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Utilization of Multiple Separation and Ionization Techniques on a Single High Resolution Mass Spectrometer for Comprehensive Screening of Environmental Water Samples with a Focus on Perfluoralkyl Substances

Ken Rosnack

ken_rosnack@waters.com

Principal Business Development Manager

Food & Environmental Markets

Overview

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- General PFAS Introduction
- Experimental
  - UPLC with PFC Kit
  - High Resolution MS with Ion Mobility (IMS)
  - Atmospheric Pressure Gas Chromatography
- Targeted vs Non-Target (Suspect) vs Unknown Screening
- High Efficiency Ion Mobility
  - Collisional Cross Section (CCS)
  - Spectral Cleanup
- PFAS in Water Example
  - UPLC ESI-
  - APGC+

Introduction: Poly and perfluoroalkyl substances (PFASs)

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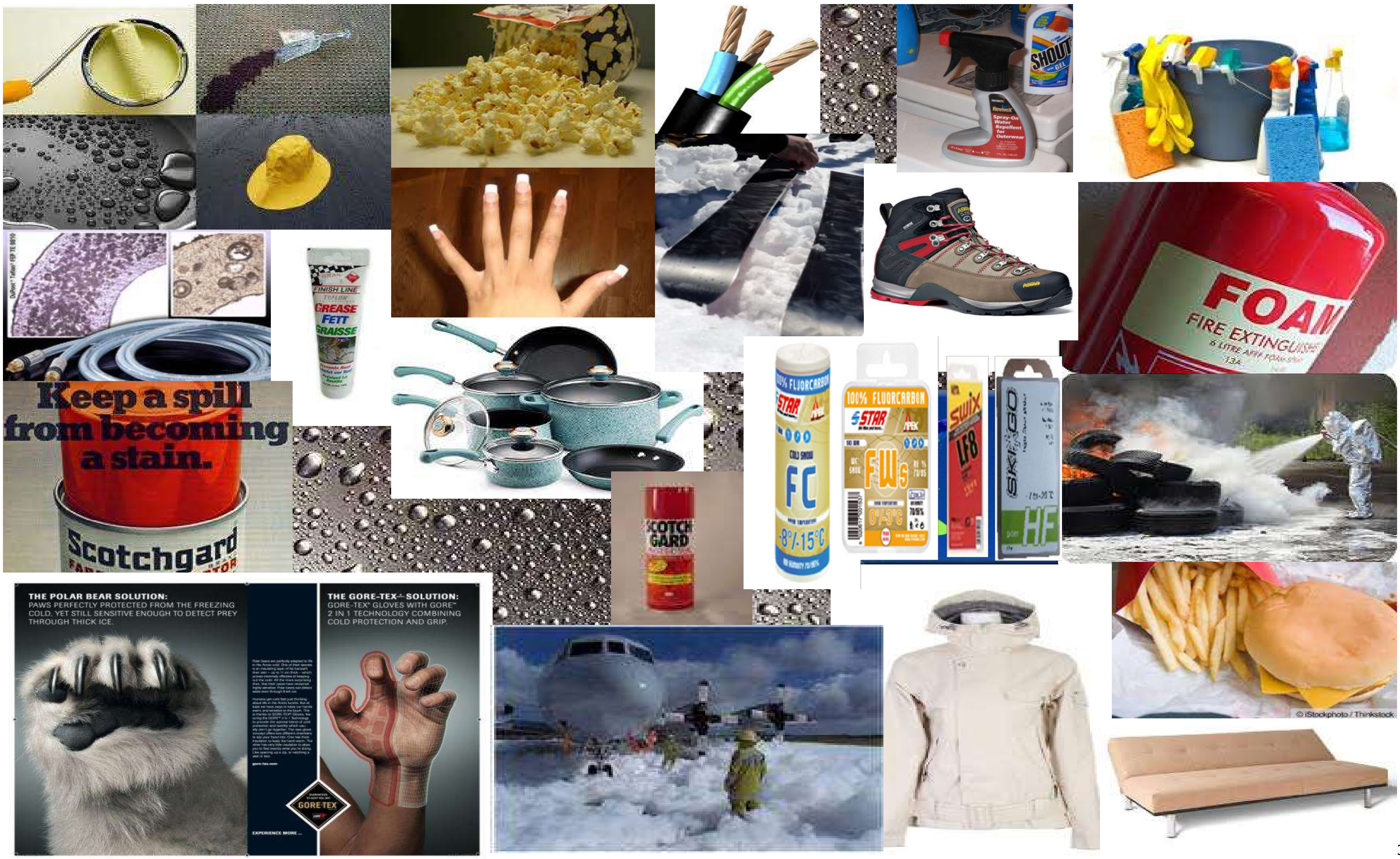
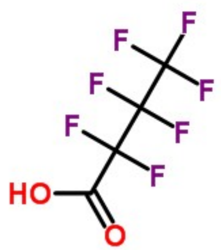
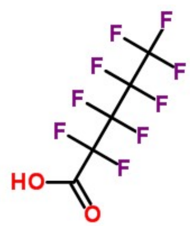


Standards in PFC Analysis Kit

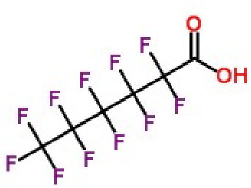
Structures Courtesy of ChemSpider



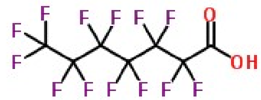
Perfluorobutyric acid (C4HF7O2)



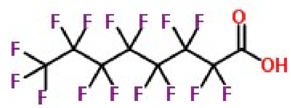
Perfluoropentanoic acid (C5HF9O2)



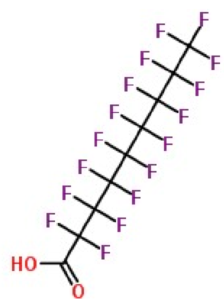
Perfluorohexanoic acid (C6HF11O2)



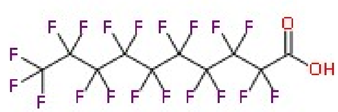
Perfluoroheptanoic acid (C7HF13O2)



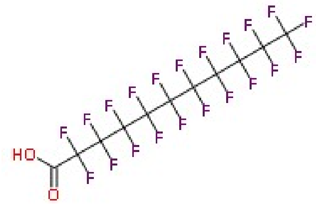
Perfluorooctanoic acid (C8HF15O2)



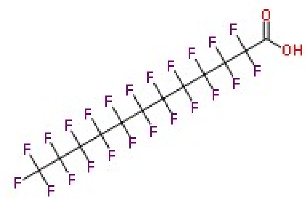
Perfluorononanoic acid (C9HF17O2)



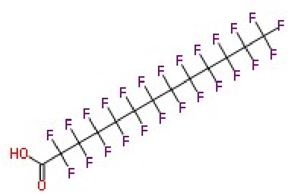
Perfluorodecanoic acid (C10HF19O2)



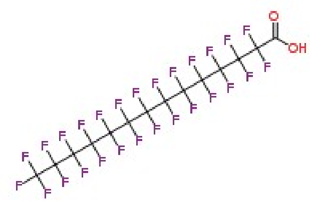
Perfluoroundecanoic acid (C11HF21O2)



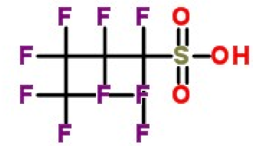
Perfluorododecanoic acid (C12HF23O2)



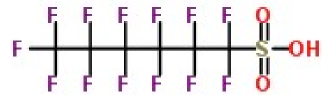
Perfluorotridecanoic acid (C13HF25O2)



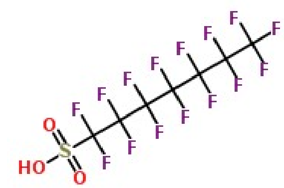
Perfluorotetradecanoic acid (C14HF27O2)



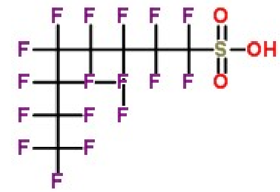
Perfluorobutanesulfonic acid (C4HF9O3S)



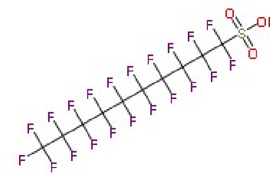
Perfluorohexanesulfonic acid (C6HF13O3S)



Perfluoroheptanesulfonic acid (C7HF15O3S)



Perfluorooctanesulfonic acid (C8HF17O3S)



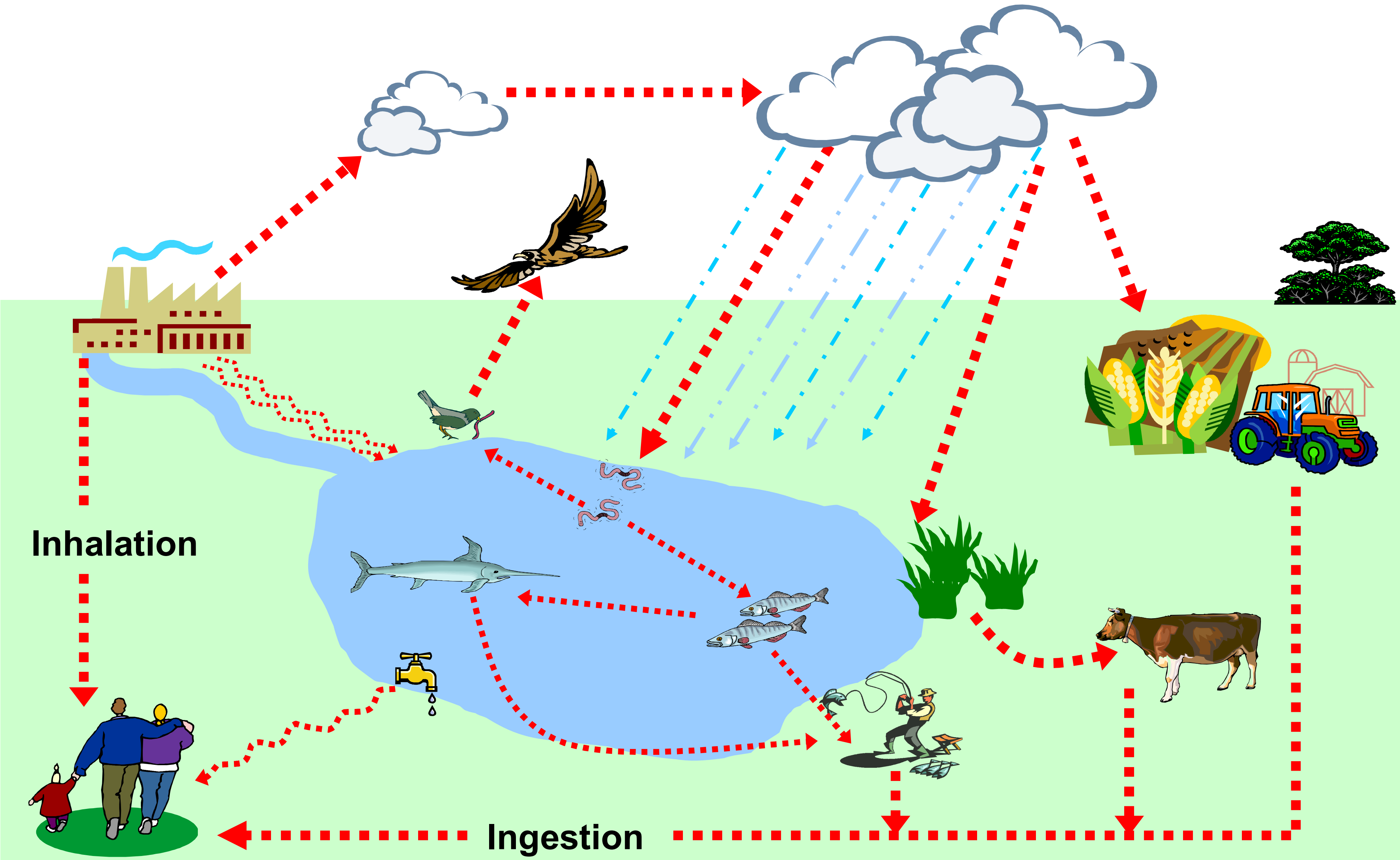
Perfluorodecanesulfonic acid (C10HF21O3S)

ISs in PFC Analysis Kit

Perfluorooctanoic acid (1,2,3,4-$^{13}C_2$)

Perfluorooctanesulfonic acid (1,2,3,4-$^{13}C_2$)

Water is One Main Route to Human Exposure of PFASs



PFOS Analysis Challenges

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- Matrix effects, retention time shifts.
- Correct PFOS isomer identification:
  - The physical, chemical and biological properties likely affected by perfluoromethyl branching.
  - Source elucidation.
  - Response factors of individual isomers.
- Increased scientific interest in toxicity, environmental transport, degradation and bioaccumulation of isomers.
- Interferences can be mistaken for PFOS and lead to a positive bias.

Residue Screening Classification

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- **Targeted screening**
  - Well defined target list of analytes
  - Selective acquisition &/or processing modes
  - Analytical standards available for every compound
- **Non-targeted (suspect) screening**
  - Usually in combination with targeted screening
  - Screen against a comprehensive library of known compounds
  - Analytical standards available for many compounds
- **Unknown screening**
  - No defined target list
  - Compound not present in the library, maybe a new chemical structure
  - Structural elucidation required

Non-Target Screening

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Manage Components

Create | Import | Paste Results | Delete | Edit Fragments... | Edit Adducts... | Add To Common Fragments | Add To Neutral Losses

	Component name	Expected RT (min)	Expected neutral mass (Da)	Expected fragment (m/z)	Excluded	Formula
1	Perfluorotetradecanoic acid	4.68	713.9545	668.9574, 127.0012, 168.9894, 218.9862	☐	C14HF27O2
2	Perfluorotridecanoic acid	4.57	663.9577	618.9606, 168.9894, 218.9855, 118.9926	☐	C13HF25O2
3	Perfluorododecanoic acid	4.43	613.9609	568.9638, 168.9894, 118.9926	☐	C12HF23O2
4	Perfluorodecanesulfonic acid	4.28	599.9311	79.9574, 229.9478, 279.9446, 168.9894	☐	C10HF21O3S
5	Perfluoroundecanoic acid	4.27	563.9641	518.9670, 168.9894, 118.9926	☐	C11HF21O2
6	Perfluorodecanoic acid	4.09	513.9673	568.9638, 168.9894, 118.9926	☐	C10HF19O2
7	Perfluorooctanesulfonic acid	3.89	499.9375	79.9574, 168.9894, 129.9535, 197.9779	☐	C8HF17O3S
8	C13PFOS	3.89	503.9509		☐	12C4(13C) 4HF17O3S
9	Perfluorononanoic acid	3.86	463.9705	418.9734, 168.9894, 155.9840, 118.9926, 218.9881, 203.9652	☐	C9HF17O2
10	Perfluoroheptanesulfonic acid	3.64	449.9407	79.9574, 368.9766, 168.9894, 118.9926, 129.9542	☐	C7HF15O3S
11	Perfluorooctanoic acid	3.60	413.9737	368.9766, 168.9894,	☐	C8HF15O2

Tips for defining expected components

ⓘ Tips for using internal standards

Non-Target vs Target Screening

- Mink Liver Extract Example

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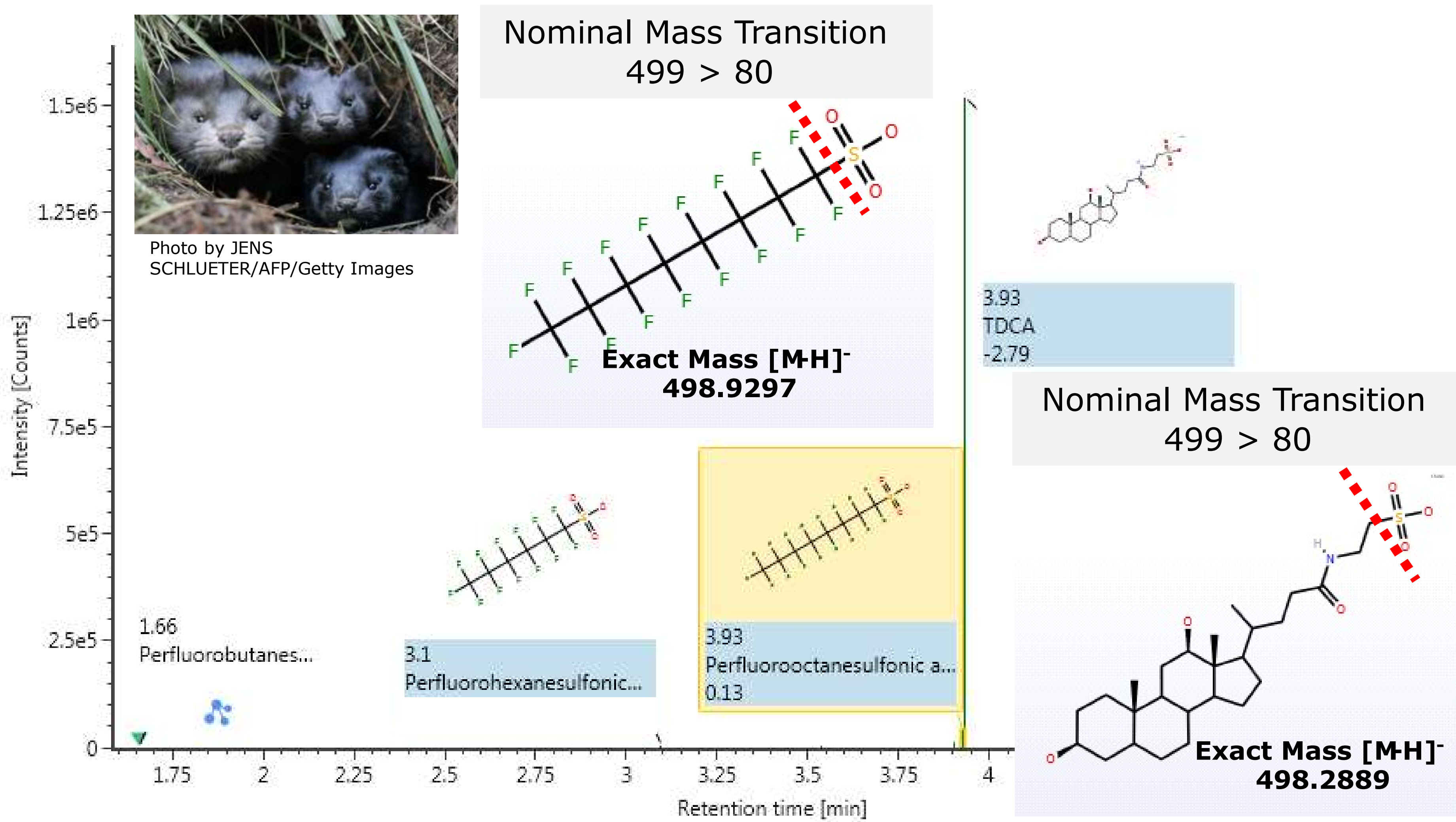

Ionization & Separation vs PFC type

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LC-ESI$^-$	APGC$^+$
Perfluorocarboxylic acids: C_{4-14}	Fluorotelomer Alcohols: 4:2, 6:2, 8:2, 10:2
Perfluorosulfonic acids: $C_{4,6,7,8,10}$	

UPLC PFC Isolation Kit

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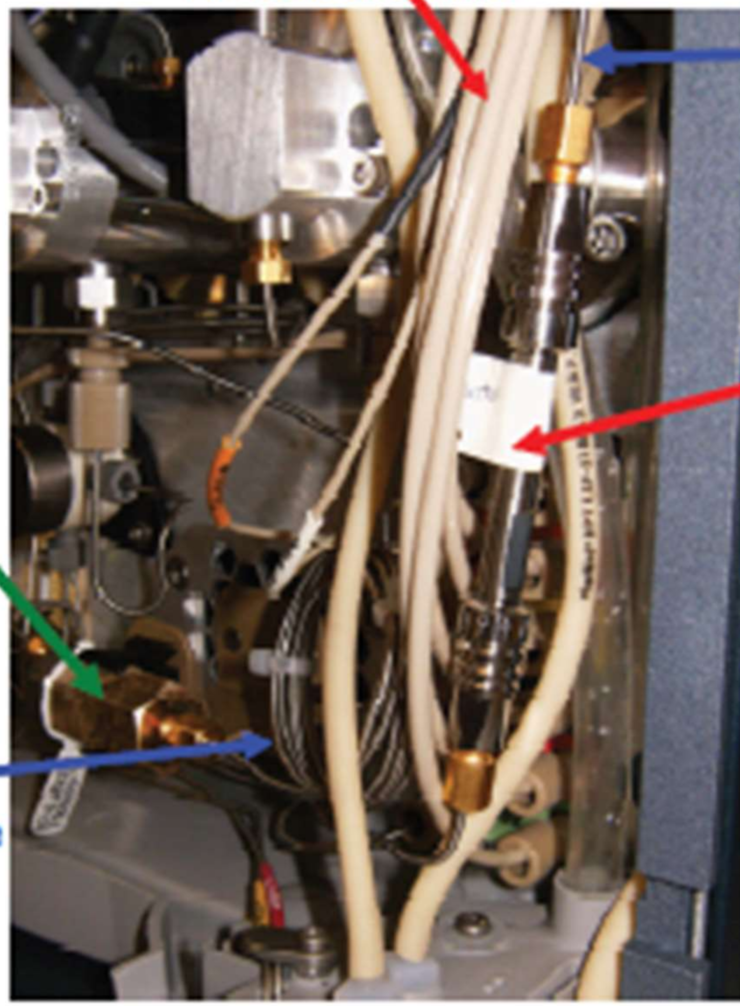

Water Extract Sample Preparation

1) The cartridge was first conditioned with 5 mL methanol, and 10 mL water.
2) Pass 250 mL water sample through HLB cartridge (200 mg).
3) Purge dry for 20 mins.
4) Elute with 2 mL methanol into PPE 15 mL collection tube.
5) Take 500 µL of this and dilute with 500 µL water (end result is a 125x enhancement)

Experimental
- LC Chromatography

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- LC system: ACQUITY UPLC I-Class with the PFC Analysis Kit
- Column: ACQUITY UPLC BEH C18, 1.7 µm 2.1 x 50 mm
- Column temp.: 55 ° C
- Mobile phase A: 98:2 Water:MeOH 2 mM ammonium acetate
- Mobile phase B: MeOH 2 mM ammonium acetate
- Gradient:

Min.	Flow Rate	%A	%B
Initial	0.65	90	10
0.5	0.65	90	10
5.1	0.65	0	100
6.6	0.65	0	100
6.7	0.65	90	10
8.5	0.65	90	10

Experimental
- APGC Chromatography

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- GC Type: Agilent 7890
- GC Injector: Multimode
- Injection Type: Splitless [Purge Flow 50 mL/min at 2.05 min]
- Injector Temp: 250 °C
- Injector Pres: 14.9 psi
- Septum Purge: 3 mL/min
- Transfer Line Temp: 240 °C
- GC Run Time: 19.6 min

	Rate	Value	Hold Time	Run Time
	°C/min	°C	min	min
(initial)	-	60	2	2
Ramp 1	10	200	0	16
Ramp 2	25	240	2	19.6

APGC Source Conditions

Corona Mode: Current
Corona Current: 3.0 µA
Sample Cone: 20 V
Source Temp: 150 °C
Cone Gas: 50 L/h
Aux Gas: 350 L/H

Portoles et al. Gas chromatography-tandem mass spectrometry with atmospheric pressure chemical ionization for fluorotelomer alcohols and perfluorinated sulfonamides determination. J. of Chrom. A 1413 (2015) 107-116

Atmospheric Pressure GC

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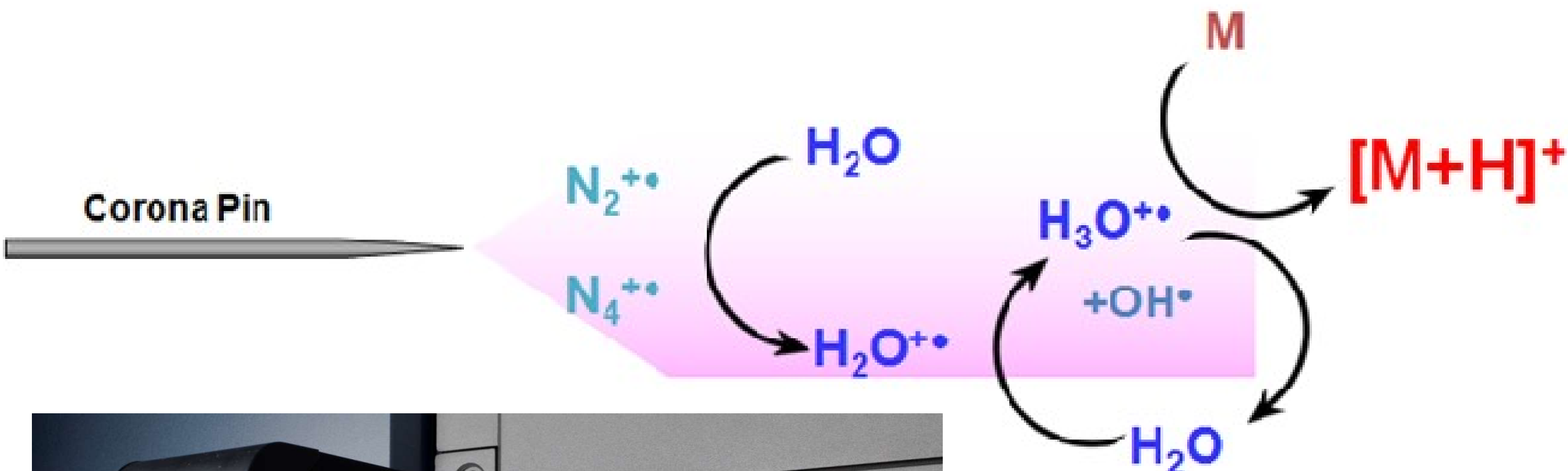
Protonation



Experimental
- Mass Spectrometer

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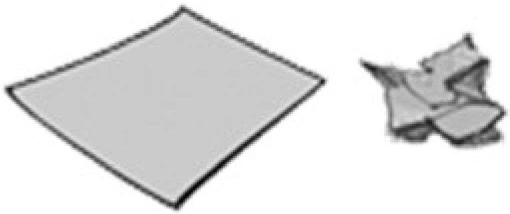
- MS system: SYNAPT G2-S
- Ionization mode: ES –
- Desolvation temp.: 550 ° C
- Acquisition mode: Ion mobility
- Mass range: 50 to 600 Da
- Acquisition rate: 10 spectra/s
- Capillary voltage: 2.3 kV
- Cone voltage: 15V
- Ion mobility gas: N_2
- Collision energy: 35 to 75 eV ramped
- IMS wave velocity: 650 m/s
- IMS wave height: 40 V
- IMS duty cycle: 10.8 ms
- Lock mass: Leucine enkephalin (554.2610)

Ion Mobility Spectrometry

- Separation of ions as they drift through a gas under the influence of an electric field
- Rate of drift is dependent on ion’s mobility through the gas
- Mobility is dependent on factors such as
  - Mass
  - Charge
  - Interaction Cross Section



Ion Mobility Spectrometry (IMS) - Another Separation Dimension

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Small, compact

Large, extended

SYNAPT G2-S High Definition MS
- Ion Mobility Explained

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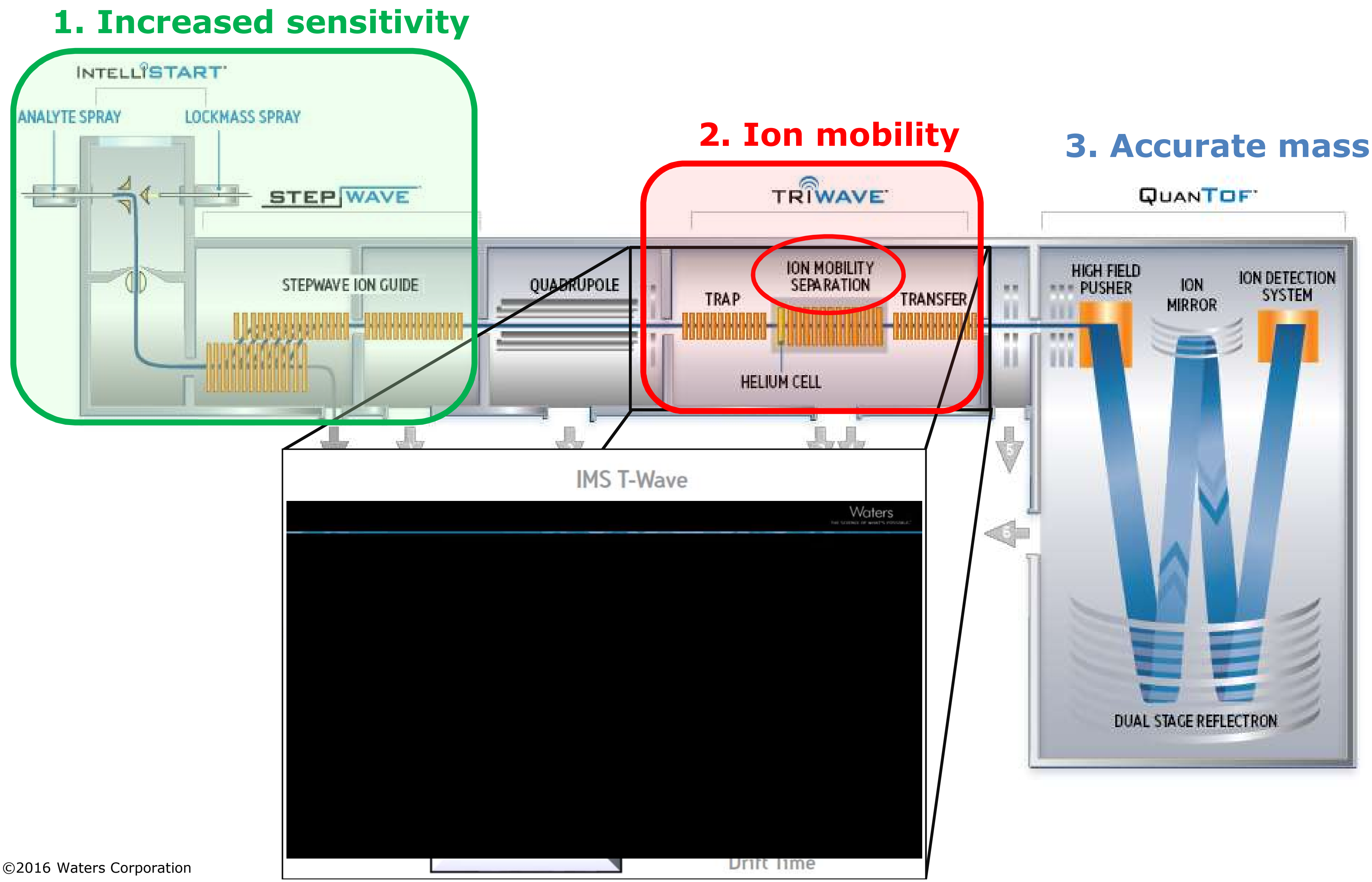

MS^E Time Aligned vs HDMS^E Time and Drift Aligned Spectra

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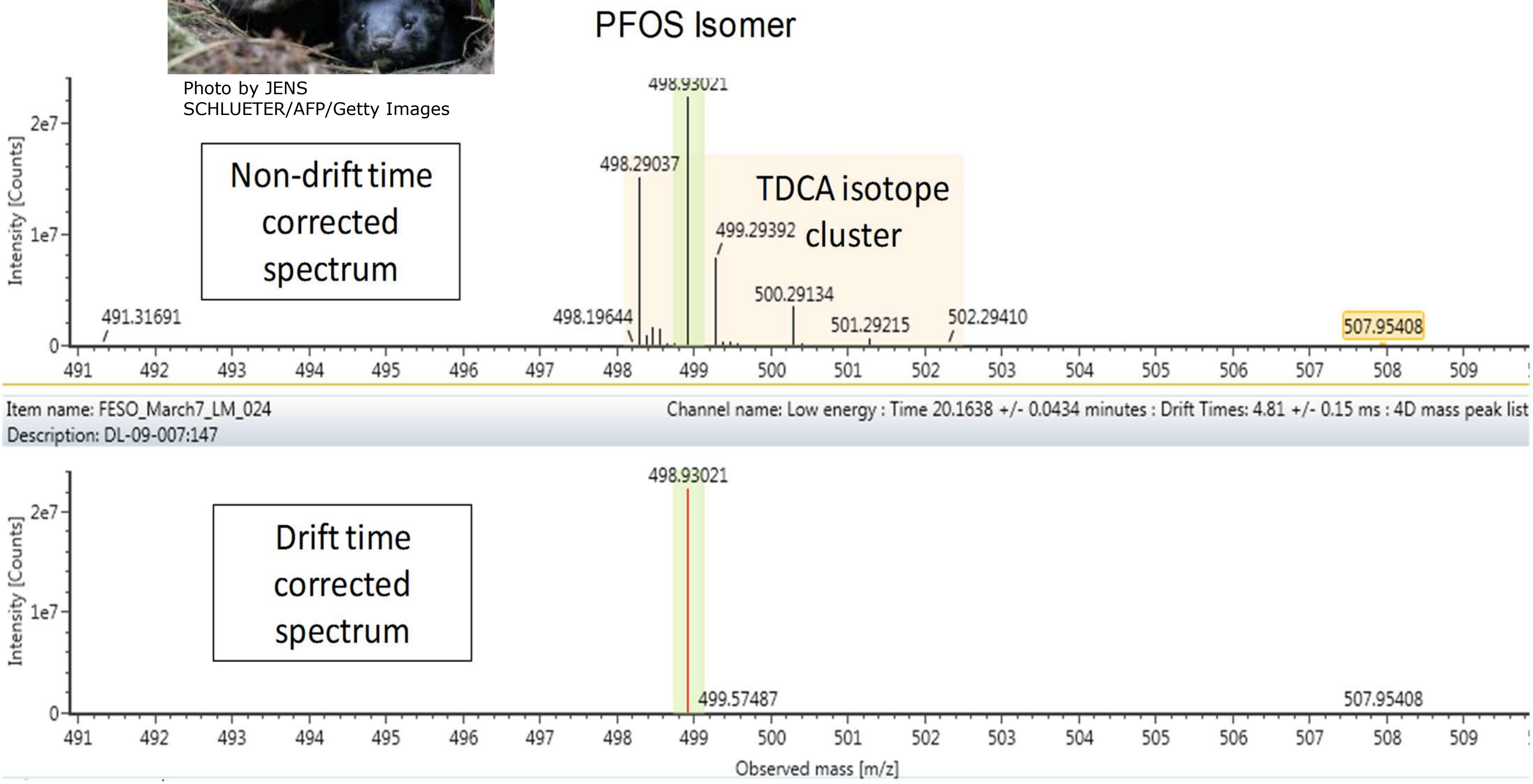
Photo by JENS SCHLUETER/AFP/Getty Images



UPLC ESI-: Lake Water (Non-Target)
- Componentized MS^E Spectra (PFOS)

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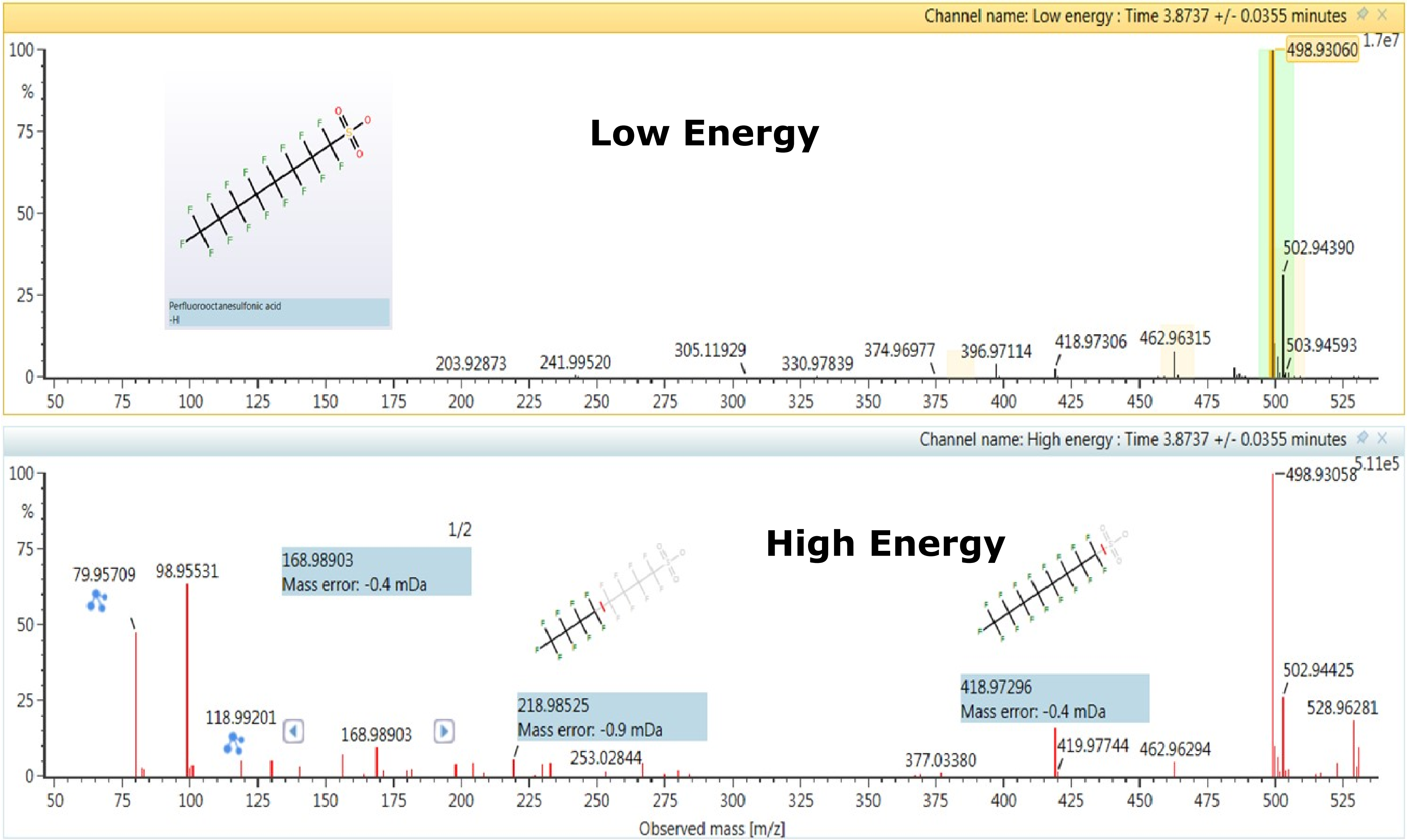

APGC+: Lake Water (Non-Target)

- Componentized MSE Spectra (FTOH)

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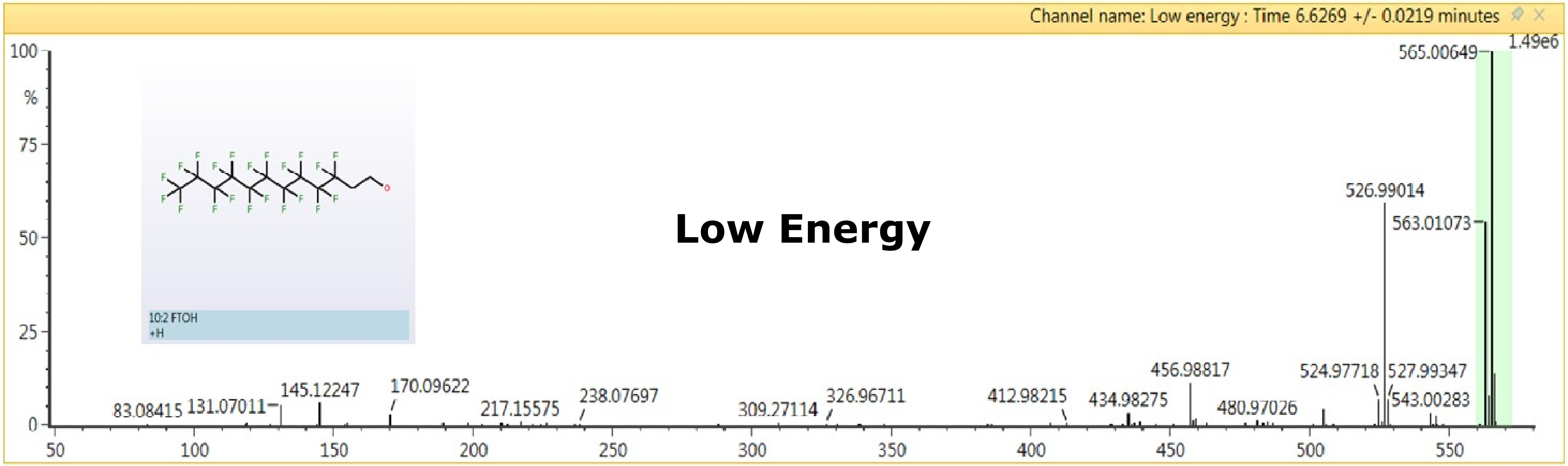

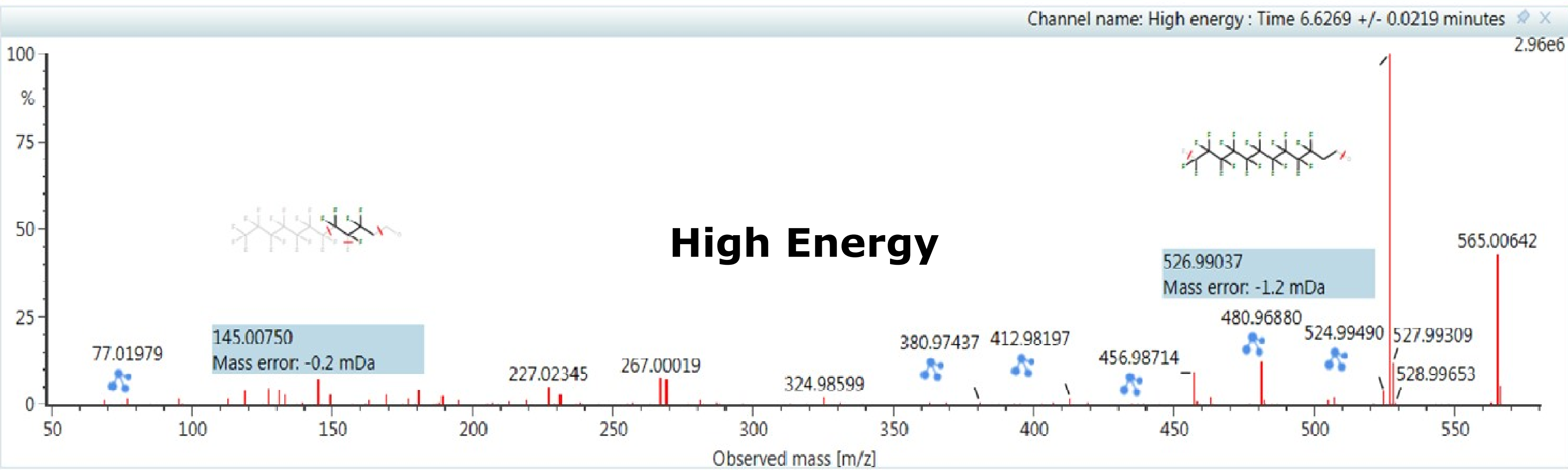

LC-ESI-: Lake Water (Non-Target)

- Screening Identification with CCS

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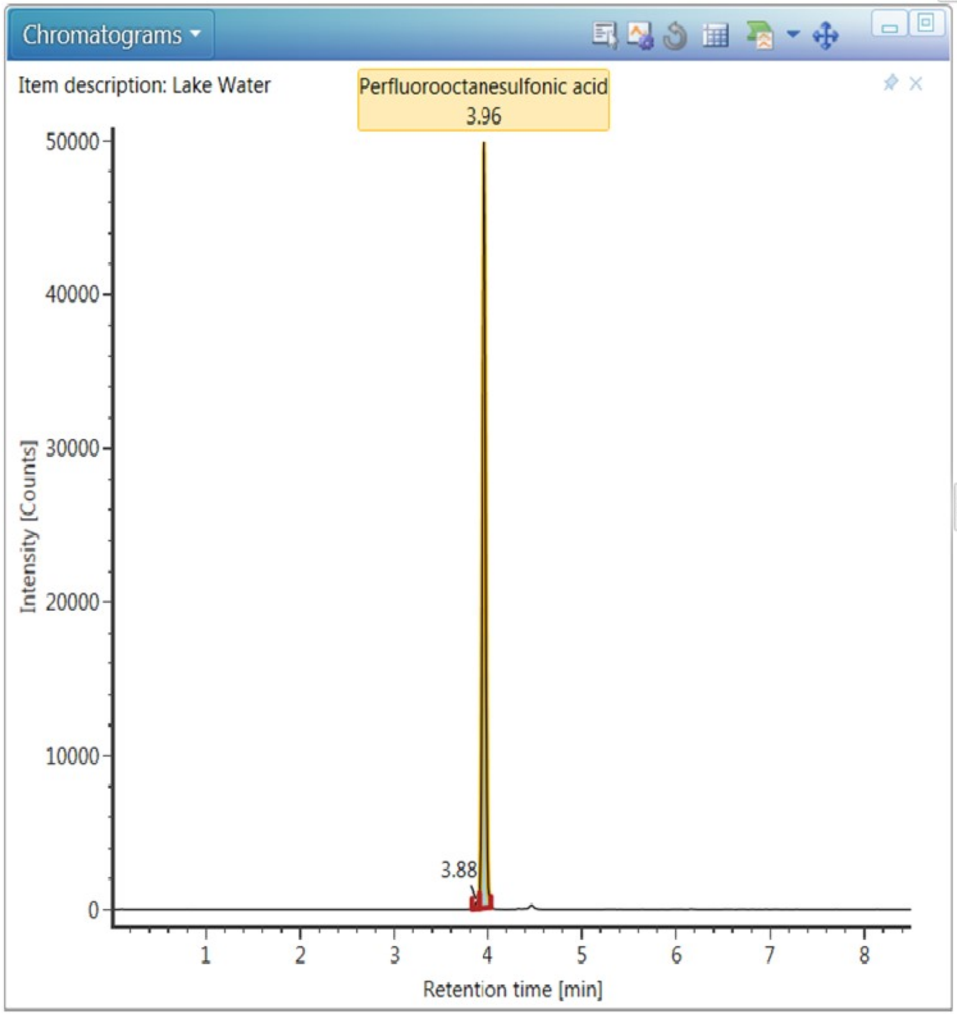
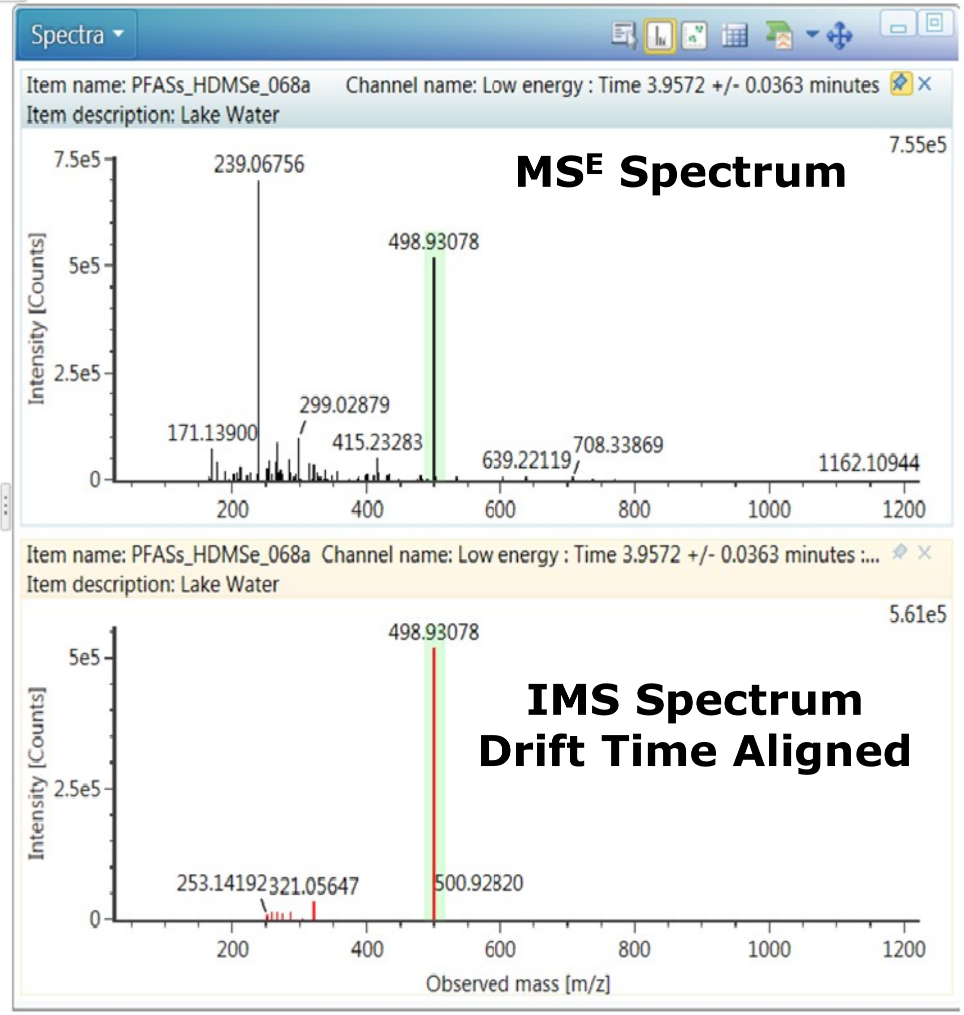
Component name	Mass error (ppm)	Expected RT (min)	Observed CCS (Å²)	Expected CCS (Å²)	Collision cross section error (%)
Perfluorooctanesulfonic acid	1.1	4.20	166.96	166.93	0.02





What is Collision Cross Section (CCS)?

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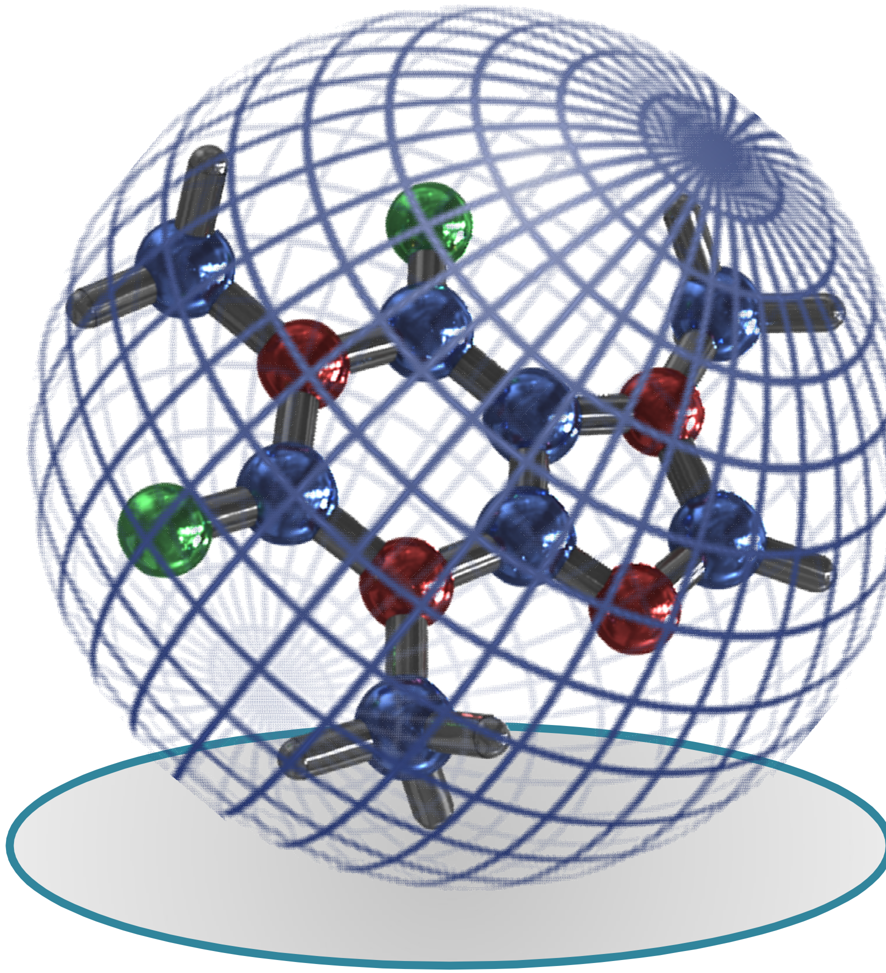
- CCS is an important distinguishing characteristic of an ion which is related to:
  - chemical structure
  - 3-dimensional conformation

- CCS is a physicochemical property of an ion.



APGC+: Drinking Water (PFOS x10 less)
- CCS Values Conserved at low conc.

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Component name	Mass error (ppm)	Expected RT (min)	Observed CCS (Å²)	Expected CCS (Å²)	Collision cross section error (%)
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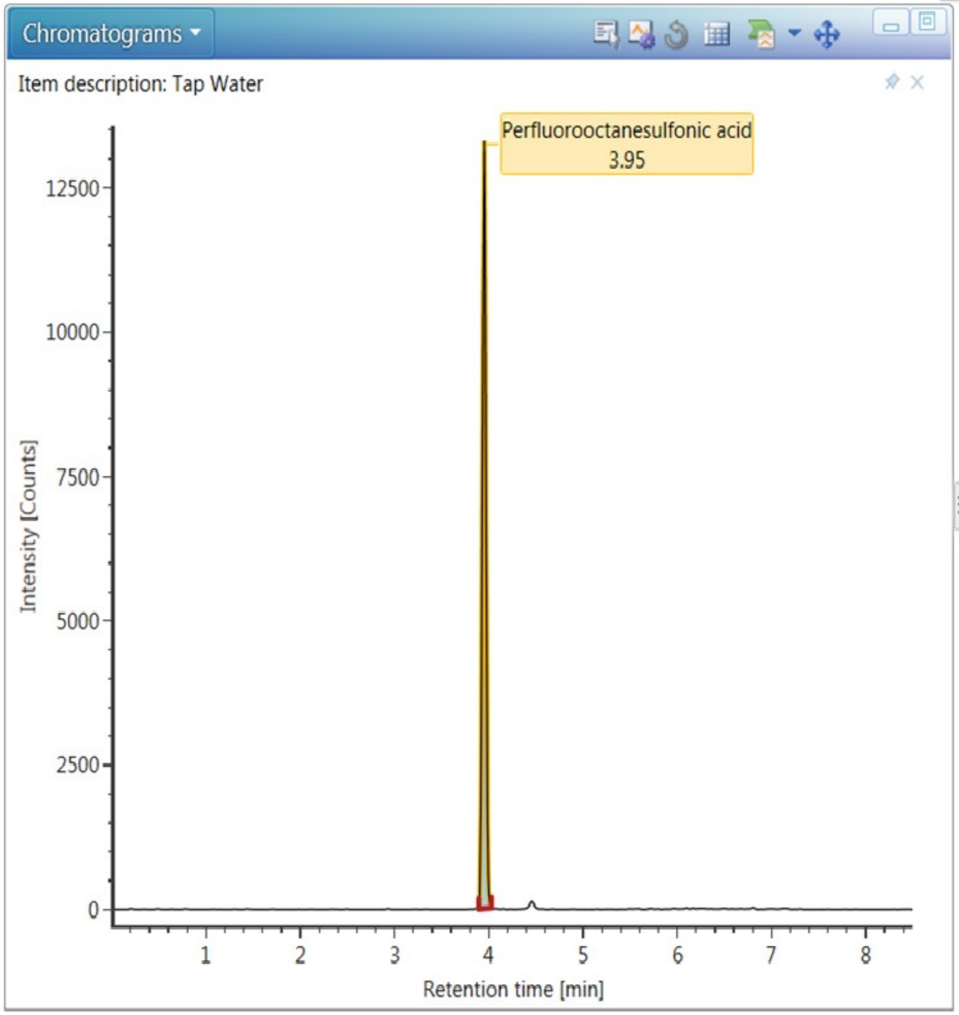

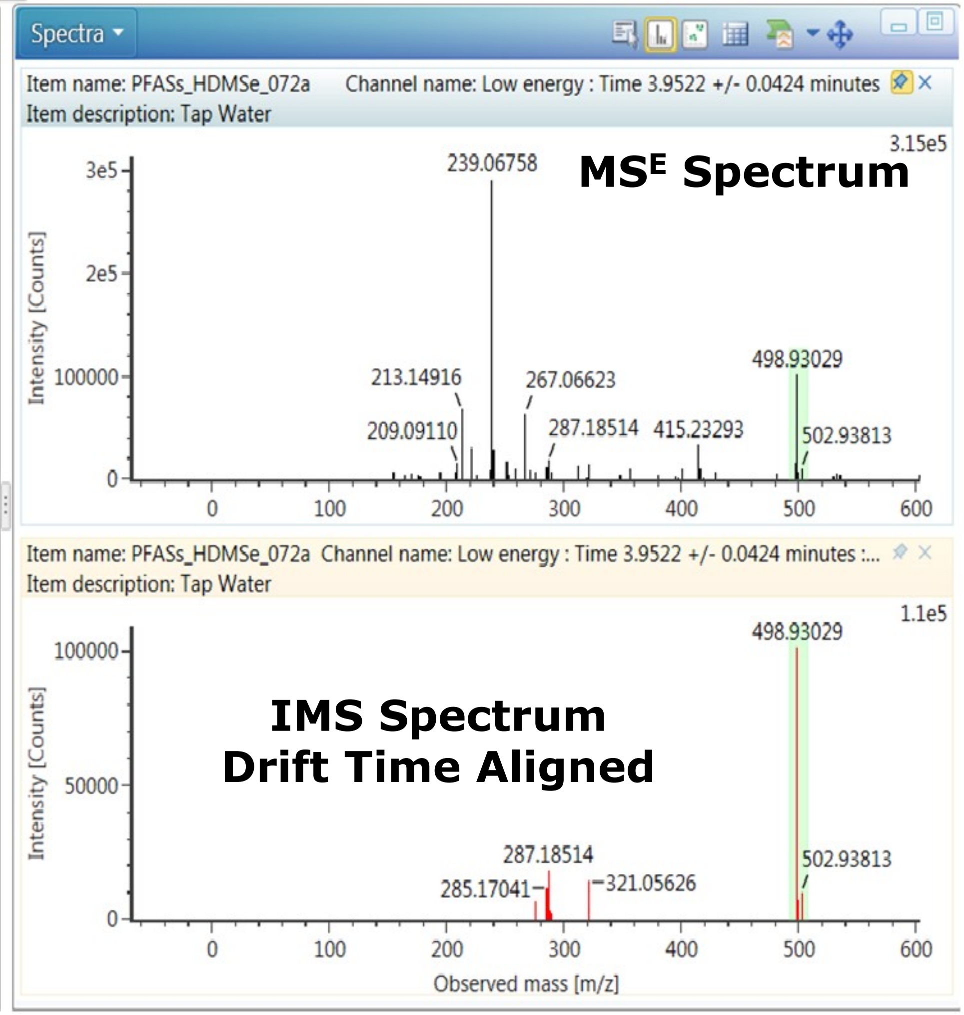

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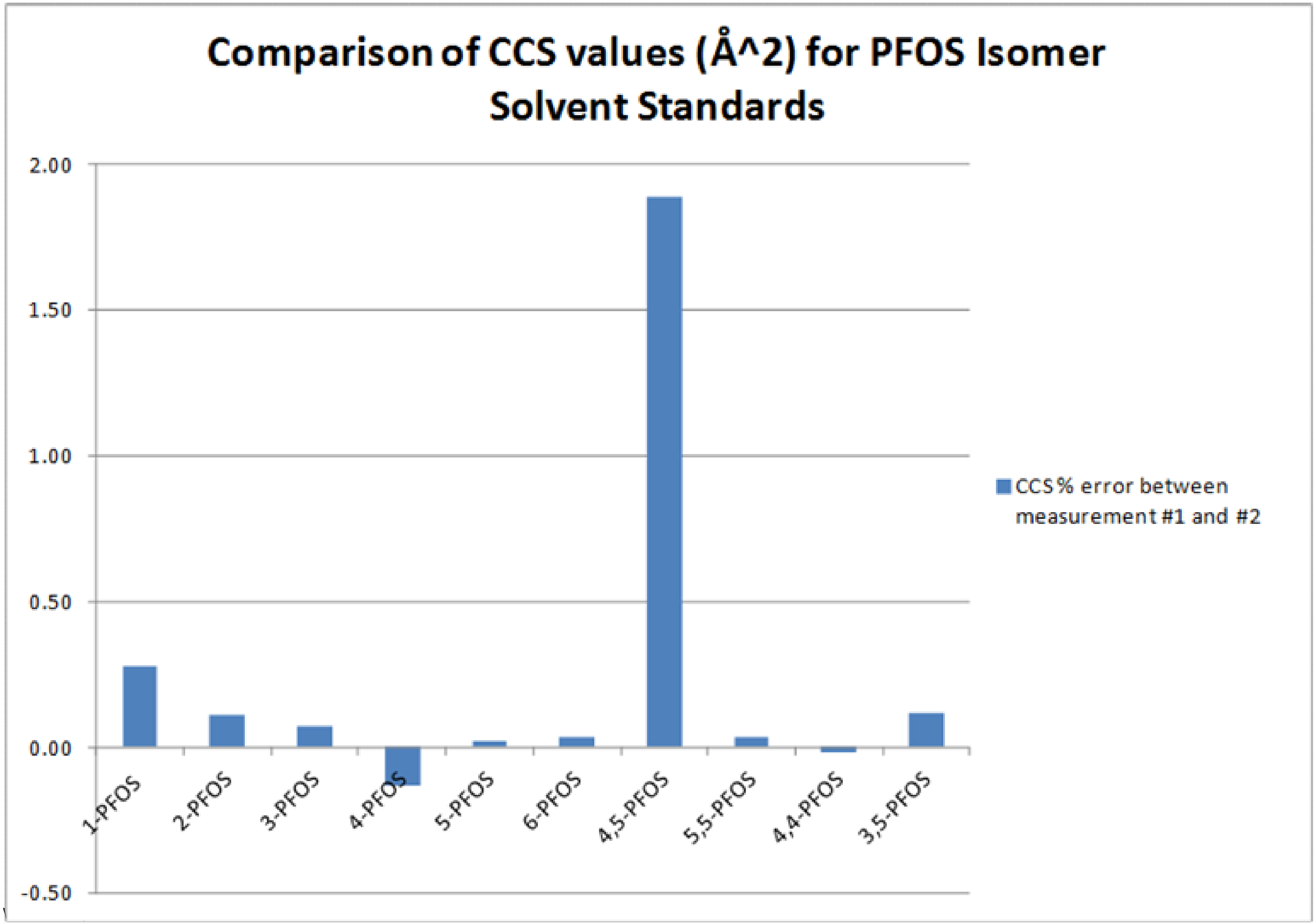

Summary

- Universal ionization coverage affords comprehensive coverage of analyte classes which vary in physical and chemical properties and require GC and LC analyses

- Accurate mass low and elevated collision energy spectrum provides fragmentation pathway information which can be used for further screening analyses and isotope fidelity for both low and high energy

- Including ion mobility, and additional dimension of separation, in the screening workflow adds an additional identification parameter, CCS, which is conserved despite ionization mechanism, concentration, and retention time.

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Acknowledgements

- Ingrid Ericson Jogsten – MTM Research Center

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MAN TECHNOLOGY
ENVIRONMENT
RESEARCH CENTRE

- Laruen Mullin, Gareth Cleland, Adam Ladak – Waters Corp.

Thank You