An online-SPE approach to analyzing broad classes of Emergent Contaminants

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Outline

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- Previous methodology and What's new
- What is online-SPE
- Optimizing online-SPE
- Applications – PPCPs and Perfluorinated Compounds
- Improving methods for matrix interference
  - Optimizing Mass Spectrometer parameters
- Closing Comments

Previous Methodology

for PPCP/CEC analysis

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- EPA 1694 (Dec. 2007)
- Four analyte classes –
  - ESI(+) Formate buffer, MeOH:ACN
  - ESI(+) Oxalic acid, MeOH:ACN
  - ESI(-) Acetate buffer, MeOH:ACN
  - ESI(+) HILIC, Basic extraction

Newer Developments

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- Eluent systems that improve ESI.
- More variety of SPE sorbents (polymeric, WAX)
- Online-SPE, decreasing analysis time
- Analytes of interest has changed
- Sample collection/preservation (Vanderford[1])
- EPA method 543 (Online SPE specific)
- Improvements to Instrumentation
  - Polarity switching (ESI+/-)
  - High duty cycles with smaller FWHM

What is Online-SPE

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- Automated solid phase extraction at the instrument
- Uses 2 HPLC pumps and 2 injection valves; sample load and LC run; can be multiplexed.
- Specialized LC column: 20-50mm, 12-40um, 2.1mm
- Loading a 2-10mL aqueous sample at 1-5mL/min
- Elution by backflushing the SPE sorbent with mobile phase
- *Turbulent flow: Specialized online SPE (Thermo Transcend Turboflow)

Equan MAX System

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Optimizing online SPE

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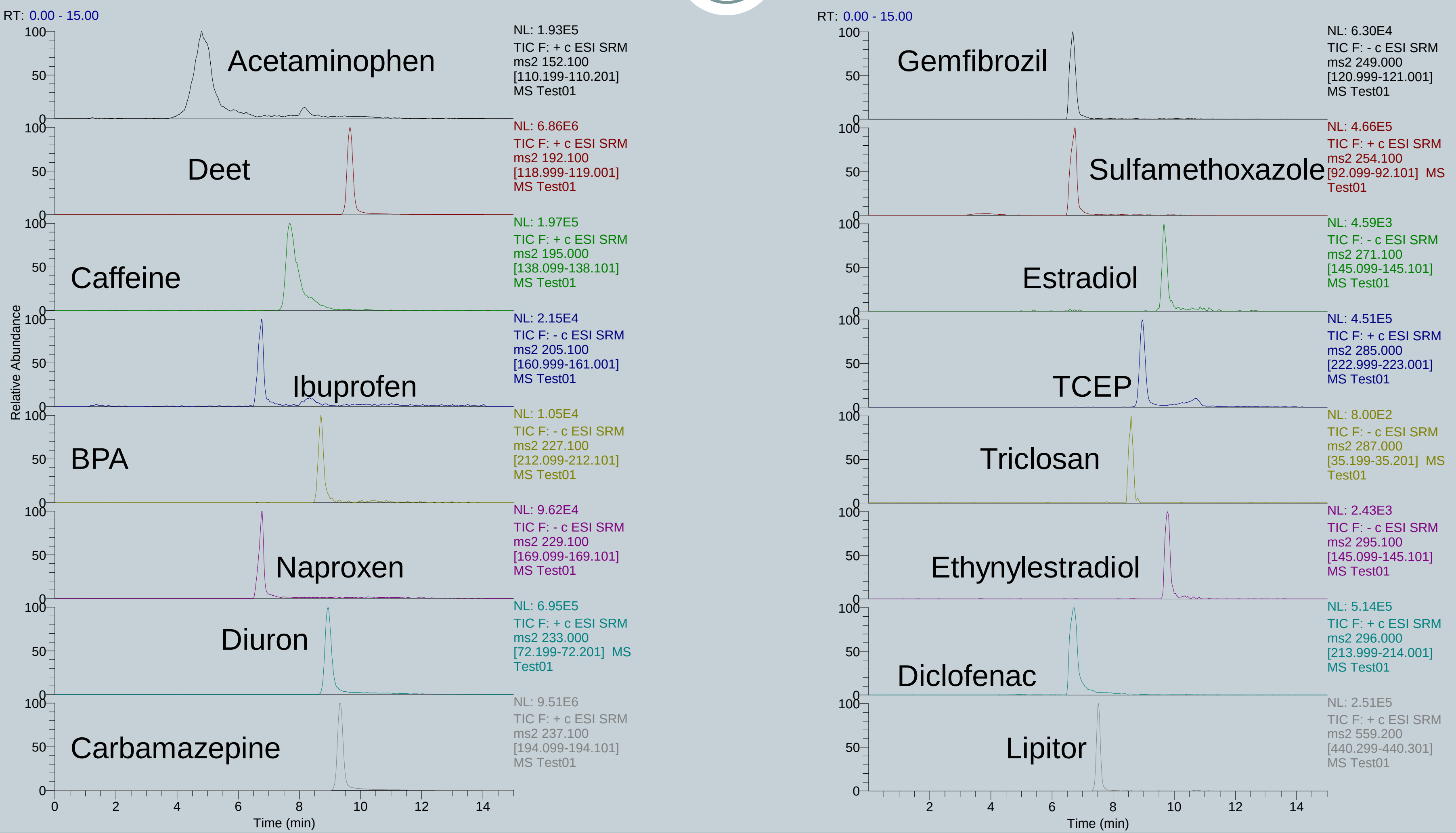
- Wash step – Allows the system to rinse the media to eliminate preservative and/or matrix salts. 2-3 sample volumes.
- Elute focus – Step gradient organic to elute highly hydrophobic analytes from C18.
- Mobile phase background elimination – Works well for PFC analysis. Mobile phase 90% organic while online SPE sample loading.
- Use short analytical columns (50mm) unless increased retention is required.
- Use fast gradients – online SPE adds retention

Compound Classes using online SPE

- Pharmaceuticals (Acetaminophen)
- Hormones (17B-Estradiol)
- Organophosphate Flame Retardants (TCEP)
- Perfluorinated Compounds (PFOA)
- Artificial Sweeteners (Sucralose)
- Iodinated Contrast Media (Iopromide)
- Antimicrobials (Triclosan)
- Pesticides (Pyrethroids down to 1.0ng/L)
- Others (nonylphenols, Bisphenol-A, Phthalates, NP Pesticides)

LIMITED by ESI more than SPE

Example Chromatogram at 50ng/L



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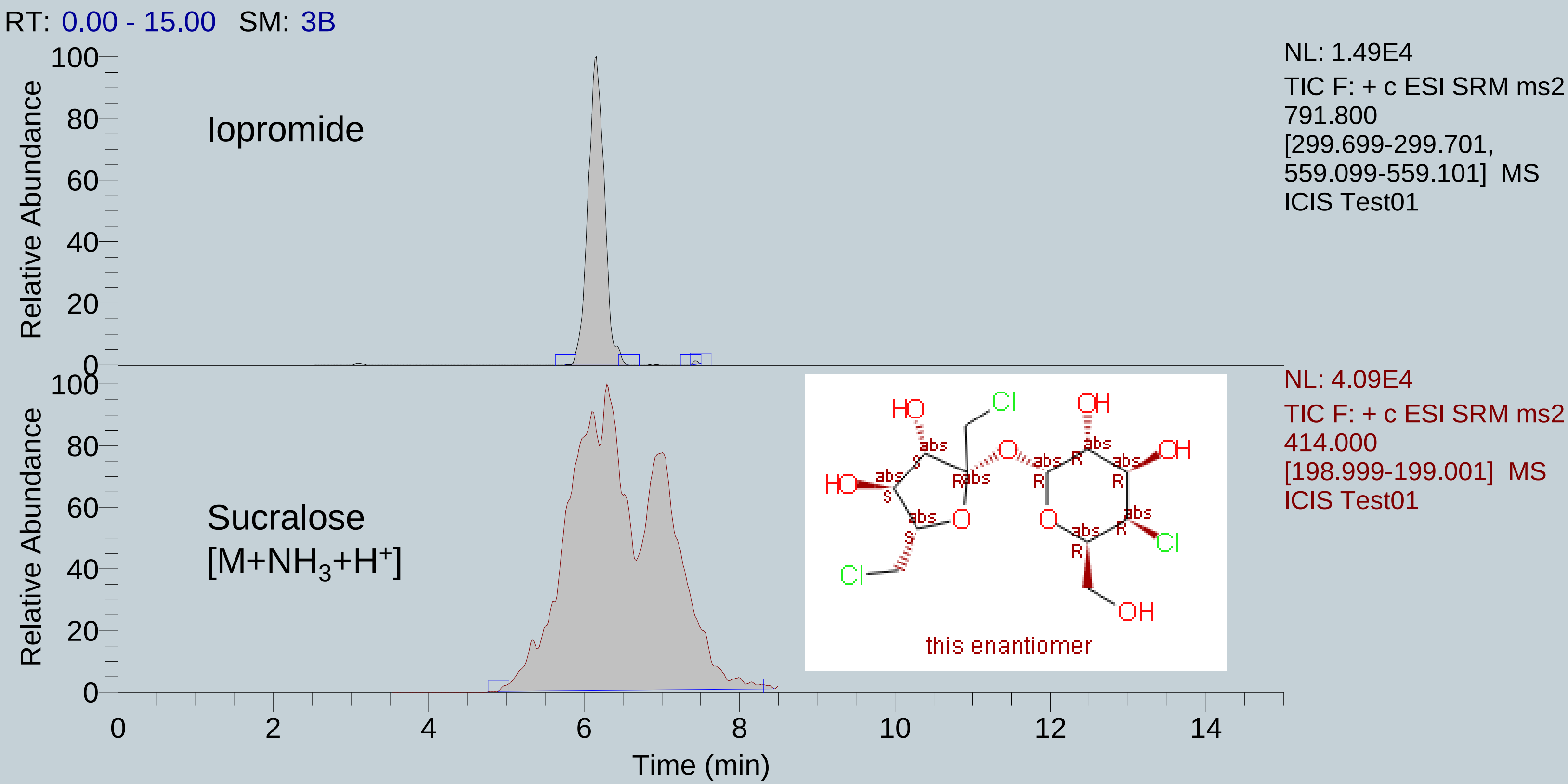
Example Chromatogram at 50ng/L

Polymeric (Strata-X) online SPE retains highly polar analytes



PPCP analysis conditions

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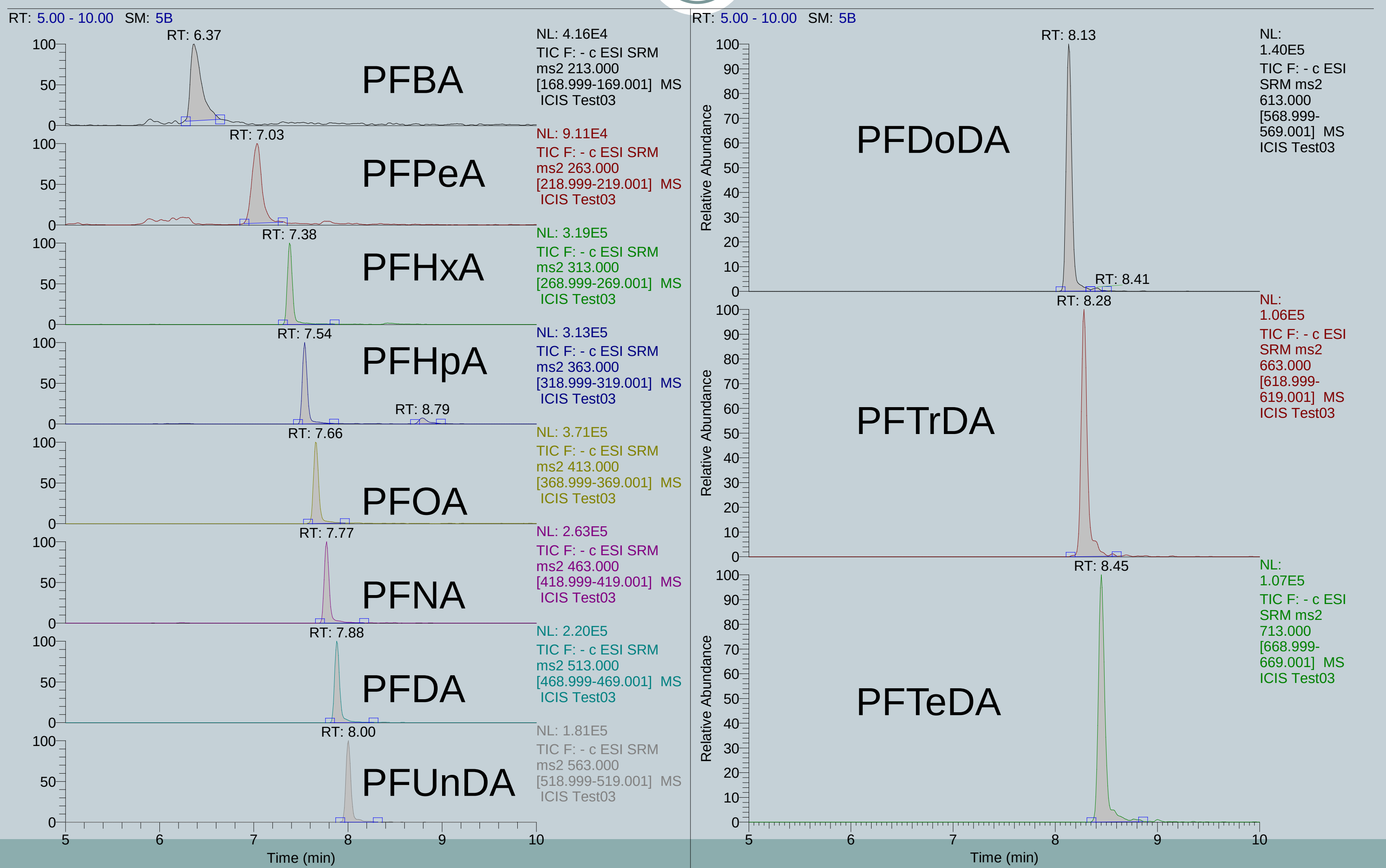
- Sample: Prepreserved with sodium azide and ascorbic acid. Add 3 drops formic acid per 40mL prior to analysis
- Mobile Phase: 300ul/min 5mM Ammonium Hydroxide with 10-80% Methanol.
- Online SPE media: Thermo Hypersil C18 and Strata-X (polymeric) for Sucralose/Iopromide (can use HyperSep Retain PEP[7])
- MS parameters: TSQ Quantum Ultra. Vap/Cap Temps at 250C, Sheath=35, Aux=15, CID=1.0mTorr Argon, Q1=0.4 FWHM
- Quantitation method is strict isotope dilution

Example: 50ng/L C4-C14 PFCAs



Example: 50ng/L C4-C10 PFSAs

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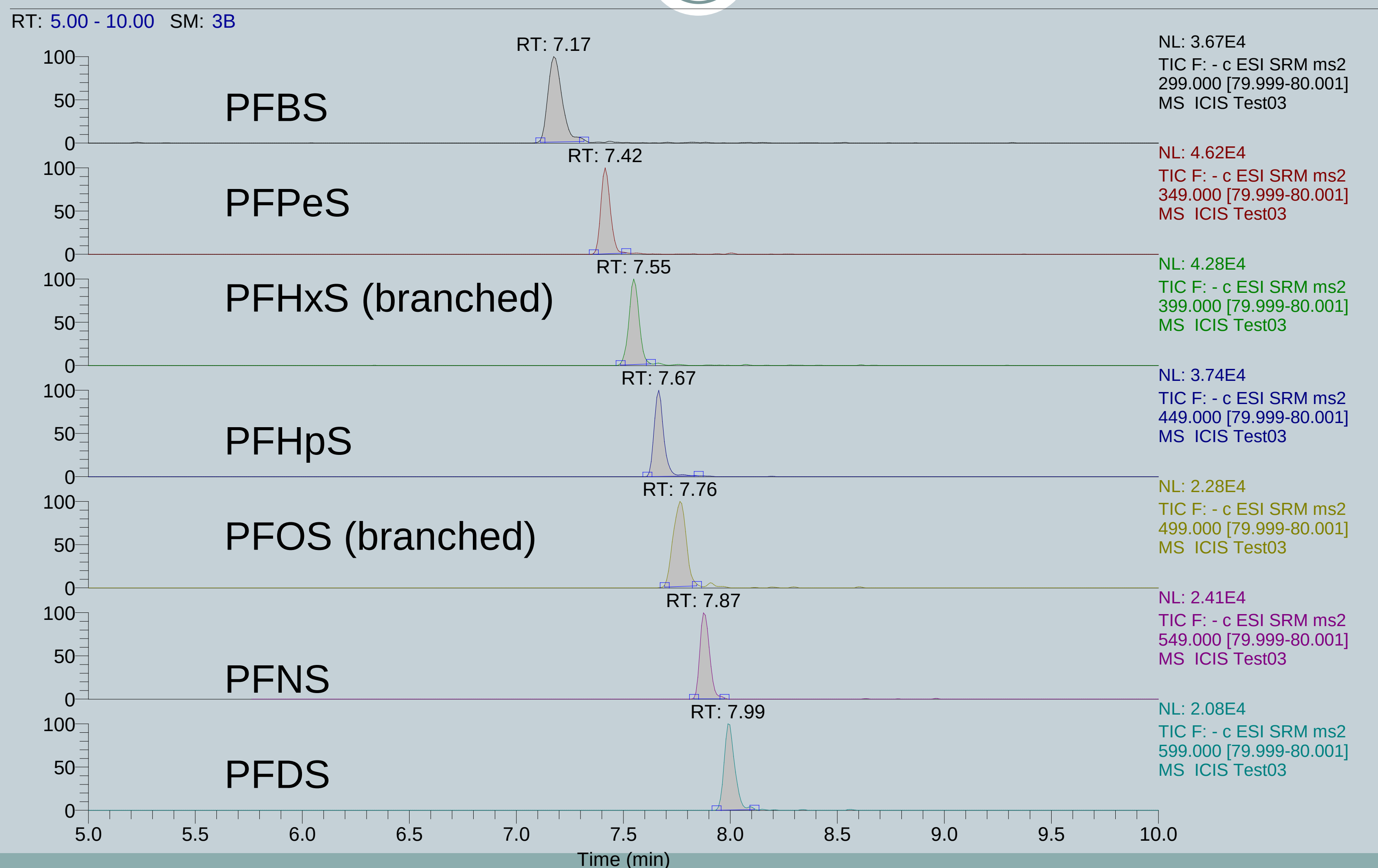
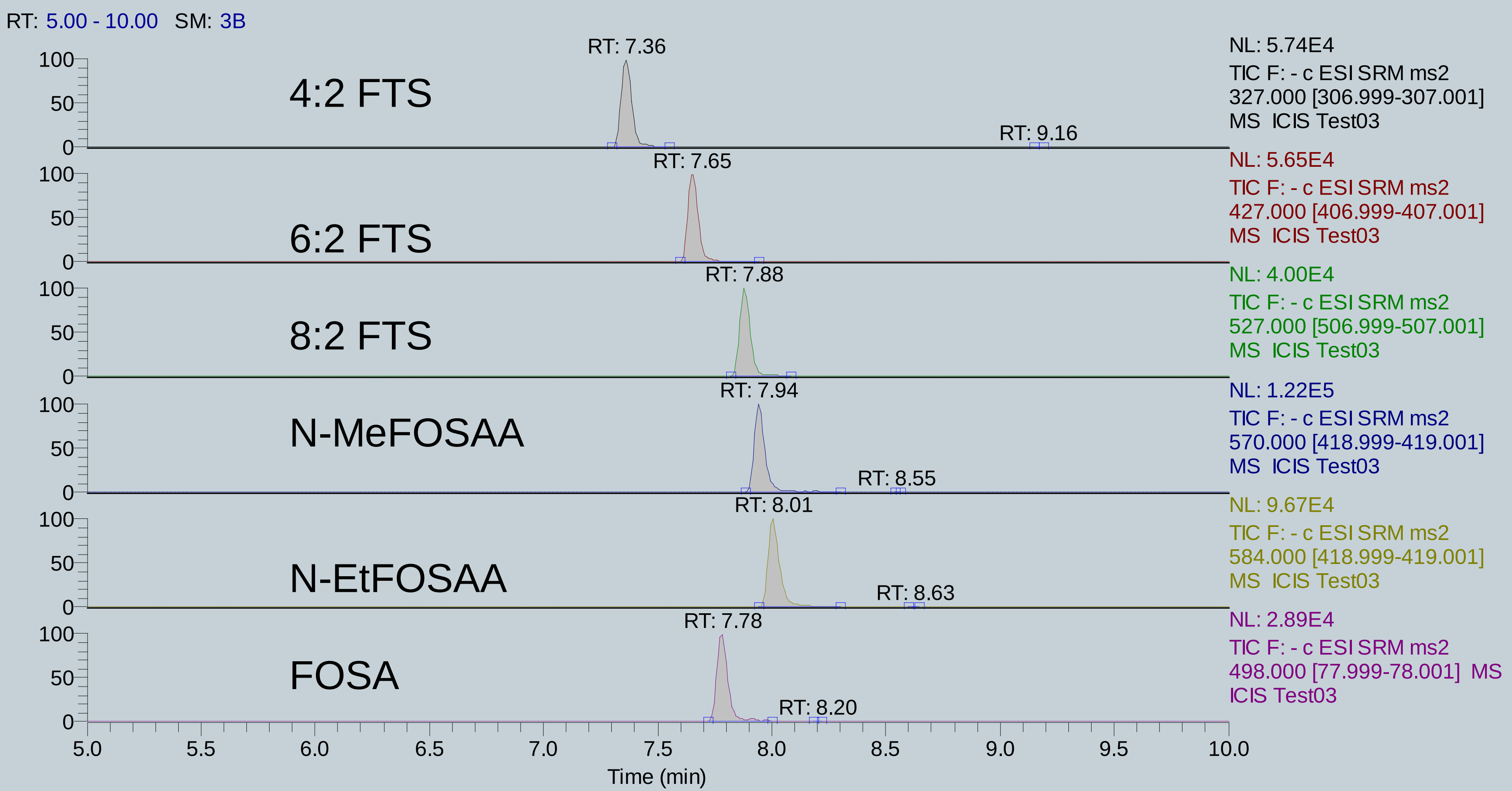


Example: 50ng/L other PFCs



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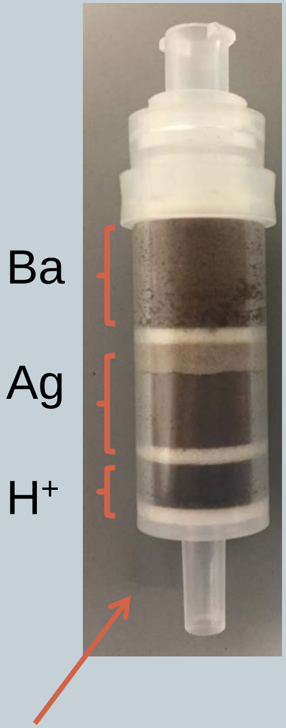
PFC analysis conditions

- Sample: Prepreserved with 5g/L Trizma
- Mobile Phase: 300ul/min 0.24-0.08% Ammonium Hydroxide with 40-80% Methanol.
- Online SPE media: Strata-X-AW (mixed mode weak anion) works especially well for PFBA/PFPeA
- LC Column: C18 EVO (stable at high pH)
- MS parameters: TSQ Quantum Ultra. Vap/Cap Temps at 250/150C, Sheath=40, Aux=40, CID=1.0mTorr Argon, Q1=0.4 FWHM
- Quantitation method is internal standard by compound class

Addressing Matrix issues

- For samples containing high salts Cl^- or SO_4^{2-}, use Dionex OnGuard II Ba/Ag/H. Check for losses.
- Have a wash step after sample loading (1-2 sample volumes)
- Filter samples with appropriate media. Glass fiber works well for PFCs, even for PFTeDA
- Use smaller FWHM; 0.2 or 0.4 instead of 0.7Da



After filtering >2000ppm Cl^-

Effect of FWHM

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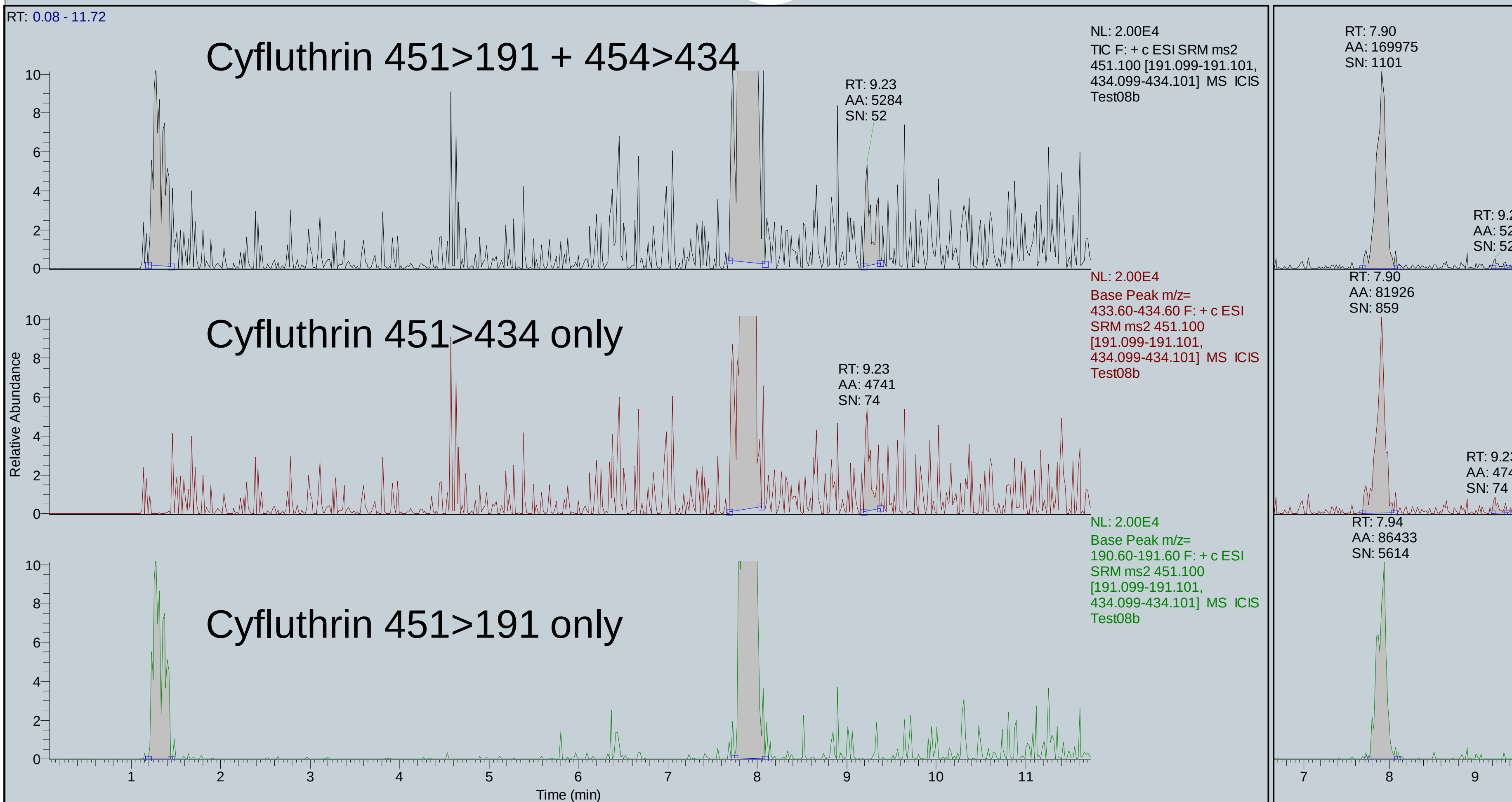
Q1 FWHM = 0.7Da

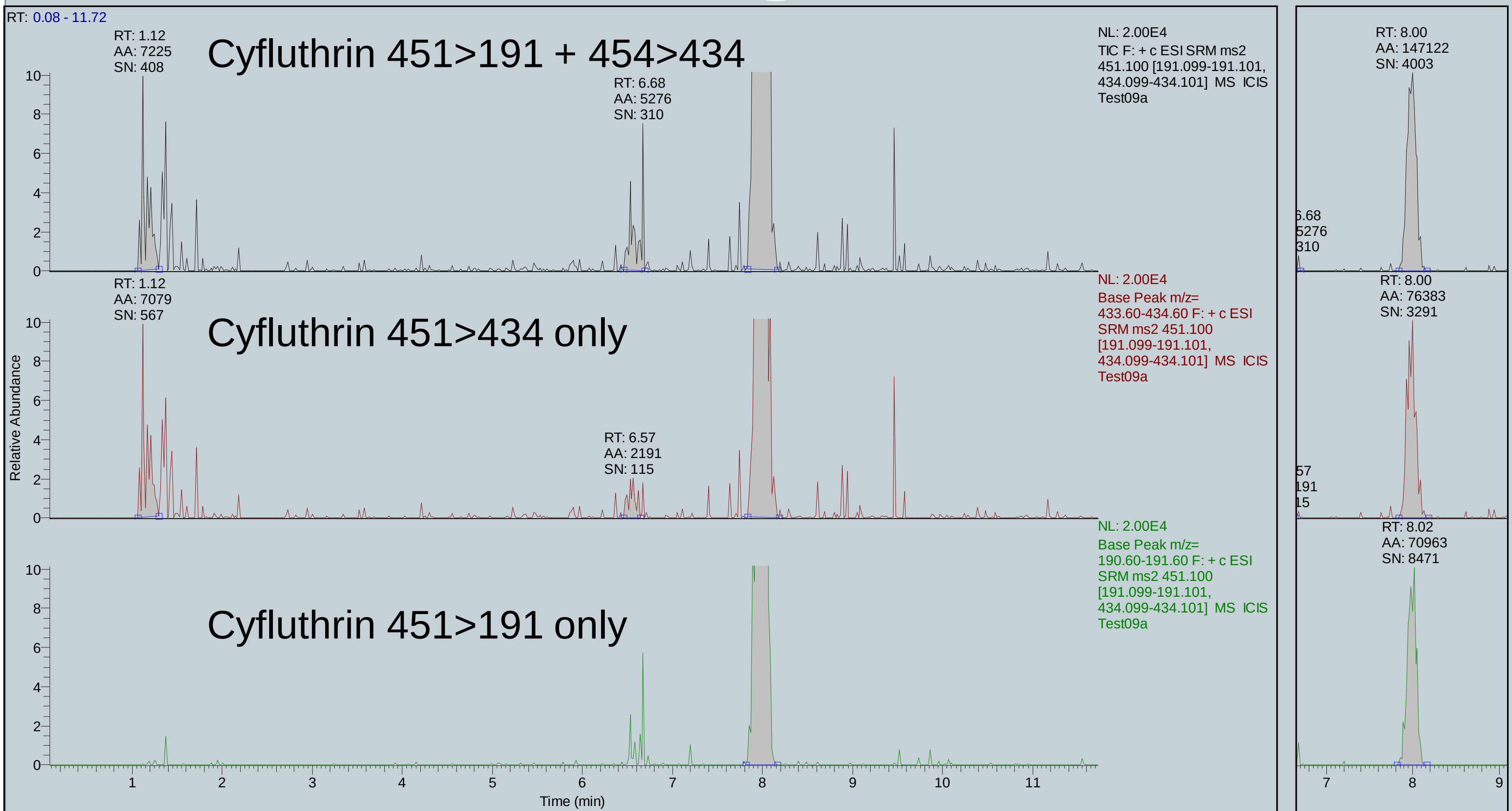


Effect of FWHM

Q1 FWHM = 0.5Da

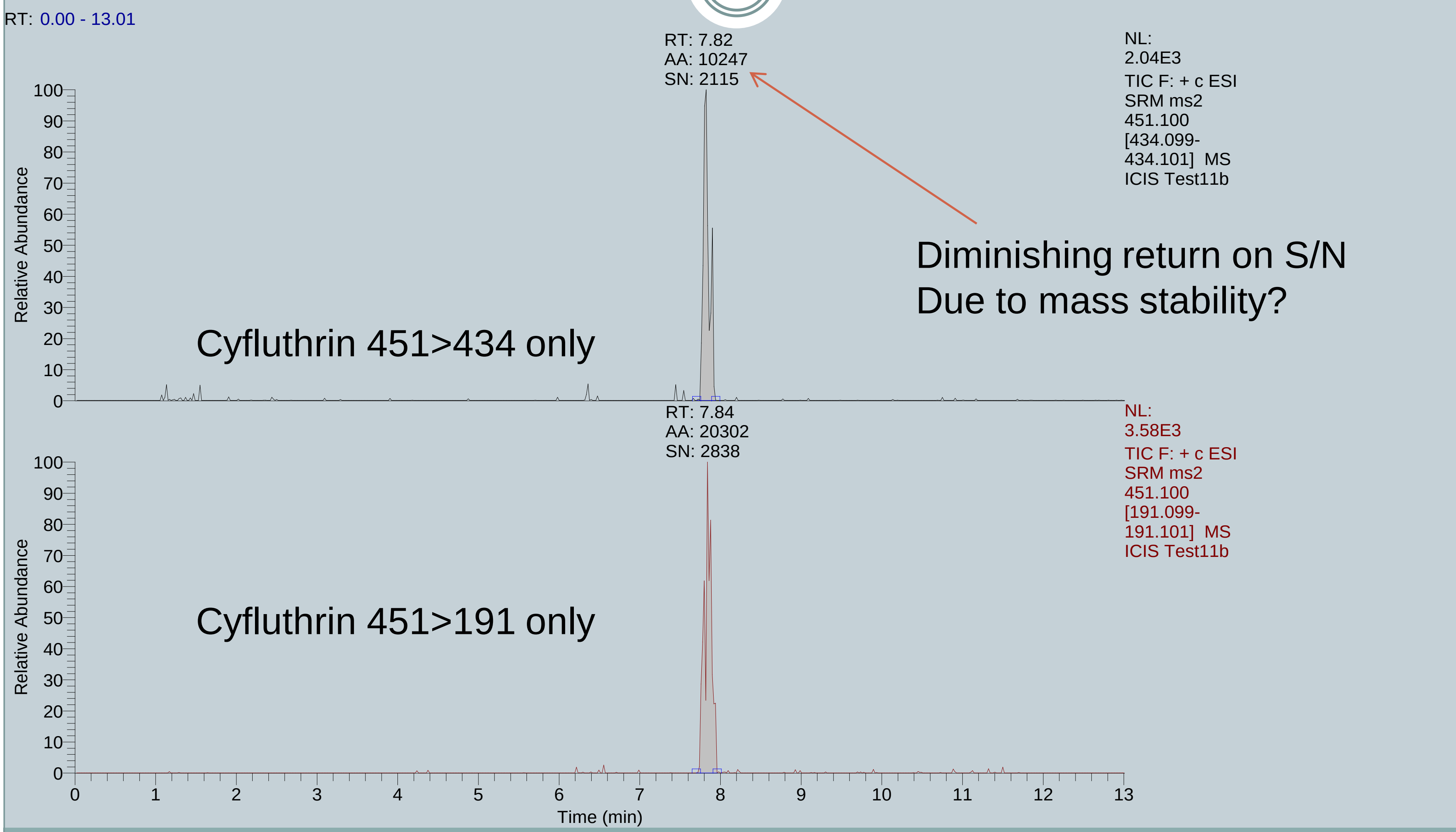
Effect of FWHM

Q1 FWHM = 0.2Da

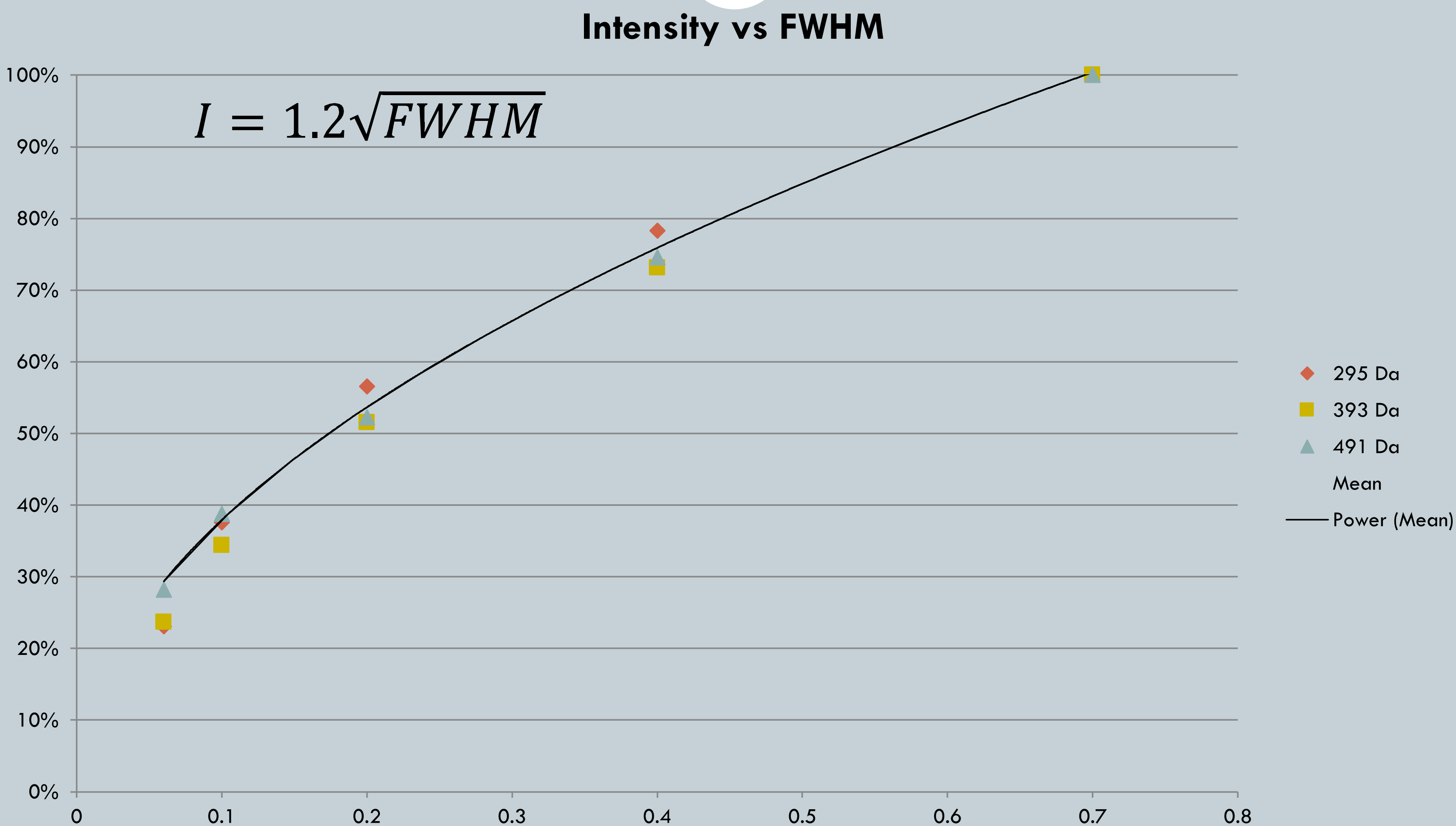


Effect of FWHM

Q1 FWHM = 0.04Da



Sensitivity vs. FWHM



$$I = 1.2\sqrt{FWHM}$$

Mass Calibration

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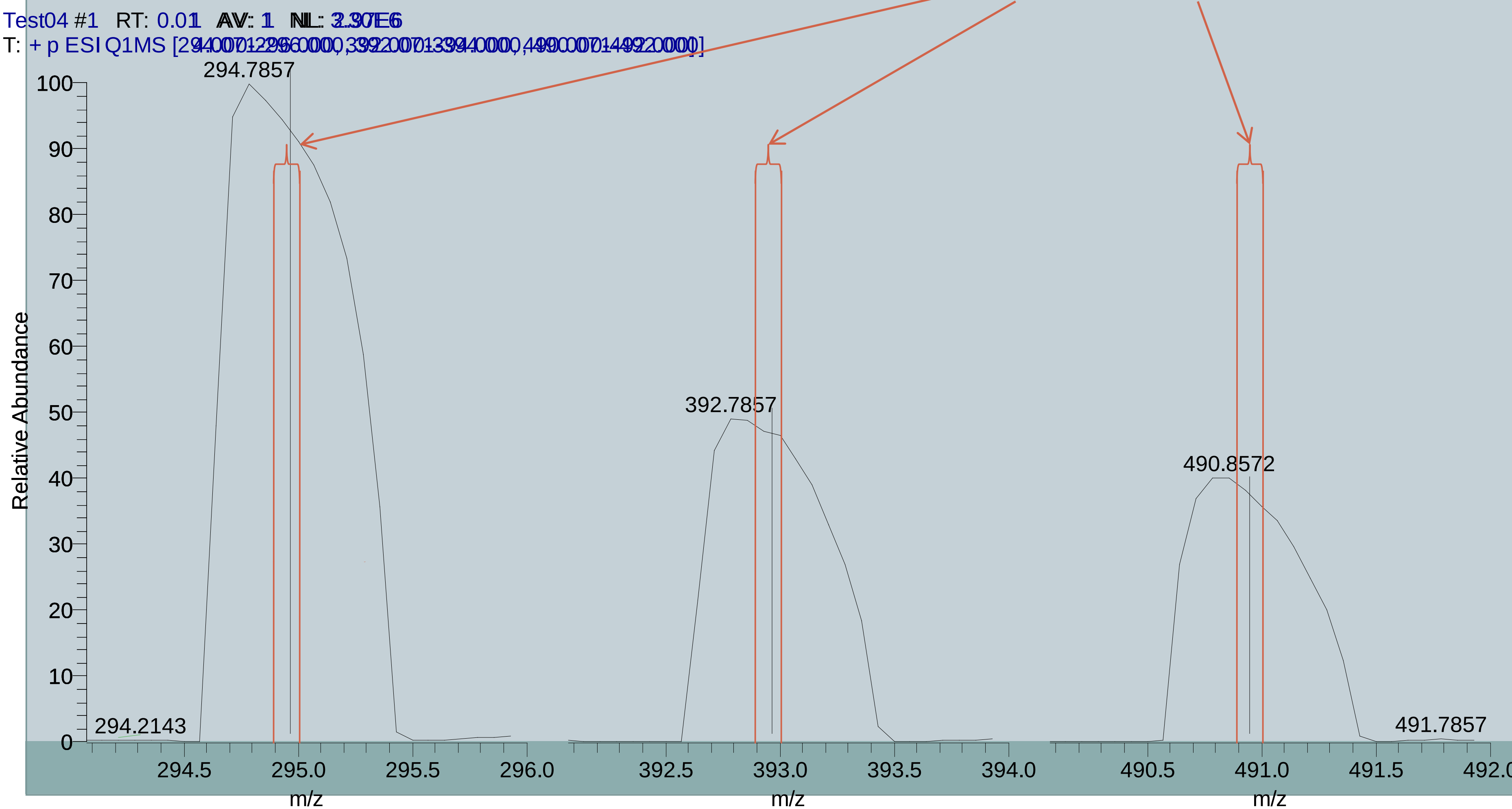
- Lowering FWHM requires special attention to mass calibration frequency.
- Mass stability for TSQ Series around ±0.05 Da ✓
- There is a limit to lowering FWHM (0.2-0.3 Da)
- Using Phosphoric acid clusters for mass calibration can improve mass calibration stability (9 mass cal points instead of 3.
- Instrument Mass stability is the limiting factor to lowering FWHM.

Mass Stability and FWHM

FWHM = 0.70 Da

Mass when analyte was infused ± 0.05Da



Mass Stability and FWHM

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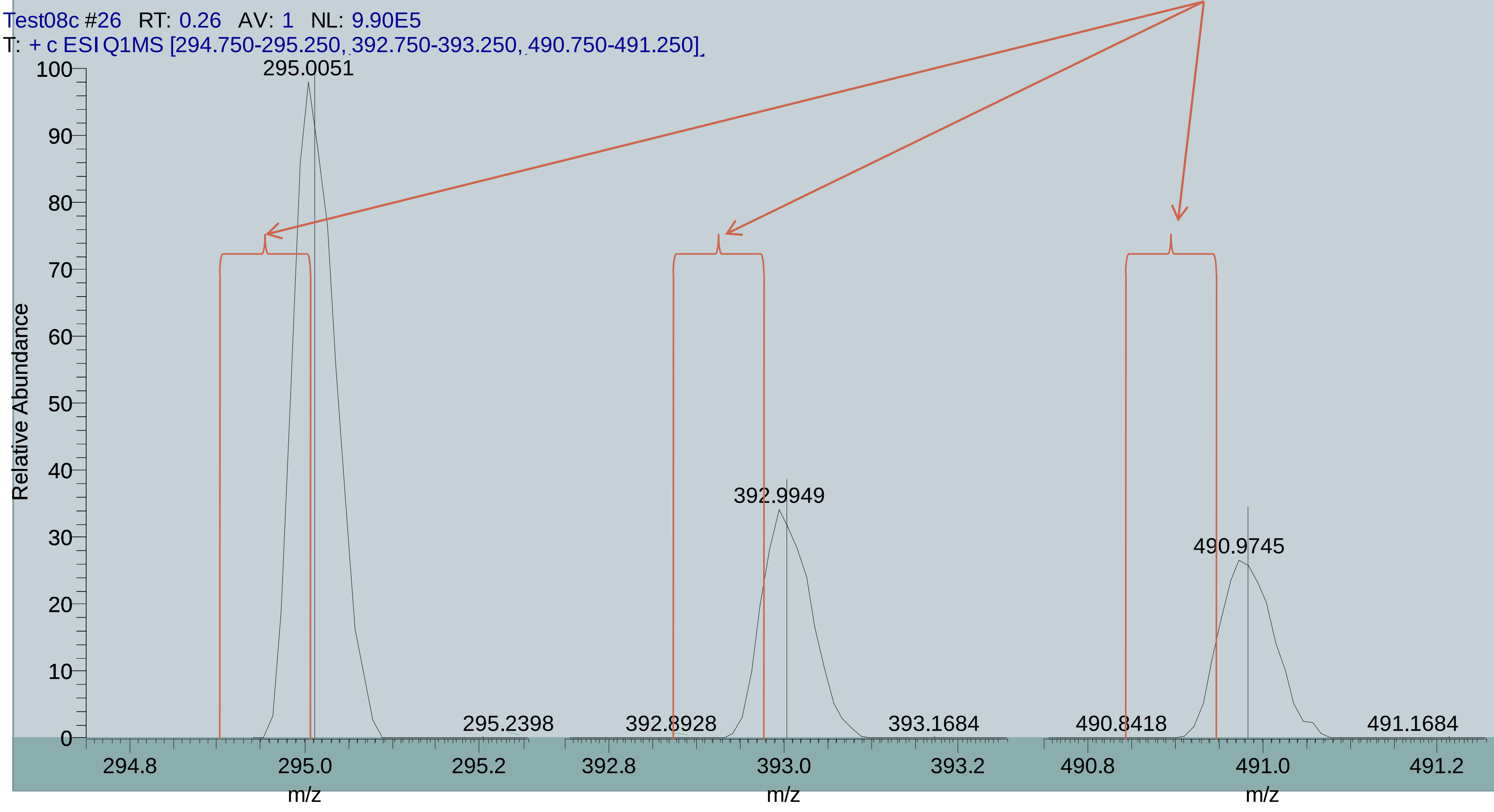
FWHM = 0.06 Da

Mass when analyte was infused ± 0.05Da



Mass Error vs FWHM

Before/After Mass Calibration

Data Applicability

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- Do results from online-SPE produce results that reflect bioavailable[3] analyte only?
  - Particles trapped in media
  - Particles pass through (turbulent flow at high flows)
- Analytes with high K_{oc} might be trapped in suspended solids
- Examples – Pyrethroids and some PFCs. Generally analytes that are VERY hydrophobic

Final Thoughts

- Most analytes can be done by online-SPE which makes analysis much faster
- There may be limitations on sensitivity and peak shape.
- Coupling online-SPE with MS/MS is a sensitive analytical tool.
- Online SPE may be adapted to soil analysis if aqueous extraction is used

References

1) Vanderford, Brett J., Douglas B. Mawhinney, Rebecca A. Trenholm, Janie C. Zeigler-Holady, and Shane A. Snyder. “Assessment of Sample Preservation Techniques for Pharmaceuticals, Personal Care Products, and Steroids in Surface and Drinking Water.” Analytical and Bioanalytical Chemistry 399, no. 6 (February 2011): 2227–34. doi:10.1007/s00216-010-4608-5.
2) Peake, Barrie M., Rhiannon Braund, Alfred Y.C. Tong, and Louis A. Tremblay. “Detection and Presence of Pharmaceuticals in the Environment.” In The Life-Cycle of Pharmaceuticals in the Environment, 77–107. Elsevier, 2016. doi:10.1016/B978-1-907568-25-1.00004-9.
3) Maund, Steve J., Mick J. Hamer, Mike C. G. Lane, Eamonn Farrelly, Jean H. Rapley, Una M. Goggin, and Wendy E. Gentle. “Partitioning, Bioavailability, and Toxicity of the Pyrethroid Insecticide Cypermethrin in Sediments.” Environmental Toxicology and Chemistry / SETAC 21, no. 1 (January 2002): 9–15.
4) EPA Method 537 v1.1, Determination Of Selected Perfluorinated Alkyl Acids In Drinking Water By Solid Phase Extraction And Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS), September 2009
5) EPA Method 543 v1.0, Determination of Selected Organic Chemicals in Drinking Water by On-line Solid Phase Extraction - Liquid Chromatography/Tandem Mass Spectrometry (On-line SPE - LC/MS/MS), March 2015
6) Yamazaki, Eriko, SachiTaniyasu, KodaiShimamura, Shunya Sasaki, and Nobuyoshi Yamashita. “Development of a Solid-Phase Extraction Method for the Trace Analysis of Perfluoroalkyl Substances in Open-Ocean Water.” Bunseki Kagaku 64, no. 10 (2015): 759–68. doi:10.2116/bunsekikagaku.64.759
7) Batchu, Sudha Rani, Natalia Quinete, Venkata R. Panditi, and Piero R. Gardinali. “Online Solid Phase Extraction Liquid Chromatography Tandem Mass Spectrometry (SPE-LC-MS/MS) Method for the Determination of Sucralose in Reclaimed and Drinking Waters and Its Photo Degradation in Natural Waters from South Florida” 7 (2013): 141.

QUESTIONS ?
COMMENTS?

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