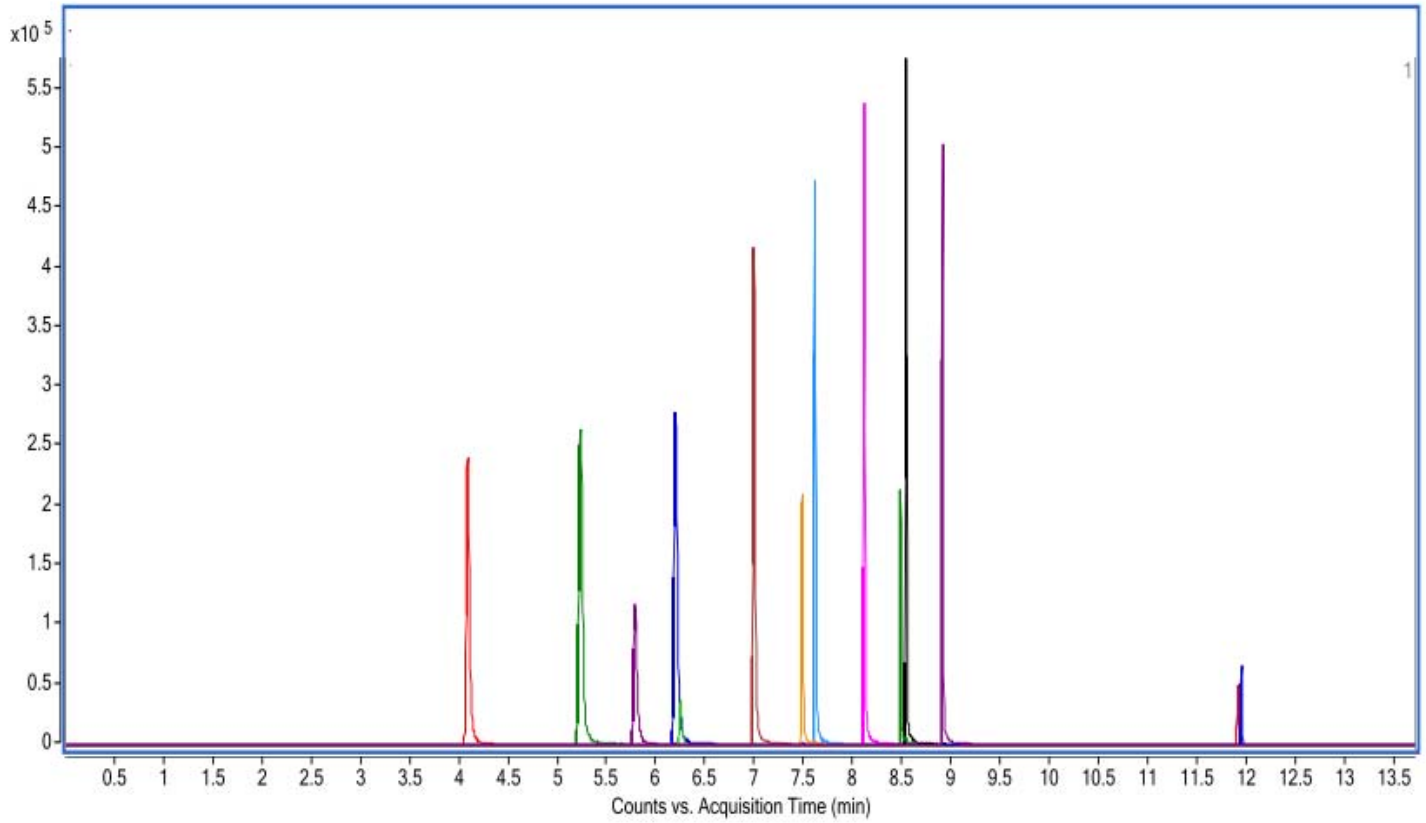
Oven Ramp:

- 60 ºC for 1 min
- 3 ºC/min to 75ºC for 0 min
- 20 ºC/min to 210 ºC for 0 min

| Acronym | Type | r.t. | Exact Mass + H |
|---|---|---|---|
| 4:2 FTOH | Target | 4.078 | 265.0269 |
| 5:1 FTOH | Target | 5.221 | 301.0081 |
| 6:2 FTOH | Target | 5.773 | 365.0206 |
| 6:1 FTOH | Target | 6.188 | 351.0049 |
| 7:2 sFTOH | Target | 6.237 | 415.0174 |
| 7:1 FTOH | Target | 6.981 | 401.0017 |
| 8:2 MFTOH | ISTD | 7.449 | 469.0334 |
| 8:2 FTOH | Target | 7.471 | 465.0142 |
| 8:1 FTOH | Target | 7.598 | 450.9985 |
| 9:1 FTOH | Target | 8.098 | 500.9953 |
| 10:2 FTOH | Target | 8.470 | 565.0078 |
| 10:1 FTOH | Target | 8.523 | 550.9921 |
| 11:1 FTOH | Target | 8.886 | 600.9889 |
| d7-MeFOSE | ISTD | 11.868 | 565.0466 |
| MeFOSE | Target | 11.889 | 558.0026 |
| d9-EtFOSE | ISTD | 11.897 | 581.0748 |
| EtFOSE | Target | 11.928 | 572.0183 |

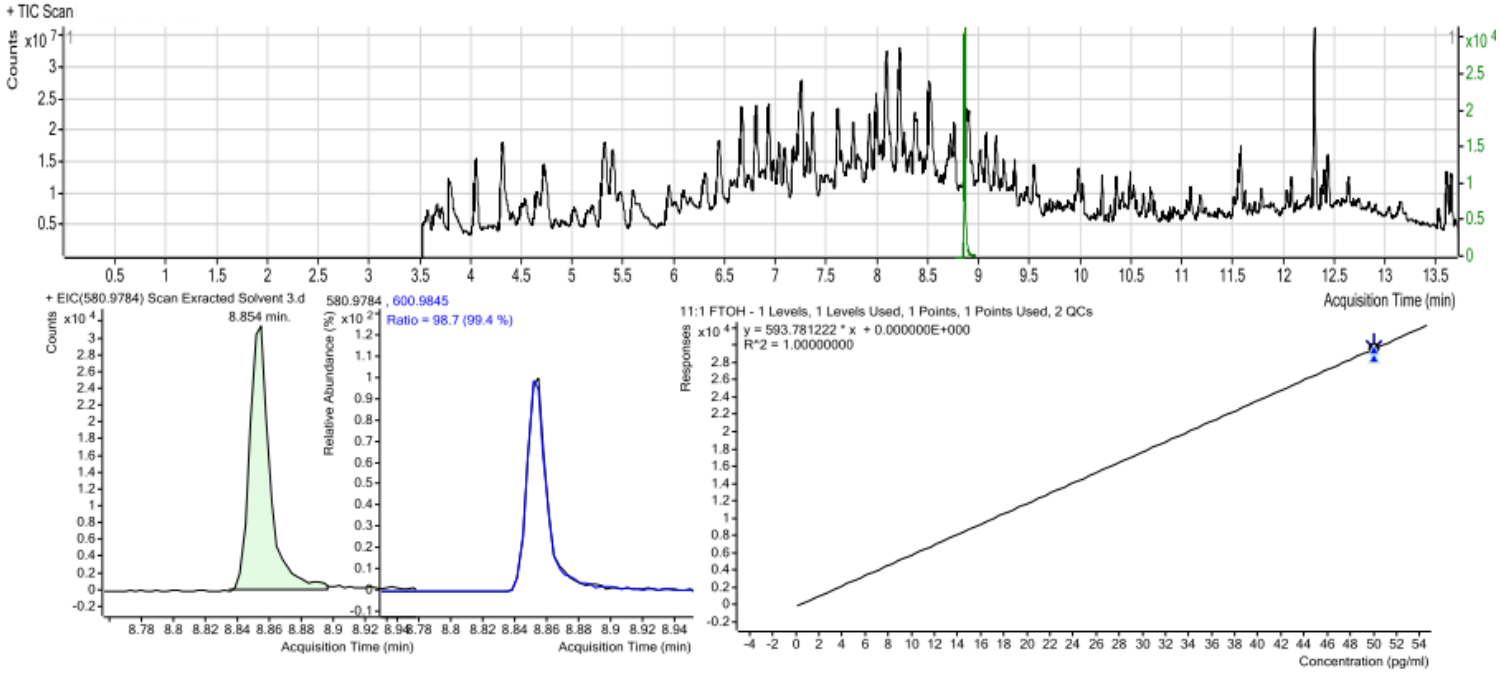
# Chromatogram

## 17 FTOHs & FOSEs



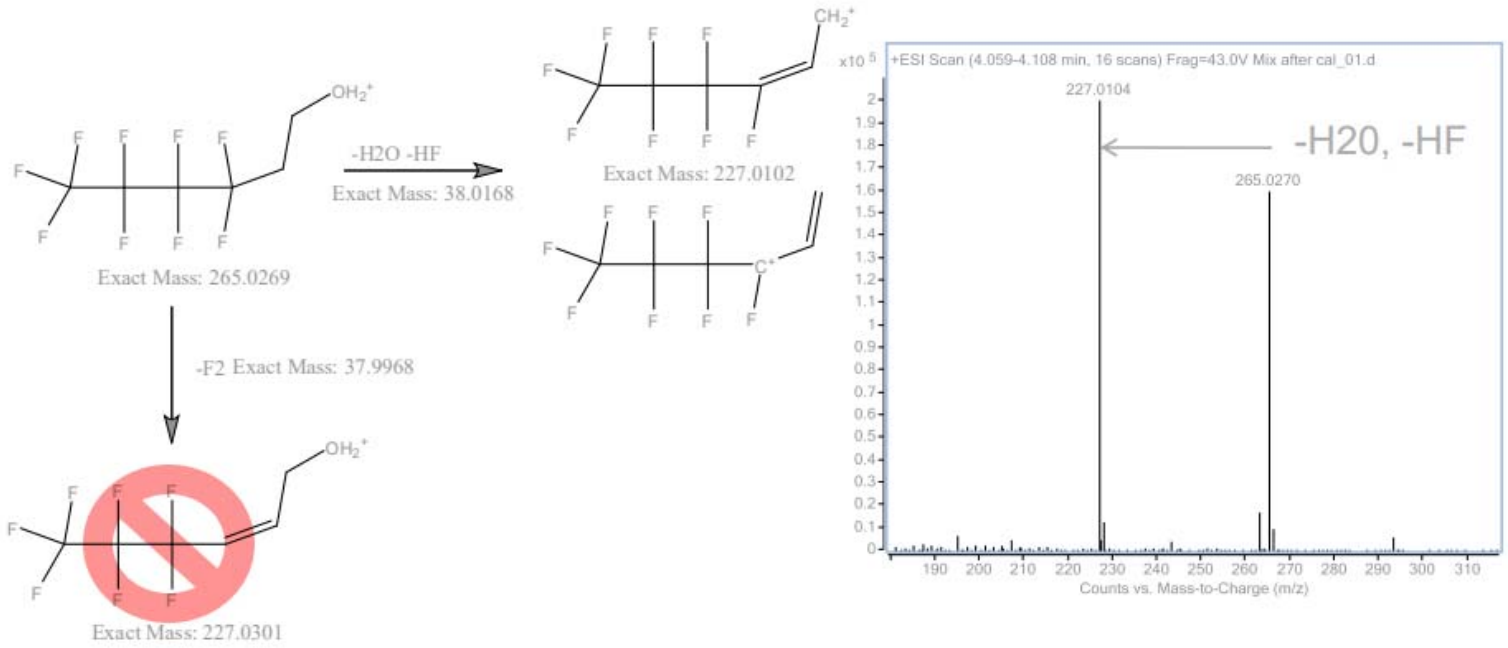
# Structure Elucidation

## 11:1 FTOH



# Structural Elucidation: Why use a HRAM?

Nominal loss = 38 m/z; $F_2$ or something else?




| Acronym | Observed Base Peak m/z | Molecular ion -$F_2$ m/z | Δppm | Molecular ion -$H_20$, -HF m/z | Δppm |
|---|---|---|---|---|---|
| 4:2 FTOH | 227.0104 | 227.0301 | 86.7726 | 227.0102 | -0.8810 |

# Loss Elucidation

## Why use a 'Q'-TOF

Mass accuracy facilitated in structural elucidation of fragment ions

- Nominal mass loss of m/z 38 for n:2 FTOHs (n=4,6,8,10…)
  - Loss of $F_2$ ruled out due to mass accuracy
  - Loss of $H_2O$ and HF confirmed
  - **MS/MS confirmed $H_2O$ and HF Losses**

- Loss of HF for n:1 FTOH
- Loss of $H_2O$ for perfluorosulfonamidoethanols

# Identifying PFAS with loss elucidation

## Mass Accuracy

| Acronym | Formula | Loss | Exact Mass-Loss | Observed Mass 2 | Δppm 2 |
|---|---|---|---|---|---|
| 4:2 FTOH | $C_6H_5F_9O$ | -H2O, -HF | 227.0102 | 227.0105 | -1.3215 |
| 6:2 FTOH | $C_8H_5F_{13}O$ | -H2O, -HF | 327.0038 | 327.0040 | -0.6116 |
| 8:2 FTOH | $C_{10}H_5F_{17}O$ | -H2O, -HF | 426.9972 | 426.9975 | -0.7026 |
| 10:2 FTOH | $C_{12}H_5F_{21}O$ | -H2O, -HF | 526.9910 | 526.9915 | -0.9488 |
| 7:2 sFTOH | $C_9H_5F_{15}O$ | -H2O, -HF | 377.0006 | 377.0011 | -1.3263 |
| 5:1 FTOH | $C_6H_3F_{11}O$ | -HF | 281.0017 | 281.0019 | -0.7117 |
| 6:1 FTOH | C7 H3 F13 O | -HF | 330.9988 | 330.9990 | -0.6042 |
| 7:1 FTOH | C8 H3 F15 O | -HF | 380.9954 | 380.9956 | -0.5249 |
| 8:1 FTOH | C9 H3 F17 O | -HF | 430.9923 | 430.9927 | -0.9281 |
| 9:1 FTOH | C10 H3 F19 O | -HF | 480.9894 | 480.9896 | -0.4158 |
| 10:1 FTOH | C11 H3 F21 O | -HF | 530.9860 | 530.9864 | -0.7533 |
| 11:1 FTOH | C12 H3 F23 O | -HF | 580.9834 | 580.9835 | -0.1721 |
| MeFOSE | C11 H8 F17 N O3 S | $-H_2O$ | 539.9920 | 539.9943 | -4.2593 |
| EtFOSE | $C_{12}H_{10}F_{17}NO_3S$ | $-H_2O$ | 554.0077 | 554.0100 | -4.1516 |

# Summary

- The GC/Q-TOF is a complementary tool for environmental monitoring that is capable of unique identification of regulated and emerging environmental contaminants.
- The 7250 GC/Q-TOF is a versatile instrument that can analyze various classes of POPs including Dioxins, SCCPs, PCBs and PFAs on the same instrument

# Acknowledgements

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