Journal of Visualized Experiments

Identifying Per- and Polyfluorinated Chemical Species with a Combined Targeted and Non-Targeted-Screening High-Resolution Mass Spectrometry Workflow

--Manuscript Draft--

Article Type:	Invited Methods Article - JoVE Produced Video
Manuscript Number:	JoVE59142R1
Full Title:	Identifying Per- and Polyfluorinated Chemical Species with a Combined Targeted and Non-Targeted-Screening High-Resolution Mass Spectrometry Workflow
Keywords:	PFASs; Mass Spectrometry; Perfluorinated Substances; GenX; HFPO-DA; Non-targeted screening; water pollution; LC-MS/MS quantitation
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Additional Information:	
Question	**Response**
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Please indicate the **city, state/province, and country** where this article will be **filmed**. Please do not use abbreviations.	Durham, North Carolina, United States

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Dear Editor,

Please find enclosed our manuscript entitled "*Identification of per- and polyfluorinated chemical species with a combined targeted and non-targeted screening high-resolution mass spectrometry workflow"* that we would like to be considered for publication in Journal of Visualized Experiments. This manuscript presents a sample preparation/concentration strategy using solid-phase extraction (SPE) tuned for short chain and hydrophilic PFASs, such as perfluorinated ethers acids, and describes the analysis of samples prepared this way in both targeted and non-targeted modes. We consider of value publishing these data in Journal of Visualized Experiments, as they provide for the quantitation of known materials with substantial public interest, while non-targeted analysis allows identification of emerging unknowns and determination of their chemical class and structure. The techniques presented in this paper and demonstrated in video format will be highly useful for researchers interested in perfluorinated chemistry for emerging compounds screening, and for those interested in expanding their targeted chemistry workflows to include investigation of novel chemical compounds.

The procedures and experiments described in this manuscript were developed and performed by James McCord and Mark Strynar, with James McCord preparing the manuscript.

Benjamin Werth invited the initial submission of this manuscript and has provided his support and assistance throughout its preparation.

Thank you for your consideration of this manuscript. We look forward to hearing from you.

Sincerely yours,

Mark Strynar

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TITLE:

Identifying Per- and Polyfluorinated Chemical Species with a Combined Targeted and Non-Targeted-Screening High-Resolution Mass Spectrometry Workflow

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KEYWORDS:

PFAS, Perfluorinated Compounds, Solid Phase Extraction, Environmental Analysis, Water Analysis, High Resolution Mass Spectrometry, Non-Targeted Analysis, LC-MS/MS

SUMMARY:

Here, we present a protocol for the sequential targeted quantification and non-targeted analysis of fluorinated compounds in water by mass spectrometry. This methodology provides quantitative levels of known fluorochemical compounds and identifies unknown chemicals in related samples with semi-quantitative estimates of their abundance.

ABSTRACT:

Historical and emerging per- and polyfluoroalkyl substances (PFASs) have garnered significant interest from the public and government agencies from the local to federal levels. The continuing evolution of PFAS chemistries presents a challenge to the environmental monitoring, where ongoing development of targeted methods necessarily lags the discovery of new chemical compounds. There is a need, therefore, to have forward-looking methodologies that can detect emerging and unexpected compounds, monitor these species over time, and resolve details of their chemical structure to enable future work in human health. To this end, non-targeted analysis by high-resolution mass spectrometry offers a broad base detection approach that can be combined with almost any sample preparation scheme and provides significant capabilities for compound identification after detection. Herein, we describe a solid-phase extraction (SPE) based sample concentration method tuned for shorter chain and more hydrophilic PFAS chemistries, such as per fluorinated ether acids and sulfonates, and describe analysis of samples prepared in this fashion in both targeted and non-targeted modes. Targeted methods provide superior quantification when reference standards are available but are intrinsically limited to expected compounds when performing analysis. In contrast, a non-targeted approach can

identify the presence of unexpected compounds and provide some information about their chemical structure. Information about chemical features can be used to correlate compounds across sample locations and track abundance and occurrence over time.

INTRODUCTION:

The class of per- and polyfluoroalkyl substances (PFASs) are persistent organic pollutants with significant public health concern. The specific compounds perfluorooctanoic acid (PFOA) and perfluorooctanesulfonate (PFOS) have drinking water health advisory levels set by the EPA[1,2] and their major US production ceased in the 2000s[3,4]. To gain a significant understanding for the properties of PFAS materials in the textile and consumer product manufacturing spheres, hundreds, if not thousands, of alternate PFAS chemistries have been developed to fill product niches, including replacements for the deprecated compounds[5-8]. There is an ongoing need to monitor the environmental levels of straight chain perfluorinated carboxylic acids and sulfonates such PFOS, PFOA, and their related homologous series, but emerging chemical compounds are not covered by established methods such as EPA Method 537[9] and frequently lack analytical standards for traditional targeted analysis. The intention of this protocol is thus two-fold. It provides a pathway for the targeted LC-MS/MS analysis of fluorochemical species in water where analytical standards are available and details the seamless integration of a non-targeted, high-resolution mass spectrometry-based approach for data analysis that enables the detection of unknown or unexpected compounds in the same samples.

Solid-phase extraction (SPE) is an established technique for the sample cleanup and concentration with applications to many analytes and sample matrices[10,11]. For PFAS analysis, multiple solid retentive phases including non-polar, functionalized polar, and ion exchange columns have been used to varying extents for subclasses of fluorinated species in a wide variety of matrices[9,12-16]. Advances in SPE sample analysis using on-line setups greatly increase the throughput of the approach and improve the reproducibility of sample handling, but the fundamental process remains consistent[17]. Some efforts to remove the offline concentration of SPE using large volume injections have also been undertaken, but these require modifications to the chromatography that place them outside the realm of casual analysis[18,19]. Our sample analysis uses a polymeric weak anion exchange (WAX) retentive phase to thoroughly separate acidic PFAS materials from the traditional organic contaminants while achieving substantial sample concentration factors. This WAX phase is important to capture the short chain perfluorinated acids such as perfluorobutane sulfonate (PFBS) or perfluorinated ethers such as hexafluoropropylene oxide dimer acid (HFPO-DA) which are more polar than the longer chain legacy perfluorinated species[20,21]. As there has been a significant shift towards shorter fluorinated chains and ether inclusion in recent PFAS chemistry[5], this phase selection enables more thorough recovery of novel compounds for MS analysis.

Targeted LC-MS/MS quantitation using authenticated standards and stable isotope labeled internal standards provides an unparalleled level of specificity and sensitivity for the quantitative analysis. While this approach is desirable in many situations, it is impractical for all-too-common situations in analysis. Targeted approaches work only for species that are expected in the sample, and for which methods have previously been established. For new and emerging compounds,

this approach is incapable of even detecting species that may be of interest, regardless of their chemistry or concentration, and low-resolution mass spectrometers are nearly incapable of providing enough information to make unequivocal chemical assignments of unknown compounds. Consequently, the field of non-targeted analysis has arisen, leveraging the power of high-resolution modern mass spectrometers to analyze samples without a presupposed hypothesis and retroactively assign chemicals to detectable features in the sample. This approach has been used extensively in the fields of biology[22-24] and environmental science[25-27] on numerous classes of chemicals. Perfluorinated chemicals are particularly straightforward to identify in this method due to their unique mass spectral patterns, and hundreds of compounds have been described in just the past few years[5,28].

The protocol discussed here is intended to align targeted LC-MS/MS PFAS quantitation with the need to identify and semi-quantitatively monitor emerging compounds of interest. The SPE phase selection and sample preparation techniques are intended to ensure capture of more hydrophilic emerging PFAS acids from water and may be less suited for longer chain polymeric species and non-ionic species. Further, the data generated by non-targeted analysis is dense and of high dimensionality, which necessitates the use of data analysis software. Such software packages are frequently vendor specific and require modification to operate between instrument platforms. Where possible, the analysis process has been described in a generic fashion and open source/freeware alternatives are referenced, but the efficiency and accuracy of any software approach must be assessed on an individual basis.

PROTOCOL:

1. Collection of water samples

1.1 Preparation of PFAS Standard Stocks

1.1.1 Prepare a PFAS standard mixture in methanol containing any targeted compounds of interest (e.g., PFOA, PFOS, HFPO-DA) at 1 ng/µL. This is the Native PFAS Mixture. Commercially prepared mixtures are also available (i.e., PFAC Mix A and Mix B).

1.1.2 Prepare a standard mixture containing matched stable isotope labeled (SIL) PFAS compounds (e.g., $^{13}C_4$-PFOA, $^{13}C_8$-PFOS, $^{13}C_3$-HFPO-DA) at 1 ng/µL. This is the IS PFAS Mixture. Commercially prepared mixtures are also available (i.e., MPAFC Mix A and Mix B).

NOTE: If an SIL version of the targeted PFAS is unavailable, a surrogate with similar structure and chain length can be used (e.g., $^{13}C_2$-PFHxA for HFPO-DA)

1.2 Preparation of Field Blank (FB), Spike Blank (SB) samples

1.2.1 Fill two, clean high-density polypropylene (HDPE) or polypropylene (PP) bottles with 1,000 mL of laboratory deionized (DI) water, known to be PFAS free.

CAUTION: PFAS materials frequently have undefined toxicity and/or carcinogenicity. Care should be taken to avoid oral or skin exposure to standards or stock solutions.

1.2.2 Add a quantity of PFAS standard mixture to one of the bottles at a final concentration equivalent to the expected sample concentrations (e.g., 100 ng/L). This is the Spike Blank (SB).

1.2.3 Add 5 mL of 35% nitric acid preservative to the Spike Blank.

1.2.4 Carry both SB sample and the unspiked field blank to the sampling location as controls.

1.3 Field sampling

NOTE: Sample collector should wear nitrile gloves and sample from flowing systems where possible. Tap samples should be allowed to flow and equilibrate prior to sampling (2-3 min).

1.3.1 Collect 500-1000 mL of water from the field location in a clean HDPE or PP bottle.

1.3.2 Add 5 mL of 35% nitric acid preservative to sample bottles and field blank.

CAUTION: Nitric acid is corrosive and a strong oxidizer

2. Sample extraction

NOTE: PFAS are ubiquitous and persistent. Ensure that all solvents are of the highest grade and have been analyzed for low level PFAS contamination. Thoroughly rinse all laboratory equipment used for preparing standards before preparing blanks and samples.

2.1 Sample pretreatment

2.1.1 Pour each sample into a separate, pre-cleaned 1 L HDPE graduated cylinder and record the exact volume.

2.1.2 Add 10 mL of methanol to the emptied sample bottle, cap it, and shake well to rinse adsorbed PFAS from the bottle interior.

2.1.3 Return the measured water sample to the rinsed bottle with the methanolic rinse.

2.2 Standard curve for quantitation

2.2.1 Fill eight, 1 L HDPE/PP bottles with PFAS-free DI water.

2.2.2 Select eight evenly spaced concentrations covering the desired quantitation range. For example: 10, 25, 50, 100, 250, 500, 750 and 1,000 ng/L for a range of 10-1,000 ng/L.

2.2.3 Add a quantity of Native PFAS mix to each bottle to yield the final PFAS concentrations in 2.2.2 (e.g., 100 µL PFAS Mix A to 1L of DI water = 100 ng/L).

2.3 Internal standard addition

NOTE: Addition of stable isotope labeled internal standard (IS) is necessary only if quantitative results are desired in addition to non-targeted analysis.

2.3.1 Add the IS PFAS mixture to each sample at a concentration approximating the midpoint of the calibration curve (e.g., 250 µL of IS PFAS mix = 250 ng/L)

2.4 Filtration

2.4.1 Filter samples through GF/A glass fiber filters (47 mm, 1.6 µm pore size) under gentle vacuum into a pre-cleaned 1 L HDPE vacuum flask.

2.4.2 If particulate matter remains in the bottle, rinse with additional deionized water into the filter. Return the filtered water to the sample bottle or a new container for solid phase extraction.

2.5 Solid phase extraction (SPE)

NOTE: Cartridge concentration described here uses a constant flow piston pump. Alternative methods of concentration using a vacuum manifold[20] or using an on-line SPE-LC-MS[17] setup are possible but not discussed.

2.5.1 Condition a weak anion exchange (WAX) cartridge with 25 mL of methanol.

2.5.2 Condition the WAX cartridge with an additional 25 mL of deionized water.

2.5.3 Position pump draw tubing in filtered sample bottles and label SPE cartridges with corresponding sample names.

2.5.4 Pump 500 mL of sample water through the cartridge at a steady flow rate of 10 mL/min (500 mL total), discarding flow-through liquid to waste.

NOTE: Larger or smaller volumes can be concentrated depending on expected sample concentrations.

2.5.5 Remove the cartridge from piston pump for elution.

NOTE: If concentrating additional samples using the same pump, the piston pump should be flushed with 25 mL of methanol before installing the next cartridge for equilibration.

2.5.6 Transfer SPE cartridge to a vacuum manifold and equip with external glass reservoir.

2.5.7 Flush SPE cartridge with 4 mL of 25 mM, pH 4.0 sodium acetate buffer under gentle vacuum. Discard flow through. Wash SPE cartridge with 4 mL of neutral methanol.

NOTE: Neutral wash fraction can be collected if specific nonionic polar analytes are expected. Otherwise, discard to waste

2.5.8 Place a 15 mL polypropylene centrifuge tube beneath each SPE cartridge to collect eluent. Elute sample with 4 mL of 0.1% ammonium hydroxide in methanol.

2.5.9 Remove elution tube and reduce eluate volume to 500 – 1,000 µL by evaporation under dry nitrogen stream in a water bath at slightly elevated temperature (40 °C).

2.5.10 Concentrated sample extracts can be stored prior to analysis at room temperature.

2.6 Targeted LC-MS/MS quantitation

2.6.1 Dilute 100 µL of sample extract with 300 µL of 2 mM ammonium acetate buffer into an HPLC sample vial.

2.6.2 Calibrate and equilibrate an HPLC and MS systems according to manufacturer's instructions.

NOTE: Background PFAS are commonly detected due to the use of fluoropolymer components of most LC systems and in sample vial septa. Confirm that the detectable levels in blanks is negligible before use. Modification of the LC system to replace Teflon components is suggested where possible. The use of an analytical "hold-up" column adjacent to the LC mixing valve is also suggested[29].

2.6.3 Prepare an analytical worklist consisting of the standard curve, samples, and an additional replicate of the standard curve to assess instrumental drift across the run. An example worklist is shown in **Table 1**.

2.6.4 Analyze the samples using LC and MS methods established for the targeted compound(s) of interest. The example LC gradient is shown in **Table 2** and MS method parameters are shown in **Table 3** and **Table 4**. Further detailed discussion can be found in McCord et al.[21].

2.6.5 Generate a standard curve from the standard samples using the peak area ratio of the analyte to the internal standard versus the concentration of analyte. Generate a quadratic regression formula with 1/x weighting for concentration prediction[9].

2.6.6 Quantitate targeted analytes in each sample using the prepared standard curves and area ratio (standard area/IS area) for each measurement.

2.6.7 If the concentration exceeds the calibration range, dilute the original sample with DI water spiked with the appropriate IS concentration and re-extract to bring the concentration into the appropriate range.

2.7 Non-targeted LC-MS/MS data collection

2.7.1 Dilute 100 µL of sample extract with 300 µL of 2 mM ammonium acetate buffer into an HPLC sample vial.

2.7.2 Calibrate and equilibrate an HPLC and high-resolution MS according to manufacturer's instructions.

2.7.3 Prepare an analytical worklist as in 2.6.2.

2.7.4 Using the instrument software, collect LC-MS data in with a wide scan MS1 in data-dependent mode to collect MS/MS. Example LC gradient in **Table 5**. Further discussion of instrument settings can be found in Strynar et al.[30] and Newton et al.[31].

NOTE: For improved MS/MS quality data-dependent analysis can be carried out with a preferred ion list of a subset of features remaining after data processing in 2.8.1-2.8.8.

2.8 Non-targeted data processing

NOTE: Data analysis can be performed with a wide range of software and these methods do not reflect the only, or best method for an arbitrary dataset. Where possible, steps provide a generic description that can be carried out in alternate software. Processing of the example data used in this manuscript was carried out using vendor specific software (Software 1 and Software 2) as detailed in Newton et al.[31].

2.8.1 Perform molecular feature extraction of chemical features using one of several open source software packages[32,33] or vendor software to identify monoisotopic masses, retention times, and integrated peak areas of chemical features.

2.8.1.1 In Software 1, select **Add/Remove Sample Files** > **Add Files** and select the raw data from the non-targeted experiment, then hit **OK**.

2.8.1.2 In Software 1 select **Batch Recursive Feature Extraction** > **Open Method…** to load a preestablished method, or manually edit software settings. Profinder settings for feature extraction are found in **Table 6**.

2.8.1.3 In **Software 1**, after feature extraction, select **File** > **Export as CSV**…, **File** > **Export as CEF**…, or **File** > **Export as PFA**… for further processing. CEF files are assumed for the remainder of the description.

2.8.1.4 In Software 2 (MPP) create a new experiment with Type **Unidentified** and Workflow type **Data Import Wizard** and click **OK**.

2.8.1.5 In MPP **Select Data Files** and locate the exported Software 1 results (either CEF or PFA) to import; then click **Next** until **Alignment Parameter** options appear.

2.8.1.6 In MPP, set the Compound Alignment values to 0.0 (alignment was already performed in the feature extraction of Software 1, step 2.8.1.2) then click **Next** through the steps until **Finish** is available.

2.8.2 Filter identifications based on analytical reproducibility. Where multiple replicate samples are available, features should be present in >80% of individual replicates and have an analytical coefficient of variation (CV) of < 30%

2.8.2.1 In MPP select **Experimental Setup** > **Experiment Grouping** and assign each raw file a group corresponding to its origin sample (i.e., replicates from the same source should be in the same group). Multiple groups can be created corresponding to nested variables (e.g., instrumental vs. technical replicates).

2.8.2.2 In MPP select **Experimental Setup** > **Create Interpretation** then select the experiment parameter (i.e., group) and click **Next** until **Finish** is available. This will create a category that future filtering can operate on.

2.8.2.3 In MPP select **Quality Control** > **Filter by Frequency**. Set Entity List to **All Entities** and the Interpretation to the sample Group(non-averaged) created in 2.8.2.2, then hit **Next**.

2.8.2.4 For Input parameters, set entity retention at 80% of sampled in at least one condition then click **Next** until **Finish** is available. Name the list Frequency Filtered Features

2.8.2.5 In MPP select **Quality Control** > **Filter on Sample Variability**. Set the Entity List to the Frequency Filtered Features from 2.8.2.4 and the interpretation to Group(non-averaged), then hit **Next**.

2.8.2.6 Select the radio button for **Raw Data** and the Range of Interest to Coefficient of variation < 30%. Click **Next** > **Finish** and save the list as CV Filtered Features.

2.8.3 Remove features where no samples have significantly higher (>3 fold) abundance than the Field Blank (FB) sample.

2.8.3.1 In MPP select **Analysis > Fold Change**. Set Entity List to **CV Filtered Features** and the Interpretation to the sample Group then hit Next. Select the fold change option to **All against single condition** and select condition **FB** or whatever the group name for the blank processed sample was.

2.8.3.2 On the following screen, set the Fold-Change cutoff to 3.0 and click through to the end of the prompts. Save the list as **FC Filtered List**.

2.8.4 Perform binary comparisons of individual samples of interest against an appropriate background sample (e.g., upstream vs. downstream of a point source) to determine fold-changes for individual chemical features.

2.8.4.1 In MPP select **Analysis > Filter on Volcano Plot**. Set the entity list to **FC Filtered List** and the Interpretation to Group.

2.8.4.2 For the fold-change condition pair choose two samples for comparison (e.g., a paired upstream and downstream sample) and select test **Mann-Whitney Unpaired**.

2.8.4.3 For preliminary analysis, do not select a value for multiple test correction on the following screen, click through to the result plot.

2.8.4.4 On the results screen select a fold-change cutoff of 3.0 and a p-value cutoff to 0.1. Then **Finish** and export the list as **Prelim Results**.

2.8.5 For each feature remaining after filtering, generate predicted chemical formula(s) from the exact mass and composite mass spectrum.

2.8.5.1 In MPP, select **Results Interpretation > IDBrowser Identification** and the **Prelim Results** entity list.

2.8.5.2 In the IDBrowser select **Identify all compounds** using molecular formula generator (MFG) as the identification method.

2.8.5.3 In the Generate Formula options add F to the Elements column and set the Maximum to 50, then select **Finish**. Following formula generation select **Save** and **Return** to return to MPP.

2.8.5.4 In MPP, right click the filtered and MFG matched Entity List and select **Export List**. Save the results.

2.8.6 Examine the monoisotopic mass of species in the reduced significant chemical feature list for those containing mass defects indicative of fluorination; see Kind and Fiehn[34].

2.8.7 Note chemical series containing common polyfluorination motifs (CF2 (m/z 49.9968), CF2O (m/z 65.9917), CH2CF2O (m/z 80.0074), etc.) using a mass defect plot or software algorithm; see discussion section, Liu et al.[17], Loos et al.[35] , and Dimzon et al.[36].

2.8.8 Search predicted chemical formulas or neutral masses against the EPA Chemistry Dashboard database and/or other databases to return potential chemical structures.

2.8.8.1 Open the EPA Comptox Chemicals Dashboard Batch Search tool (https://comptox.epa.gov/dashboard/dsstoxdb/batch_search) and paste the list of identifiers (either formulas or masses) into the identifier box, after selecting the identifier type (i.e., MS-ready Formula or Monoisotopic Mass).

2.8.8.2 Select **Download Chemical Data…** and also select any physical/chemical/toxicology data desired for potential matches from the dropdown.

2.8.9 Using chemical intuition and available reference data, remove unlikely matches from the potential chemical structure list for each formula based on feasibility due to chemical stability, physical properties such as ionizability or hydrophobicity, the presence of manufacturing chemicals from nearby sources, etc. In the absence of additional data, spectral feasibility can be ranked purely on the basis of literature prevalence; see McEachran et al.[37].

2.8.10 Confirm structures using available standards and/or targeted high-resolution MS/MS matching of fragments against spectra from databases, *in silico* theoretical spectra, or manual curation.

REPRESENTATIVE RESULTS:

Quantitative LC-MS/MS results are in the form of ion-chromatograms for the total ion chromatogram (TIC) and the extracted ion chromatograms (EIC) of specific chemical transitions for measured chemicals (**Figure 1**). The integrated peak area of a chemical transition is related to the compound abundance and can be used to calculate the exact concentration using a calibration curve normalized to an internal standard (**Figure 2**). Low or flat response of individual analytes indicates that the calibration range is outside the linear range of the mass spectrometer, or that the instrument requires tuning/calibration. Poor precision of replicates indicates an issue with sample injection or inconsistent chromatography that requires modification of LC parameters.

Non-targeted analysis using a full MS1 scan yields a TIC for samples (**Figure 3**), which allows for ad hoc generation of EICs for individual ions (**Figure 4**). Any given chromatographic time point contains signals for chemical species, and when using a high-resolution mass spectrometer, the isotopic fingerprint of the compound. Identifying compounds from the MS1 scan is performed programmatically by a peak-picking algorithm using one of several approaches[38-40]. Peak picking yields chemical features with a measured accurate mass and chromatographic retention time, as well as the mass spectrum of the ion and the chromatographic peak area. This information is typically stored in a digital database format for further processing and filtering, but the nested and interconnected nature of the data can be understood conceptually (**Figure 5**).

The feature list is filtered for compounds meeting one of several criteria to be selected for further investigation. The first and most straightforward is filtering by mass defect (the difference between the exact mass of a feature and its nominal mass). PFAS compounds have negative mass defects (**Figure 6**) due to their preponderance of fluorine atoms, and polyfluorinated compounds have positive, but substantially smaller mass defects than homologous organic materials[31,34]. A

second method filtering step is to identify homologous series containing repeating units common to PFAS species, such as CF2 or CF2O. Identifying these can be done using Kendrick Mass defect plots[17,36], or software packages such as R's *nontarget* package[35] (**Figure 7**).

Following filtering, assignment of chemical identity on the shortlist of highly differentially observed and/or tentatively per/polyfluorinated species can begin. Accurate mass provides a relatively small list of potential chemical formulas for matching but is insufficient for identification without the addition of spectral matching to isotope pattern of the mass spectrum[41]. From high resolution MS1 data, one or more putative chemical formulas are matched against the isotopic fingerprint of the mass spectrum and scored (**Figure 8**). Formulas for matching can be generated ab initio using a defined pool of atoms or can be sourced from a combination of literature reported compounds and the contents of one or more databases. The US EPA Chemistry Dashboard (https://comptox.epa.gov/dashboard/) hosts a constantly updated list of PFAS compounds identified by the agency, as well as lists compiled by other organizations such as the NORMAN Network[42].

Chemical formulas can be further confirmed, and some structural information can be garnered from MS/MS spectra (**Figure 9**). Candidate structures are available from large chemical databases such as the EPA chemistry dashboard, Pubchem, the CAS registry, etc. Predicted spectra can be generated or acquired using a variety of fragmentation programs and assigned,[43] or MS/MS spectra can be interpreted manually.

An example data matrix is available in the Supplemental Information containing a whole feature matrix from ten samples (5 upstream, 5 downstream) collected upstream and downstream of a fluorochemical point source. Each row represents a chemical feature with associated retention time, neutral mass, mass spectrum, and raw abundance for each sample. (**Supplemental Table**, **Sheet 1**). Initial filtering (**Supplemental Table**, **Sheet 2**) for negative mass defect and statistical significance in an unpaired t-test between upstream and downstream reduces the number of "interesting" chemical features to ~120. Predicted chemical formulae were obtained from Agilent IDBrowser and searched against the EPA Comptox Chemicals Dashboard, which returned possible matches (**Supplemental Table**, **Sheet 3**). The "top-hit" for each chemical formula based on data sources[37] was assigned (**Supplemental Table**, **Sheet 4**). Note that more than half of the remaining features do not have high quality matches. Identified features with no matches can be the result of in-source fragmentation/adduct formation, poor formula assignment, or the identification of PFASs not found in the source database. Interpretation of the raw spectra in order to validate assignments is beyond the scope of this manuscript but more information can be found in the works cited[15,30,31,44,45].

FIGURE AND TABLE LEGENDS:

Table 1: Example worklist for Targeted Analysis and quantitation of PFAS using LC-MS/MS

Table 2: Example gradient for LC separation in targeted analysis

Table 3: Ionization source parameters for targeted analysis

Table 4: Example transition table and MS/MS parameters for the contents of PFAC-MXA, along with HFPO-DA

Table 5: Example gradient for LC separation in non-targeted analysis

Table 6: Molecular feature extraction and alignment settings for Profinder software. All unlisted values retained their default settings for data processing.

Table 7: Comparison of sample processing time and chemical feature identifications for different feature extraction thresholds.

Figure 1: Total ion chromatogram and extracted ion chromatograms for a subset of perfluorinated ether standards

Figure 2: Representative calibration curves for compounds demonstrating decreasing quality of analytical curve construction. Left-most panel indicates a high quality calibration; Middle panel indicates a compound with poor precision across preparation duplicates, particularly at the higher concentrations; Right Panel indicates a curve with poor precision and a low linear dynamic range, resulting in flat response at the high end of the calibration range, and no detectable signal at the lower end.

Figure 3: Overlaid total ion chromatograms (TIC) for surface water extracts collected upstream and downstream of a fluorochemical production site

Figure 4: Extracted ion chromatograms (EIC) for all identified chemical features from a surface water sample containing multiple fluorochemical classes. Each chemical trace is a different color for differentiation.

Figure 5: Conceptual diagram of raw and predicted information for a chemical feature identified as hexafluoropropylene oxide dimer acid (HFPO-DA). Chemical features are compiled from software extraction of raw data from MS measurements and contain chromatographic (e.g., retention time (RT)) and mass spectrometry information. Predicted formula, structures, and chemical identities are generated from raw measurement data for each feature.

Figure 6: Mass defect plot for chemical features identified in a manufacturing outfall (red, left) and reference surface water (blue, right). Fluorinated compounds fall near and below the dashed zero line. Note the persistent PFOA/PFOS series in the background surface water sample (right).

Figure 7: Mass vs mass defect plot for unidentified chemical features from a surface water sample with homologous series identified and labeled by the *nontarget* R package.

Figure 8: Mass spectrum of an unknown chemical features with predicted isotopic intensities of three possible chemical formula with the same monoisotopic mass.

Figure 9: Fragmentation spectrum of a perfluorinated ether compound with annotated fragment peaks.

Figure 10: Graphical representation of filtering thresholds. From left to right, ion abundance threshold for chemical feature mass spectra, feature abundance threshold for extracted chromatographic features, and replicate threshold for feature detection frequency in a triplicate injection experiment.

DISCUSSION:

Sample Handling and Preparation

The inclusion of reference/spike standards are of paramount importance to any targeted analysis, as they provide a backstop for checking analytical validity. Lack of QC samples prevents any assessment of the accuracy of the results; the ubiquitous nature of fluorochemicals means that chance contamination of field samples, processing materials, or LC-MS system is not uncommon and must be accounted for. Further, it allows for the validation of the protocol regardless of variation in the day-to-day sample processing, as many of the steps can be highly variable, particularly the SPE and sample concentration steps. The extraction of both legacy and novel perfluorinated chemicals can be heavily influenced by the choice of stationary phase for concentration, and components of the source samples, such as pH and salinity[46]. The influence of sample conditions should be considered if particular classes of pefluorinated chemicals are of interest. Alternative sample preparation schemes for water extracts can be used if the laboratory setup is available and the downstream data analysis remains similar.

Targeted Data Analysis

For compounds with available standards and matched, stable isotope labeled internal standards, the primary concerns for data analysis are instrumental and determination of method detection limits and suitable reporting ranges can be determined on a laboratory-by-laboratory basis using standard approaches, such as signal-to-noise ratio from low-level standard spikes[47]. In the absence of matched internal standards errors from mismatched matrix effects can occur, and accurate back-prediction of spiked samples can be used to estimate the accuracy of the measurements. When lacking standards to prepare a curve, a quantitative estimate of an unknown can be made by treating it identically to a closely matched standard compound, but errors in the estimate are on the order of 10+ fold with limited ability to quantify the uncertainty, see McCord, Newton, and Strynar[21]. In these cases, trend data can still be collected, but concentration estimates are inherently unreliable.

Non-targeted Data Analysis

Peak picking settings have a substantial impact on the number of chemical features identified, but the quality of feature selection is also heavily impacted. The decisions of interest in peak picking are 1) intensity of individual masses to be included in spectra, the ion abundance threshold 2) the intensity of extracted chromatogram peaks to be considered features, the

feature abundance threshold 3) feature detection frequency, the replicate threshold, and 4) analytical variation, the CV threshold (**Figure 10**).

Setting unrealistically low thresholds for peak picking results in an exponential increase in sample time to resolve additional features of increasingly low abundance (**Table 7**). The ion-abundance threshold filters mass spectral features where enough of the individual isotope abundances do not pass the threshold. This ideally selects only for features with quality MS spectra, ensuring they are real chemical features rather than instrumental noise, and allowing for formula prediction in downstream processing. An appropriate threshold is based on instrumental noise, ideally at least 3x the noise threshold for MS1 scans. Feature abundance threshold filters chemical features based on the intensity or area of the chromatographic feature extracted. This step enables rejection of low abundance peaks, which are typically of poor chromatographic quality, have high variances, or are the result of other poor software extraction. An appropriate threshold must be determined per experiment, and per matrix based on an acceptable level of poor feature generation (e.g., features below the threshold exhibit unacceptably poor chromatography). Further analytical QC can be used to reject features at the chromatographic level based on inconsistent identification in analytical and/or preparatory replicates (replicate threshold) or based on poor reproducibility across replicates (CV threshold). Appropriate levels depend on the quality of the peak integration software used and the chemical entities under investigation. For water soluble perfluorinated compounds and lightly optimized integration protocols, features should be identified in 80+% of analytical replicates and CVs are expected to fall below 30%, as detailed in the methods section.

The peaks detected from non-targeted analysis do not yield quantitative estimates of the concentrations of the materials detected. Further, the identity of true unknowns can be difficult to confirm because novel compounds are absent from publicly available databases. Novel structural determination requires extensive analysis with multiple methods and requires expertise in both mass spectrometry and chemistry. However, normalizing the peak areas of chemical features can provide semi-quantitative estimates of concentrations of unknowns from known species[21]. If consistent sampling and preparation steps are employed, time trend information for individual species can be generated to monitor the persistence of a chemical into the future as the response for an individual species should be consistent barring large variations in the matrix[21].

The primary benefit of this method is the extensibility of the sample treatment to allow both targeted and nontargeted analysis. While targeted analysis provides equivalent or superior quantitative information, it greatly lacks breadth of analysis desired when dealing with new and emerging materials, as well as their relationship to matrix materials. Applying a targeted methodology, or even a suspect screening method based only on known materials and limited databases is completely blind to previously unobserved species, even if they may have significant health effects. As software improves and databases become more robust, the accuracy of unknown identification will continue to increase, with a concomitant decrease in the time investment and level of expertise necessary to analyze the multidimensional data generated by this approach. Nevertheless, data generated presently is of significant future value because data

banking allows for post-hoc analysis with newly developed software and enables comparison across time even if the identity of a detected compound is currently unknown.

ACKNOWLEDGMENTS:

The U.S. Environmental Protection Agency, through its Office of Research and Development, funded and managed the research described here. This document has been reviewed by the U.S. Environmental Protection Agency, Office of Research and Development, and approved for publication. The views expressed in this article are those of the authors and do not necessarily represent the views or policies of the U.S. Environmental Protection Agency. This research was supported in part by an appointment to the Postdoctoral Research Program at the National Exposure Research Laboratory administered by the Oak Ridge Institute for Science and Education through Interagency Agreement DW89992431601 between the U.S. Department of Energy and the U.S. Environmental Protection Agency.

DISCLOSURES:

The authors have nothing to disclose.

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Figure 1

Click here to access/download;Figure;Figure 1 - PFECA Standard Chromatograms.tif

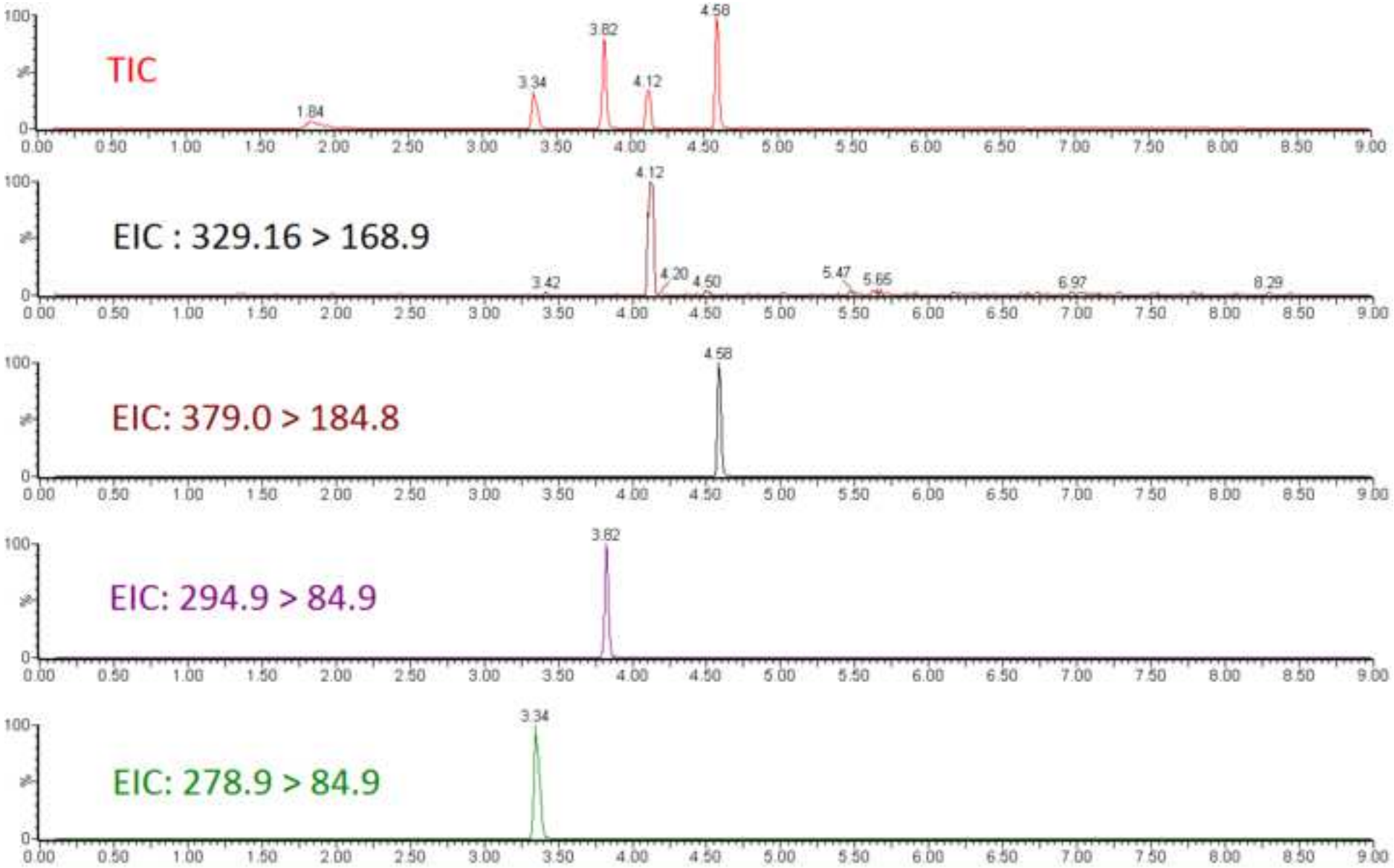
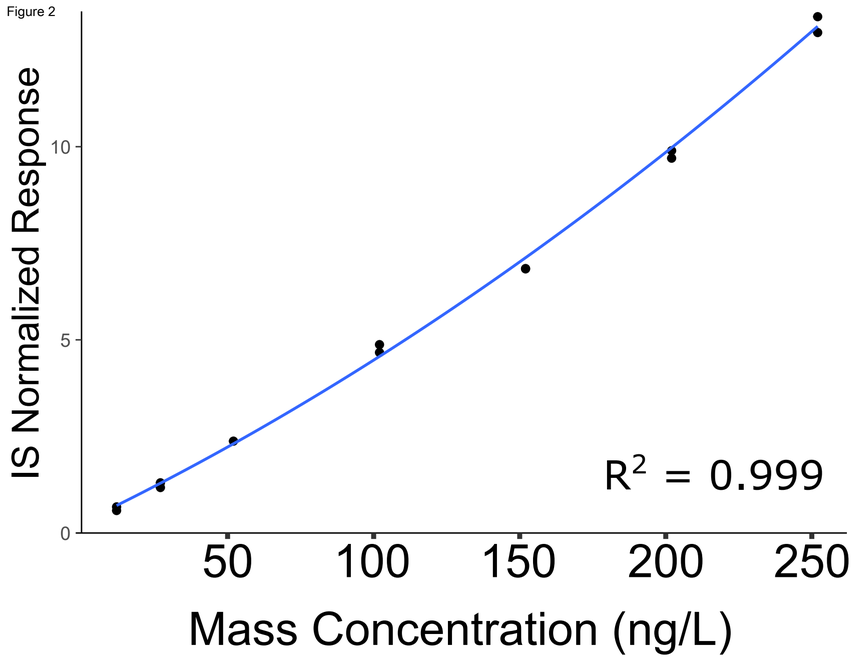
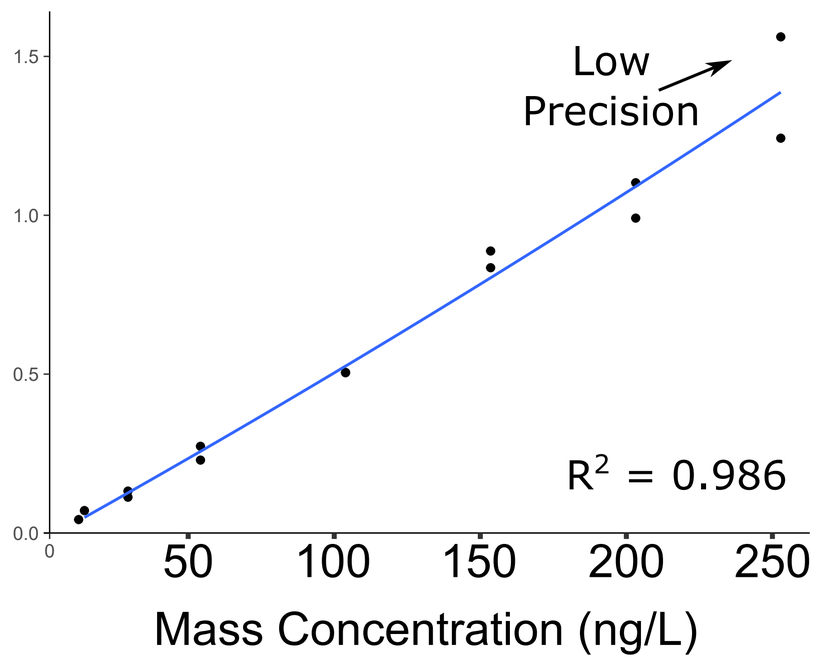


Figure 2

Click here to access/download;Figure;Figure 2 - Sample Calibration.pdf





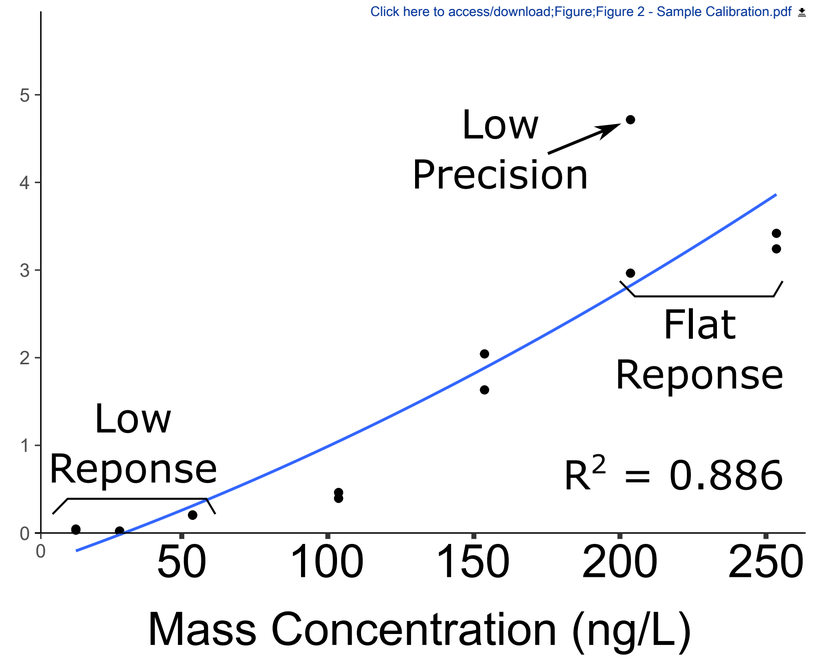


Figure 3

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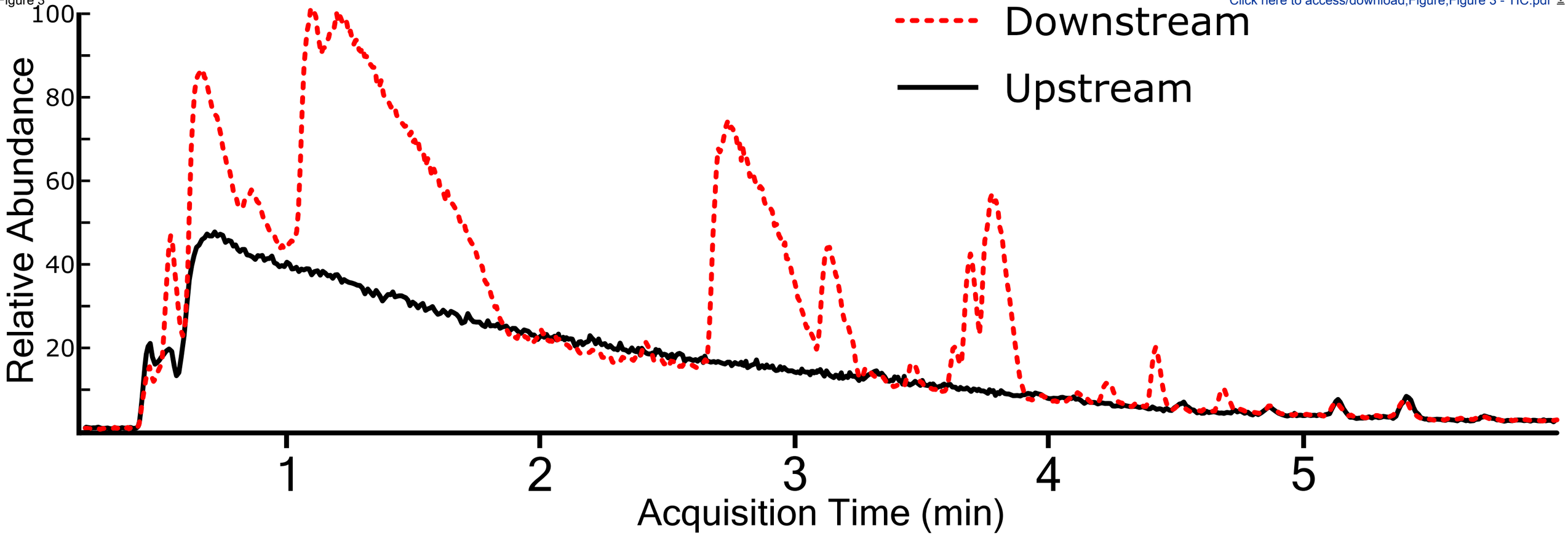

Figure 4

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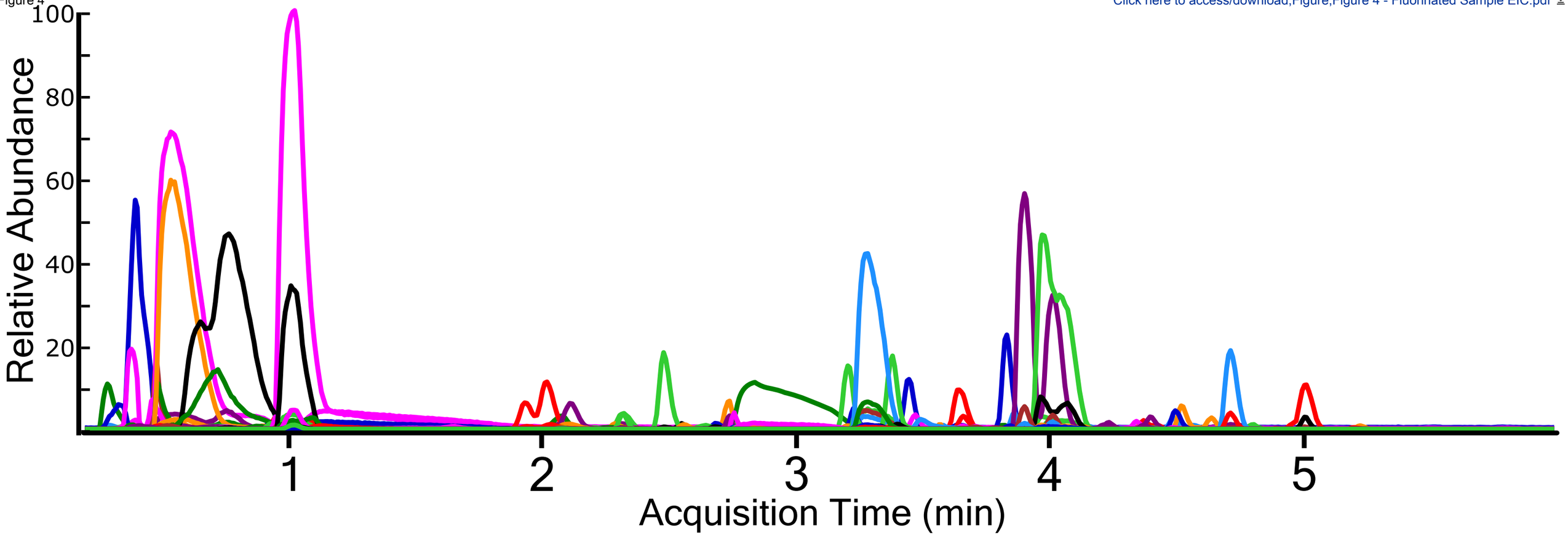

Figure 5

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Chemical Feature

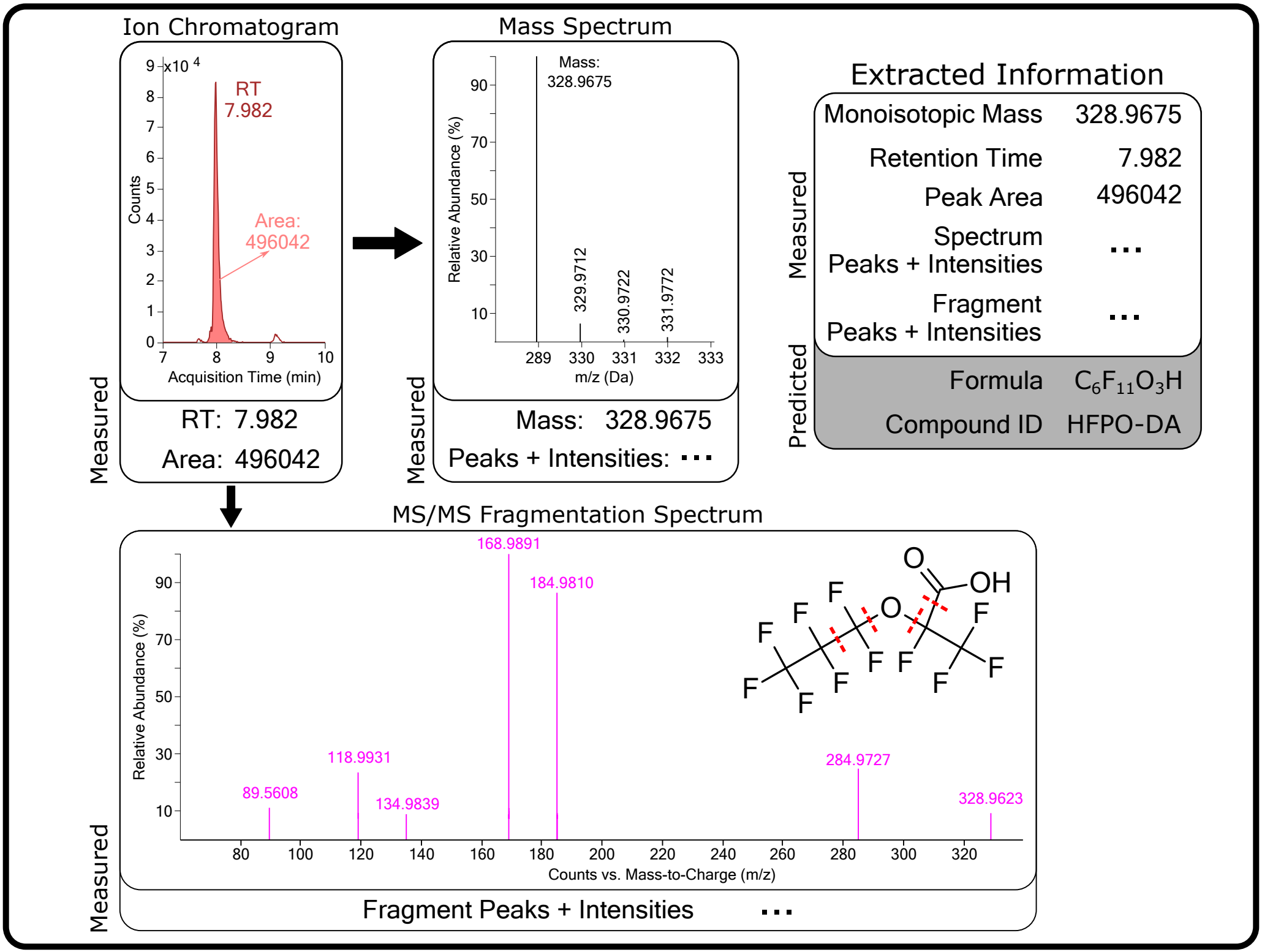

Figure 6

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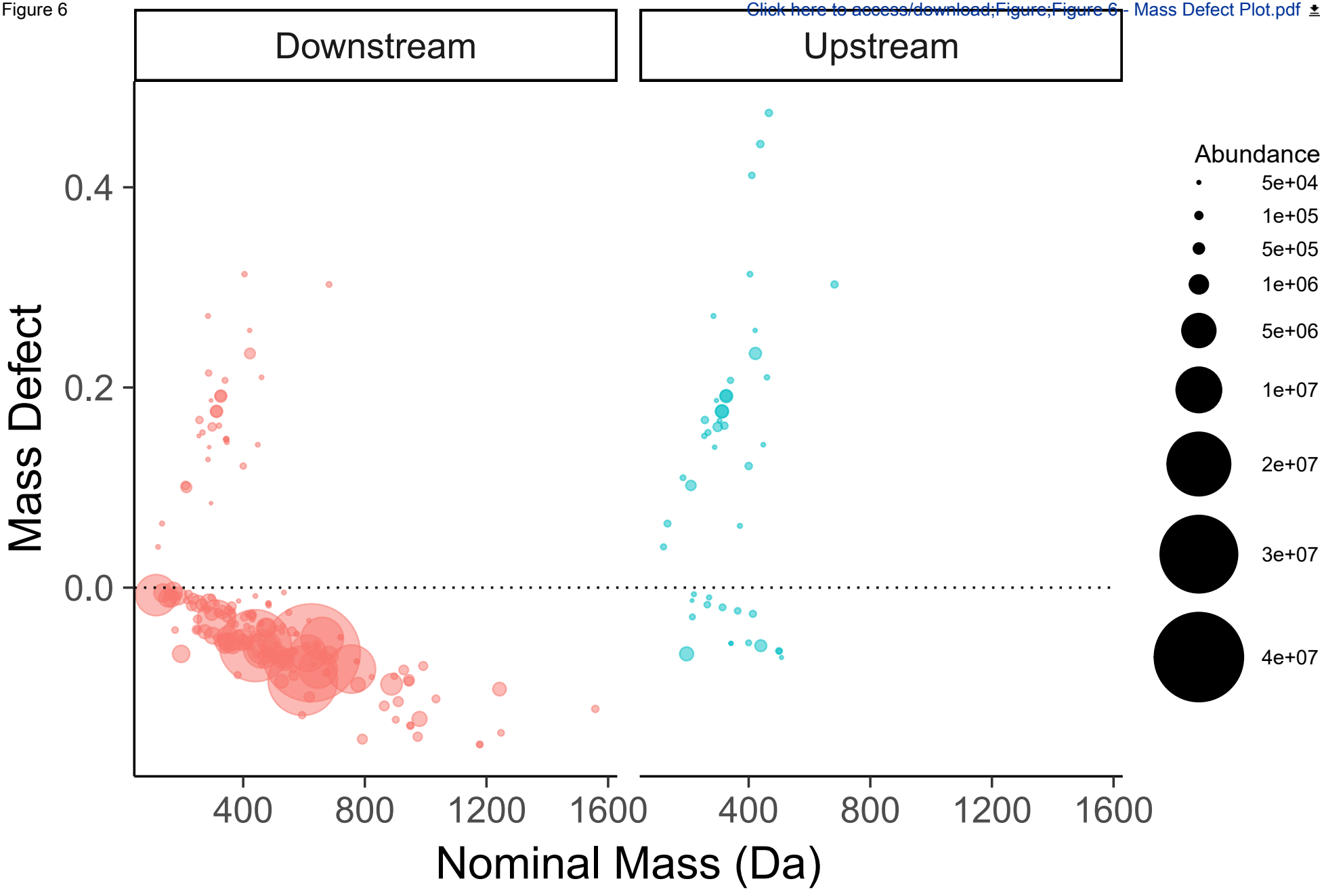

Figure 7

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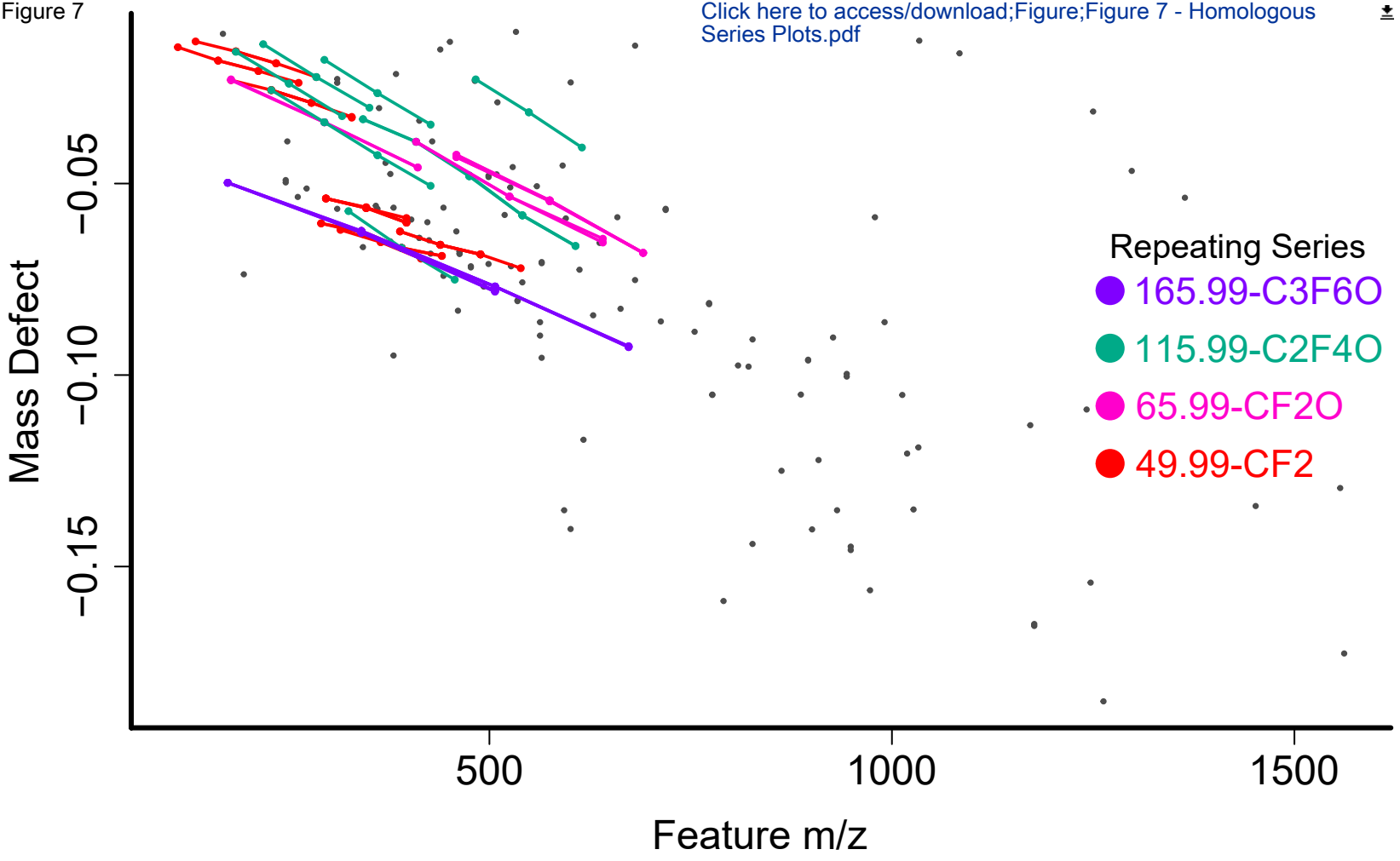

Figure 8

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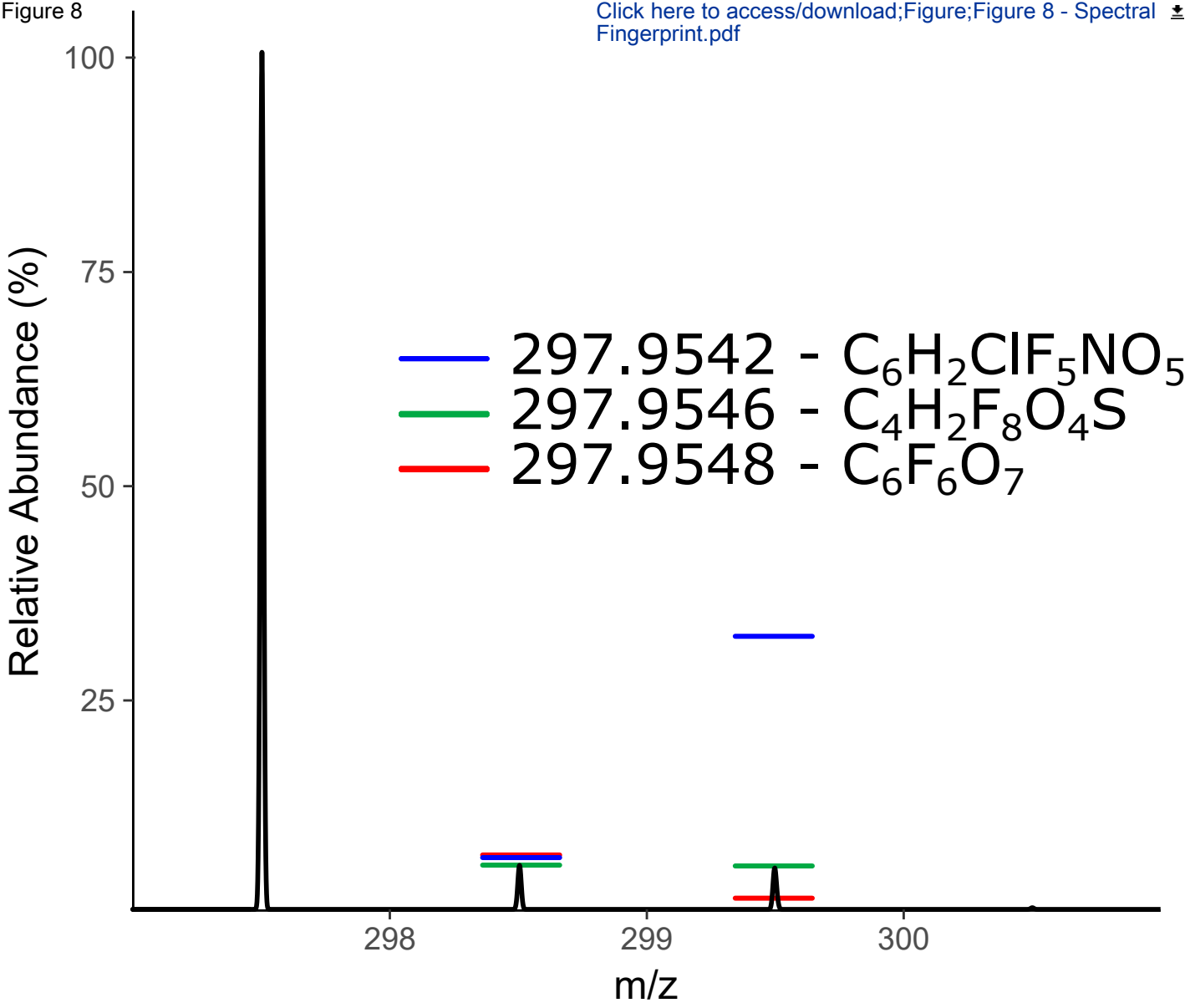

Figure 9

Click here to access/download;Figure;Figure 9 - Annotated MSMS Spectrum.pdf

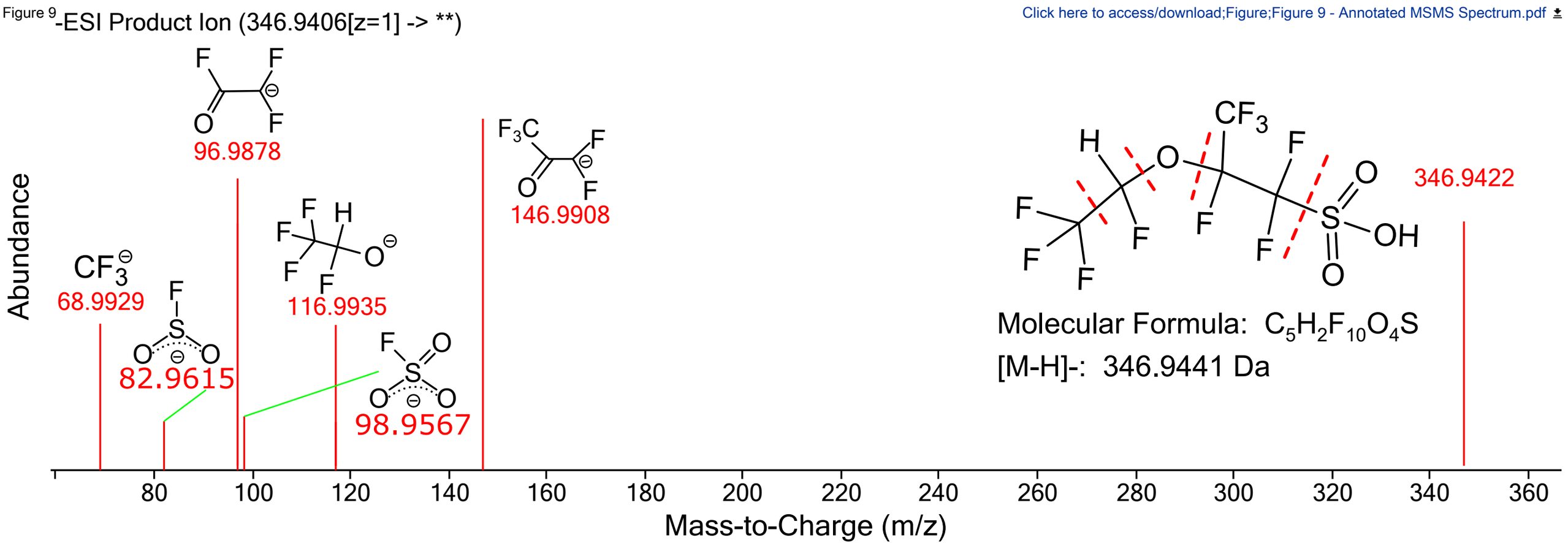

Figure 10

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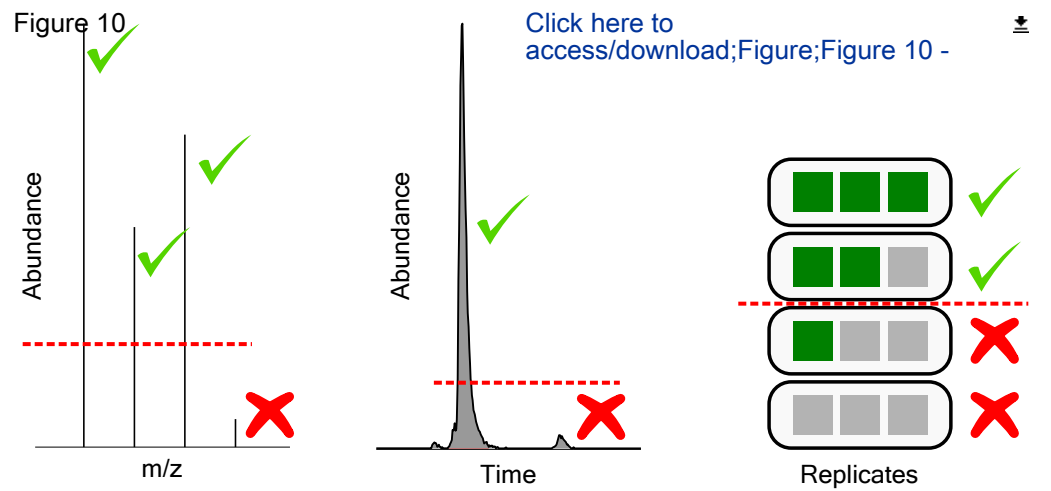

Table 1

Click here to access/download;Table;Table 1 - Targeted Sample List.xlsx

ID	Sample Name	Sample Type	Std Conc	Vial	LC Method
1	DB_001	Blank		1:A,1	PFAS grad 400uL/min - 9 min run
2	DB_002	Blank		1:A,1	PFAS grad 400uL/min - 9 min run
3	DB_003	Blank		1:A,1	PFAS grad 400uL/min - 9 min run
4	DB_004	Blank		1:A,1	PFAS grad 400uL/min - 9 min run
5	DB_005	Blank		1:A,1	PFAS grad 400uL/min - 9 min run
6	FB	Blank		1:A,2	PFAS grad 400uL/min - 9 min run
7	10 std	Standard	10	1:A,3	PFAS grad 400uL/min - 9 min run
8	25 std	Standard	25	1:A,4	PFAS grad 400uL/min - 9 min run
9	50 std	Standard	50	1:A,5	PFAS grad 400uL/min - 9 min run
10	100 std	Standard	100	1:A,6	PFAS grad 400uL/min - 9 min run
11	250 std	Standard	250	1:A,7	PFAS grad 400uL/min - 9 min run
12	500 std	Standard	500	1:A,8	PFAS grad 400uL/min - 9 min run
13	750 std	Standard	750	1:B,1	PFAS grad 400uL/min - 9 min run
14	1000 std	Standard	1000	1:B,2	PFAS grad 400uL/min - 9 min run
15	DB_006	Blank		1:B,3	PFAS grad 400uL/min - 9 min run
16	SB_DUP1	Analyte		1:B,4	PFAS grad 400uL/min - 9 min run
17	SB_DUP2	Analyte		1:B,5	PFAS grad 400uL/min - 9 min run
18	SW Site 03	Analyte		1:B,6	PFAS grad 400uL/min - 9 min run
19	SW Site 16	Analyte		1:B,7	PFAS grad 400uL/min - 9 min run
20	SW Site 30	Analyte		1:B,8	PFAS grad 400uL/min - 9 min run
21	DB_007	Analyte		1:C,1	PFAS grad 400uL/min - 9 min run
22	SW Site 19	Analyte		1:C,2	PFAS grad 400uL/min - 9 min run
23	SW Site 48	Analyte		1:C,3	PFAS grad 400uL/min - 9 min run
24	SW Site 49	Analyte		1:C,4	PFAS grad 400uL/min - 9 min run
25	SW Site 05	Analyte		1:C,5	PFAS grad 400uL/min - 9 min run
26	SW Site 47	Blank		1:C,6	PFAS grad 400uL/min - 9 min run
27	DB_008	Analyte		1:C,7	PFAS grad 400uL/min - 9 min run
28	SW Site 19_DUP	Analyte		1:C,8	PFAS grad 400uL/min - 9 min run
29	SW Site 20	Analyte		1:D,1	PFAS grad 400uL/min - 9 min run
30	SW Site 21	Analyte		1:D,2	PFAS grad 400uL/min - 9 min run
31	SW Site 46	Analyte		1:D,3	PFAS grad 400uL/min - 9 min run
32	SW Site 47	Analyte		1:D,4	PFAS grad 400uL/min - 9 min run
33	DB_009	Blank		1:D,5	PFAS grad 400uL/min - 9 min run
28	SW Site 32	Analyte		1:D,6	PFAS grad 400uL/min - 9 min run
29	SW Site 50	Analyte		1:D,7	PFAS grad 400uL/min - 9 min run
30	SW Site 25	Analyte		1:D,8	PFAS grad 400uL/min - 9 min run
31	SW Site 21_DUP	Analyte		1:E,1	PFAS grad 400uL/min - 9 min run
32	SW Site 52	Analyte		1:E,2	PFAS grad 400uL/min - 9 min run
33	DB_010	Blank		1:E,3	PFAS grad 400uL/min - 9 min run
34	FB	Blank		1:A,2	PFAS grad 400uL/min - 9 min run
35	10 std	Standard	10	1:A,3	PFAS grad 400uL/min - 9 min run
36	25 std	Standard	25	1:A,4	PFAS grad 400uL/min - 9 min run
37	50 std	Standard	50	1:A,5	PFAS grad 400uL/min - 9 min run
38	100 std	Standard	100	1:A,6	PFAS grad 400uL/min - 9 min run
39	250 std	Standard	250	1:A,7	PFAS grad 400uL/min - 9 min run
40	500 std	Standard	500	1:A,8	PFAS grad 400uL/min - 9 min run

41	750 std	Standard	750	1:B,1	PFAS grad 400uL/min - 9 min run
42	1000 std	Standard	1000	1:B,2	PFAS grad 400uL/min - 9 min run
43	DB_011	Blank		1:B,2	PFAS grad 400uL/min - 9 min run
44	DB_012	Blank		1:E,4	PFAS grad 400uL/min - 9 min run

MS Method
PFCMXA + HFPO-DA MS/MS - 9 min
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PFCMXA + HFPO-DA MS/MS - 9 min

Table 2

Click here to access/download;Table;Table 2 - Targeted LCMSMS Gradient.xlsx

Time (min)	% A (2.5mM Ammonium Acetate in 5% MeOH)	% B (2.5mM Ammonium Acetate in 95% MeOH)
0	90	10
5	15	85
5.1	0	100
7	0	100
7.1	90	10
9	90	10

Table 3

Click here to access/download;Table;Table 3 - Targeted Source Params.xlsx

Capilary Voltage (kv)	1.97
Cone Voltage (V)	15
Extractor Voltage (V)	3
RF Lens (V)	0.3
Source Temp	150
Desolvation Temp	40
Desolvation Gas Flow (L/hr)	300
Cone Gas Flow (L/hr)	2

Table 4

Click here to access/download;Table;Table 4 - Targeted Instrument Method.xlsx

Cmp	Precursor	Product	Dwell Time	Cone Volta	Collision Energy (eV)
PFBA	212.80	168.75	0.01	15	10
13C4-PFBA IS	216.80	171.75	0.01	15	10
PFPeA	262.85	218.75	0.01	15	9
PFBS °1	298.70	79.90	0.01	40	30
PFBS °2	298.70	98.80	0.01	40	28
PFHxA °1	312.70	118.70	0.01	13	21
PFHxA °2	312.70	268.70	0.01	13	10
13C2-PFHxA IS	314.75	269.75	0.01	13	9
HFPO-DA 1°	329.16	168.90	0.01	10	12
HFPO-DA 2°	329.16	284.90	0.01	10	6
HFPO-DA IS 1°	332.16	168.90	0.01	10	12
HFPO-DA IS 2°	332.16	286.90	0.01	10	6
PFHpA °1	362.65	168.65	0.01	14	17
PFHpA °2	362.65	318.70	0.01	14	10
PFHxS °1	398.65	79.90	0.01	50	38
PFHxS °2	398.65	98.80	0.01	50	32
13C4-PFHxS IS	402.65	83.90	0.01	50	38
PFOA °1	412.60	168.70	0.01	15	18
PFOA °2	412.60	368.65	0.01	15	11
13C4-PFOA IS	416.75	371.70	0.01	15	11
PFNA °1	462.60	218.75	0.01	15	17
PFNA °2	462.60	418.60	0.01	15	11
PFNA IS	467.60	422.60	0.01	15	11
PFOS °1	498.65	79.90	0.01	60	48
PFOS °2	498.65	98.80	0.01	60	38
13C4-PFOS IS	502.60	79.70	0.01	60	48
PFDA °1	512.60	218.75	0.01	16	18
PFDA °2	512.60	468.55	0.01	16	12
13C2 - PFDA IS	514.60	469.55	0.01	16	12

Table 5

Click here to access/download;Table;Table 5 - NTA Gradient.xlsx

Time (min)	% A (2.5mM Ammonium Acetate in 5% MeOH)	% B (2.5mM Ammonium Acetate in 95% MeOH)
0	90	10
0.5	90	10
3	50	50
3.5	50	50
5.5	40	60
6	40	60
7	0	100
11	0	100

Table 6

Click here to access/download;Table;Table 6 - Profinder Settings.xlsx

Profinder Parameter	
Extraction Peak Height Filter	
Permitted Ion(s)	
Feature Extraction Isotope Model	
Allowed Charge States	
Compound Ion Count Threshold	
Alignment RT Tolerance	
Alignment Mass Tolerance	
Post-Processing Absolute Height Filter	
Post-Processing MFE Score Filter	
Peak Integration Algorithm	
Peak Integration Height Filter	
Find by Ion Absolute Height Filter	
Find by Ion Score Filter	

Setting Value
800 counts
-H/+H
Common organic molecules
2-Jan
Two or more ions
0.40min + 0.0%
20.00ppm + 2.0mDa
>= 10000 counts in one sample
>= 75 in one sample
Agile 2
>= 5000 counts
>= 7500 counts in one sample
>= 50.00 in one sample

Table 7

Click here to access/download;Table;Table 7 - Profinder Results Comparison.xlsx

Ion Abundance Threshold	Feature Thresholds	Replicate Threshold (n =5)	Run Time	Features
1x S/N	2000	None	8.15	987
2x S/N	5000	None	5.02	707
3x S/N	10000	None	2.3	308
1x S/N	2000	100%	3.3	603
2x S/N	35000	100%	1.58	310
3x S/N	10000	100%	1.45	202

Pass Replicate Threshold	Pass CV Threshold	Features to 90% of TIC
505	421	91
357	313	93
249	230	93
339	297	92
248	229	93
190	182	92

Table of Materials

Click here to access/download;Table of Materials;JoVE Materials.xlsx

Name of Material/ Equipment	Company	Catalog Number
Acqity ultra-high performance liquid chromatography system	Waters Corporation	
Ammonium acetate	Fluka	17836
Ammonium Hydroxide	Sigma-Aldrich	338818
Balance	Mettler	AB204S
BEH C18 reverse phase UPLC column, 2.1×50 mm, 1.7 µm	Waters Corporation	186002350
Dual piston syringe pump	Waters Corporation	SPC10-C
Glacial Acetic Acid	Sigma-Aldrich	ARK2183
Glass Microfiber Filters	Whatman	1820-070
High density polyethelye sample bottle	Nalgene	2189-0032
High Resolution Mass Spectrometer	Various	
Methanol	Sigma-Aldrich	
	Thermo Fisher	
Nitric Acid (35% w/w)	Scientific	SVCN-5-1
Polypropylene Buchner funnel	ACE Glass	12557-09
Polypropylene cenitrfuge tube and cap	BD Falcon	352096
Polypropylene Vacuum Flask (1 L)	Nalgene	DS4101-1000
Quattro Premier XE triple quadrupole mass spectrometer	Waters Corporation	
Reagent Water		
Sodium Acetate	Sigma-Aldrich	W302406
TurboVap nitrogen evaporator	Caliper Life Sciences	103198
Weak anion exchange SPE cartridge (Oasis WAX Plus)	Waters Corporation	186003519
Standard Solutions		
2,3,3,3-Tetrafluoro-2-(1,1,2,2,3,3,3-heptafluoropropoxy)propanoic acid (HFPO-DA)	Wellington	HFPO-DA
Additional targeted compound standards of interest		
Mass labeled HFPO-DA	Wellington	M2HFPO-DA
Native PFCA/PFAS Mixture (2 ug/mL)	Wellington	PFAC-MXA
Stable Isotope Labeled PFCA/PFAS Mixture (2 ug/mL)	Wellington	MPFAC-MXA

Software

Mass Profiler Professional	Agilent
Profinder	Agilent

Comments/Description

Modified with PFCs analysis kit (176001744); equivalent UPLC system is acceptible if PFAS background is checked and confirmed to be low
Mass spectrometry grade >99% pure

Mass Spectrometer should be capable of providing accurate mass to <10ppm and collecting MS/MS data. Agilent 6530 qTOF and Thermo Fisl

Can be prepared in house using concentrated nitric acid and reagent water

Equivalent triple-quadrupole or better system can be used instead, should provide high sensitivity and stability for targeted analysis
Any source determined to be PFAS free

Equivalent systems or rotary vacuum evaporator may be used instead

to be determined based on preliminary analysis and standard availability

or PFAC-MXB; or individually prepared mixture containing compounds of interest
or MPFAC-MXB; or individually prepared mixture containing compounds of interest as appropriate for Native PFASs

Or open source software packages
Or open source software packages

her Orbitrap Fusion were used in this work

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Author(s): James McCord & Mark Strynar

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Editorial comments:

Changes to be made by the author(s) regarding the manuscript:

1. Please take this opportunity to thoroughly proofread the manuscript to ensure that there are no spelling or grammar issues.

2. Please add more details to your protocol steps. There should be enough detail in each step to supplement the actions seen in the video so that viewers can easily replicate the protocol. Please ensure you answer the “how” question, i.e., how is the step performed? Alternatively, add references to published material specifying how to perform the protocol action. See examples below.

3. 1.1.2: Please specify the PFAS standard used.

Methods have been altered to reference Wellington PFC Mix A (or custom mixture)

4. 2.2.1: Please specify the PFAS standards and the range of concentrations selected.

Methods have been altered to reference Wellington PFC Mix A (or custom mixture) and specify the points for an example calibration range (0-1000ng/L)

5. 2.3.1: Please specify the stable isotope labeled PFAS standard used.

Methods have been altered to reference Wellington MPFC Mix A (or custom mixture of SIL standards)

6. 2.4.1: Please spell out GF/A. What is the pore size of the filter?

Details of the Whatman GF/A filters have been moved from materials to methods

7. 2.6.3: Please provide more details about how to analyze the samples using an LC-MS/MS method.

A sample worklist, LC, and MS settings for comparison have been added to the manuscript. The manuscript assumes basic user competence at operation of an LC-MS/MS per the manufacturer instructions.

8. 2.6.5: Please describe how to obtain the IS corrected responses.

Manuscript now refers to the Raw/ISTD response ratio when generating calibration curves.

9. 2.7.3: Please specify the LC-gradient used.

LC gradient table now properly included.

10. 2.8 and substeps: Software must have a GUI (graphical user interface) and software steps must be more explicitly explained ('click', 'select', etc.). Please add more specific details (e.g. button clicks for software actions, numerical values for settings, etc.) to your protocol steps.

In depth substeps for the use of Agilent vendor software for data analysis are now included for reference.

11. Please combine some of the shorter Protocol steps so that individual steps contain 2-3 actions and maximum of 4 sentences per step.

12. After you have made all the recommended changes to your protocol (listed above), please highlight 2.75 pages or less of the Protocol (including headings and spacing) that identifies the essential steps of the protocol for the video, i.e., the steps that should be visualized to tell the most cohesive story of the Protocol.

13. Please highlight complete sentences (not parts of sentences). Please ensure that the highlighted part of the step includes at least one action that is written in imperative tense.

14. Please include all relevant details that are required to perform the step in the highlighting. For example: If step 2.5 is highlighted for filming and the details of how to perform the step are given in steps 2.5.1 and 2.5.2, then the sub-steps where the details are provided must be highlighted.

15. Please number the figures in the sequence in which you refer to them in the manuscript text. Currently Figure 6 (line 290) is introduced before all other figures.

Figures have been relabeled as appropriate

16. Tables 1-3: These tables are missing from the current submission. Please upload each Table individually to your Editorial Manager account as an .xls or .xlsx file.

Tables are now properly included

17. Figure 2: Please describe the left, middle and right panels in more detail.

Caption updated

18. Figures 3 and 4: Please add the y-axes. Please describe what different colors represent in the figure legend of Figure 4.

Y-axes are normalized and arbitrary, but now included. Colors in figure 4 are also arbitrary, but now described in the figure caption.

19. Figure 5: Please define RT in the figure legend.

RT now correctly referred to as retention time in the figure legend.

Reviewers' comments:

Reviewer #1:

Manuscript Summary:

The manuscript presents a useful sample preparation and analytical procedure capable of performing quantitation of known presence of polyfluorinated organic compounds as well as semi-quantitation of unknown fluorinated compounds. The targeted quantification method appears to be similar to other analytical methods leveraging the specificity and sensitivity of the isotope-labeled internal standards. The value of the proposed method is especially revealing in its ability to semi-quantify the presence of unknown fluorinated contaminants in water. The authors explained the sampling, sample preparation, instrumental analysis procedure, and the data treatment and screening credentials. It is to this reviewer the significant advance of the proposed method to perform the identification and semi-quantification of the trace amount of polyfluorinated organic contaminants in the absence of commercial database

packages. Experimentally, readers should be able to follow the depicted procedure step-by-step to obtain an analytical output. The challenge, however, may rest upon the experience needed to use the ion chromatograph and ions fragmentation data to effectively and correctly screen the possible compounds. In general, the paper is well written, and all procedures are clearly depicted.

Major Concerns:

Again, the primary challenge for an experimentalist to follow the proposed method is the lack of experience needed to use the ion chromatograph and ions fragmentation data to effectively and correctly screen the possible compounds. This paper uses several conceptual diagrams to explain the steps and credentials for a valid data interpretation. Providing an example, step by step, with a real sample and analysis would significantly enhance the success rate of the knowledge dissemination.

Unfortunately, the validation of novel compound discovery is currently outside the range of novice use of mass spectrometry. An example data file and results are included in the supplemental information, and references are made to manuscripts detailing the confirmation of structures using mass spectrometry, which is beyond the scope of this manuscript.

Minor Concerns:

Text description of Figure 6 is missing.

In-text reference to Figure 6 was in the methods description. Figures have been renumbered in-text.

Reviewer #2:

Manuscript Summary:

General comments.

In the paper Identification of per-/polyfluorinated chemical species with a combined targeted and non-targeted screening high-resolution mass spectrometry workflow, authors describe a method able to detect fluorinated and others untargeted compounds by coupled solid phase extraction with hight resolution mass spectrometry HPLC.

Report is good discussed, unfortunately, tables are mistake, and some errors were reported.

Major Concerns:

Authors report only Internal Standards. Why authors not use standard in order to monitor recovery?

PFAS standards were used to construct spike blanks for recovery assessment in step 1.1.2. Recovery assessment does not affect quantitation for IS spiked, extracted calibration curves and is relegated quantitative method development, where it can affect limit of detection and method reporting limits.

Where are Tables?

Please add a paragraph concerning to material and methods. Moreover, please add the instrument used.

JoVE materials table is included, which indicates instrumental platforms and a summary of reagent materials

Line 5 line 81

Please, add some recent references concerning to Online Solid-Phase Extraction and perfluorinated compounds

Page 2 line 124.

Have authors verified absence of fluorinated substances from Laboratory deionized? Often fluorinated compounds were detected in DI water.

Our laboratory water has been validated as being PFAS free, text to this effect has been added to the manuscript.

Page 3 line 144.

Are authors sure that nitric acid does not cause the oxidation of some untargeted compounds?

Nitric acid oxidation of traditional organic compounds is possible, PFAS materials are typically non-reactive under mild acidification.

Page 3 line 163

Please, report low levels used in calibration curve different from 0.

Calibration levels are to be determined by the experimentalist on the basis of desired quantitation range and the sensitivity of their instrumental platform.

Page 3 line 171

Add stable isotope labeled PFAS standard to approximate the midpoint of the LC-MS standard curve (e.g. 100 ng/L).

In line 163 authors reported that calibration curve ranged from 0 to 100 ng/L.

In line 163 the example calibration range was from zero to **1000 ng/L**. 100 ng/L undershoots the midpoint of that calibration curve but was a hypothetical example.

Page 4 line 177.

For analyses concerning to monitoring activities, samples must not be filtered. Moreover, authors not reported filter diameter. On another hand, by using SPE techniques, filtration can be avoided.

Pre-filtration is used to prevent clogging of the SPE cartridge system for dirty surface water samples, matched extracted calibration curves eliminate filtration related biases in quantitation. The filter diameter is not an analytically relevant parameter as it only affects the backpressure of filtration. However, the filter used is listed in the JoVE Materials table.

Minor Concerns:

have authors used column trap to avoid PFAS contamination from system?

Modifications to the LC system, including the PFAS hold-up column, are mentioned in the JoVE Materials table.

Reviewer #3:

Manuscript Summary:

The manuscript "Identification of per-/polyfluorinated chemical species with a combined targeted and non-targeted screening high-resolution mass spectrometry workflow" presents a protocol, as the title indicates, for targeted and non-targeted identification of per-/polyfluorinated compounds (PFASs) using high-resolution mass spectrometry. The article responds to the need of having forward-looking methodologies that can detect emerging and unexpected compounds (in this case PFASs and related compounds), monitor these species over time, and resolve details of their chemical structure.

Minor Concerns:

The manuscript is clear, well organized and in general very informative. Even if objectives are not clearly presented, they can be easily deduced from text. The references used are updated and they support properly the introduction and discussion sections. The number of figures and tables is correct. My only comment is related to these figures. They are not presented in ascending order (first figures 6 and 7 are cited and later figures 1, 2 , 3…). Additional information about them would also be of interest. For instance in figure 1, which are the perfluorinated ether standards used for obtaining this total ion chromatogram (TIC), or a small description of the fluorochemical production site which upstream and downstream surface waters were collected and extracted to obtain the overlaid TIC presented in this figure. This is a very good paper and I only suggest MINOR changes before it can be accepted for publication in the Journal of Visualized Experiments.

Figure references have been updated as appropriate. The Figure 1 TIC is a wide-scan total ion chromatogram and is not constructed from standards. Further description of the fluorochemical production site is not included due to privacy concerns, but references are provided in

Reviewer #4:

Manuscript Summary:

This work describes a method to extract, identify, and potentially quantify PFAS found in environmental water samples. The authors describe a method that allows for the detection of PFASs not included in the standard EPA 537 method. This work is an excellent candidate for visual representation given the current climate surrounding PFAS in the United States. These methods will help researchers to implement these methods when typical journal articles do not provide detailed information.

Major Concerns:

None

Minor Concerns:

- Providing a couple of lists or example lists of a full set of method blanks, calibration curve, etc might be useful for researchers new to LC-MS/MS.

An example worklist has been added as a table.

- Add to the discussion of the types of PFAS extracted in this method - talk about specifics of the types of compounds that are more likely to be extracted

A brief discussion of these points occurs in the introduction (Lines 83-91 and 110-113)

- Highlight potential places in the method where PFAS contamination is particularly likely

A few key points of contamination are now included as notes

Supplemental Table 1

Click here to access/download;Supplemental File (Figures, Permissions, etc.);Supplemental Table 1 - Chemical Feature

Compound	MS1 Comp	Mass	Mass Defect	Retention Time	Downstrea	Downstrea	Downstrea
1	(878.8772,	439.942	0.942	0.83000004	2.55E+07	2.57E+07	2.68E+07
2	(340.9372,	341.9443	0.9443	0.408	1596613	1692183	1625315
3	(194.9263,	195.9338	0.9338	0.30999997	1271930	1292899	1251504
4	(623.3441,	312.1761	0.1761	5.5250006	528863	538280	545181
5	(340.9369,	341.9443	0.9443	0.453	593904	679026	664279
6	(843.4599,	422.234	0.234	5.8310003	403345	414017	413813
7	(651.3749,	326.1918	0.1918	5.806	459043	478143	464575
8	(526.9574,	263.983	0.983	2.1119998	475443	479333	518468
9	(419.193, 3	210.1021	0.1021	2.281	253913	250811	260133
10	(595.3115,	298.1606	0.1606	5.2249994	213000	220821	220758
11	(681.2958,	682.3029	0.3029	9.953001	271625	254878	136234
12	(412.9663,	413.9738	0.9738	4.3850007	76875	85600	82860
13	(218.9862,	219.9933	0.9933	2.1119998	188550	186792	197391
14	(283.2641,	284.2714	0.2714	11.156999	51958	50818	46428
15	(799.2396,	400.1215	0.1215	3.532	98217	96656	101481
16	(255.1601,	256.1675	0.1675	2.6499999	158488	152557	162069
17	(403.3063,	404.3132	0.3132	11.488999	94109	85147	65383
18	(437.4359,	438.4432	0.4432	13.880999	1	1	1
19	(498.9303,	499.9367	0.9367	4.82	91056	94683	98384
20	(626.9553,	313.9803	0.9803	3.129	1	1	1
21	(132.0568,	133.064	0.064	1.898	47731	48532	48532
22	(465.467, 4	466.4743	0.4743	14.497999	1	1	1
23	(339.2, 169	340.2071	0.2071	6.1619997	81247	85035	84942
24	(506.9234,	507.9302	0.9302	1.691	123401	123966	73094
25	(362.9698,	363.9768	0.9768	3.843	86928	86158	87357
26	(409.4047,	410.4119	0.4119	13.423002	1	1	1
27	(426.9569,	213.9871	0.9871	1.014	101023	104576	106834
28	(265.1478,	266.155	0.155	5.0779996	59117	60254	61373
29	(319.1551,	320.1618	0.1618	2.4459999	27290	30231	63211
30	(398.938, 2	399.9448	0.9448	3.884	126319	129615	128848
31	(213.9635,	214.9707	0.9707	0.697	1	1	1
32	(253.1443,	254.1516	0.1516	2.8150003	20614	53261	22053
33	(459.2026,	460.2101	0.2101	4.8440003	36944	37360	39104
34	(183.1024,	184.1098	0.1098	1.4230001	1	1	1
35	(239.0275,	120.0407	0.0407	0.37599996	37544	57700	69118
36	(325.1845,	326.1912	0.1912	5.8009996	68204	558617	68368
37	(311.1689,	312.1761	0.1761	5.5180006	56622	60039	528838
38	(498.9295,	499.9366	0.9366	4.7119994	21383	21966	23333
39	(370.0537,	371.0617	0.0617	3.0410001	1	1	1
40	(268.9826,	269.9902	0.9902	3.132	1	1	1
41	(447.1346,	448.1428	0.1428	4.341	42961	42373	43064
42	(303.1603,	304.1667	0.1667	3.966	1	1	1
43	(293.1788,	294.187	0.187	5.6959996	14922	14794	15051
44	(420.249, 6	421.257	0.257	4.3790007	30841	30608	31606
45	(287.1327,	288.1403	0.1403	3.5680003	16539	15630	16113

46	(622.9279,	623.9349	0.9349	2.857	4.67E+07	4.74E+07	4.99E+07
47	(296.946, 1	595.9069	0.9069	1.047	2.41E+07	2.44E+07	2.47E+07
48	(754.9113,	755.9186	0.9186	3.993	1.10E+07	1.14E+07	1.19E+07
49	(658.9419,	659.9491	0.9491	3.301	8623775	8758179	9140374
50	(226.9789,	113.9925	0.9925	0.41800004	1	8163230	8297380
51	(926.8723,	463.9386	0.9386	3.9220004	6893698	7000858	7181519
52	(644.9111,	645.9188	0.9188	2.8729997	6629912	6830686	7102419
53	(611.9272,	612.9344	0.9344	1.045	6427296	6449398	6493521
54	(310.9611,	311.9686	0.9686	2.8619998	6586903	6762269	7037287
55	(926.8723,	463.9391	0.9391	4.035	4468358	4562471	4705168
56	(886.8955,	887.9034	0.9034	4.7309995	1920297	1980926	2076225
57	(396.9402,	397.9477	0.9477	3.852	1943535	1981885	2031345
58	(474.9521,	475.9597	0.9597	3.4030004	1650174	1681813	1686338
59	(346.9438,	347.9512	0.9512	2.507	1795315	1784648	1832358
60	(266.9574,	535.9269	0.9269	2.045	1456760	1473889	1521578
61	(680.9254,	681.9324	0.9324	3.301	1333354	1374786	1432478
62	(474.9524,	475.9598	0.9598	3.2280002	1341068	1355810	1398775
63	(134.9873,	135.9947	0.9947	0.57600003	1406384	1406477	1471601
64	(150.9825,	151.9897	0.9897	2.8699996	1306963	1341051	914935
65	(184.9843,	185.9917	0.9917	5.028	1339702	1345596	1380999
66	(326.972, 3	163.9889	0.9889	0.499	1197042	1190458	1234381
67	(168.9893,	169.9965	0.9965	3.301	1371779	1353170	1290535
68	(298.9446,	299.952	0.952	1.047	1172497	1188579	1208202
69	(250.9764,	251.984	0.984	3.6630006	1134946	1144691	1157293
70	(284.9781,	285.9853	0.9853	3.2970004	1094947	1103242	1113333
71	(364.9348,	365.9421	0.9421	3.468	1074486	1086525	1108636
72	(978.8607,	979.8688	0.8688	2.865	803177	856945	893983
73	(269.9588,	541.9316	0.9316	2.1380002	820848	815566	870332
74	(776.8949,	777.9031	0.9031	3.993	771929	874099	904415
75	(590.9386,	295.9738	0.9738	2.7630002	619486	626790	672007
76	(266.9568,	535.9267	0.9267	1.964	681725	669121	733315
77	(790.841, 2	791.8486	0.8486	1.1619998	966832	701058	602465
78	(1241.8911	1242.899	0.8986	1.045	683444	687338	730577
79	(272.9487,	273.9559	0.9559	0.35599998	766610	790304	788818
80	(261.9462,	525.9063	0.9063	0.992	740591	735992	735187
81	(380.9438,	381.9509	0.9509	0.591	707519	730975	760569
82	(328.9679,	329.9755	0.9755	3.301	680824	720039	740594
83	(943.8996,	944.9076	0.9076	3.9220004	421986	428906	428137
84	(476.9283,	477.9354	0.9354	4.5429997	475877	480076	496652
85	(926.91, 57	927.9177	0.9177	1.0430001	329462	330632	411828
86	(458.9551,	229.9818	0.9818	0.97599995	309185	314678	356458
87	(616.8835,	617.8908	0.8908	1.12	338379	387608	382350
88	(234.981, 8	235.9887	0.9887	2.3419998	475582	469013	505949
89	(342.9342,	343.9414	0.9414	0.77799994	412054	409749	431806
90	(212.093, 8	213.1003	0.1003	0.7020001	395438	401350	418951
91	(690.9324,	345.9701	0.9701	3.6630006	244932	253912	273575
92	(492.9233,	493.9307	0.9307	4.5159993	383435	386893	396699

93	(592.8657,	593.8726	0.8726	0.54300004	409413	297123	280558
94	(526.9285,	527.9358	0.9358	3.5149999	400418	390514	407403
95	(908.878, 8	909.8861	0.8861	4.7309995	315303	329944	339536
96	(396.9411,	397.9484	0.9484	3.25	335494	330211	364166
97	(854.9375,	427.9719	0.9719	4.5120006	287088	287504	308930
98	(943.8994,	944.9063	0.9063	4.035	283675	290881	294321
99	(862.8746,	863.8818	0.8818	4.029	285172	301116	307667
100	(246.9508,	247.9579	0.9579	0.5960001	307146	326670	356057
101	(972.8437,	973.851	0.851	2.8619998	282094	290454	297668
102	(506.9216,	507.9284	0.9284	1.0720001	261396	244891	277373
103	(412.9358,	413.9428	0.9428	3.492	290960	294002	302953
104	(558.9493,	559.9562	0.9562	2.347	284054	284738	303561
105	(564.9041,	565.9121	0.9121	1.111	327944	336666	317030
106	(494.9523,	991.9217	0.9217	5.029	241902	254362	269941
107	(360.9736,	361.9812	0.9812	3.6810002	274793	283972	288849
108	(476.9282,	477.9354	0.9354	4.6559997	237470	238864	243948
109	(640.9359,	641.9431	0.9431	3.258	242563	239257	219839
110	(316.9677,	317.975	0.975	4.4179997	261060	263991	266419
111	(562.9114,	563.919	0.919	2.1340003	188810	186736	211174
112	(854.9373,	427.9734	0.9734	3.881	231327	238411	241203
113	(606.9341,	607.9414	0.9414	2.7800002	232207	223845	237900
114	(278.9709,	279.9784	0.9784	2.3419998	220939	220121	241064
115	(360.9664,	361.9737	0.9737	0.5630001	240197	239714	254057
116	(350.9696,	351.9771	0.9771	4.5100007	231188	231920	236622
117	(281.961, 8	565.936	0.936	2.0639997	190525	193474	199582
118	(442.9443,	443.9504	0.9504	4.749	189338	199774	222789
119	(722.9221,	361.9649	0.9649	3.881	164834	167752	172991
120	(248.961, 4	249.9684	0.9684	0.421	176196	187958	199304
121	(1032.8811	1033.889	0.8888	3.2990003	173047	183178	187320
122	(184.9842,	185.9918	0.9918	0.974	203613	198207	226939
123	(376.9346,	377.942	0.942	0.74499995	197390	194964	210047
124	(672.9075,	673.9152	0.9152	3.322	161147	176817	164487
125	(948.8549,	949.862	0.862	4.029	157523	158261	157899
126	(1556.8715	1557.879	0.8788	1.047	144399	145097	164085
127	(422.9399,	423.9469	0.9469	0.908	169989	175142	183897
128	(458.9375,	459.9446	0.9446	4.3649993	153613	154475	157301
129	(672.9078,	673.9148	0.9148	3.426	112643	114877	148771
130	(370.9555,	371.9634	0.9634	3.2970004	150870	156255	155342
131	(895.9042,	896.9115	0.9115	0.77400005	120202	119409	142772
132	(360.9434,	361.9508	0.9508	0.967	134967	135959	145462
133	(442.9261,	443.9332	0.9332	4.2519994	141795	140772	139765
134	(426.9652,	427.9725	0.9725	3.9509997	127666	128252	134681
135	(1176.8351	1177.844	0.8435	4.0789995	102592	99596	108794
136	(294.9819,	295.99	0.99	2.316	139777	140503	145384
137	(460.9167,	461.9244	0.9244	0.77	168838	173054	177336
138	(948.8543,	949.8623	0.8623	3.9179997	107132	109924	105520
139	(402.9631,	403.9706	0.9706	0.61399996	157244	159666	193364

140	(380.9052,	381.9126	0.9126	3.5919995	145358	138102	142509
141	(506.9217,	507.9293	0.9293	0.98599994	98799	106023	102028
142	(1246.8452	1247.855	0.8549	1.066	119891	118185	115210
143	(314.9381,	315.9449	0.9449	2.712	129183	128285	132512
144	(390.9336,	391.9417	0.9417	0.514	92205	102372	102120
145	(1018.8791	509.943	0.943	5.223	70966	75034	80478
146	(310.9433,	311.9502	0.9502	2.5739996	135773	134045	138538
147	(285.2071,	286.2144	0.2144	4.5390005	110313	106762	106343
148	(456.9248,	457.9321	0.9321	0.9610001	102021	105231	106682
149	(900.86, 63	901.868	0.868	0.776	107438	112839	116337
150	(1176.8348	1177.843	0.843	3.993	91095	103823	107985
151	(346.9442,	347.9511	0.9511	2.7910001	115398	114390	115855
152	(346.9439,	347.9515	0.9515	2.667	113798	112958	119594
153	(822.916, 3	411.9615	0.9615	4.4119997	82090	83595	91633
154	(174.9504,	175.9576	0.9576	0.313	104823	106107	110332
155	(358.9447,	359.9517	0.9517	2.5839999	101675	105680	105036
156	(281.9629,	565.9369	0.9369	1.9750001	73274	78999	89089
157	(293.0811,	294.0844	0.0844	0.512	18987	8930	1
158	(854.9171,	427.9587	0.9587	4.5849996	87333	1	1
159	(462.9322,	463.9397	0.9397	4.389	85995	92407	101617
160	(402.942, 2	403.9499	0.9499	0.55499995	90015	102469	90380
161	(246.9507,	247.9579	0.9579	0.923	81933	84727	85791
162	(342.9356,	343.9428	0.9428	0.40299994	81120	87338	88978
163	(548.9687,	549.9751	0.9751	4.7729993	59596	57365	68925
164	(388.9381,	389.9456	0.9456	0.52299994	73800	82465	82369
165	(310.9771,	311.9845	0.9845	2.3250003	87472	85734	91698
166	(348.9414,	349.949	0.949	2.507	86032	83979	87624
167	(718.9433,	719.9504	0.9504	3.7280002	69601	74367	77863
168	(772.919, 1	773.9261	0.9261	4.154	71211	75508	72951
169	(180.968, 8	363.9515	0.9515	0.77799994	80729	72109	88060
170	(343.1408,	344.1481	0.1481	2.1790001	59984	57287	60000
171	(574.9461,	575.9538	0.9538	3.8629994	67660	66862	74153
172	(528.9545,	529.9615	0.9615	5.243	71869	75460	77130
173	(640.9348,	641.9426	0.9426	2.7940001	50694	52870	57166
174	(343.1427,	344.1486	0.1486	2.0739996	56017	58455	59802
175	(412.9358,	413.943	0.943	3.6810002	67704	68647	70724
176	(482.9766,	483.9821	0.9821	4.146	44302	48276	56663
177	(821.9021,	822.9107	0.9107	1.0720001	69364	72459	57882
178	(481.9769,	482.9844	0.9844	2.7059999	61752	60337	62478
179	(324.9423,	325.95	0.95	0.41500002	63345	69511	65074
180	(525.9488,	526.9561	0.9561	2.229	65699	64404	68354
181	(458.9571,	459.9646	0.9646	3.7280002	60938	69229	62513
182	(328.9677,	329.9746	0.9746	4.2459993	66561	67160	69861
183	(428.9613,	429.9682	0.9682	4.563	60914	61741	65361
184	(508.9523,	509.9592	0.9592	2.0989997	64093	64444	59874
185	(343.1427,	344.148	0.148	1.999	47812	48399	49522
186	(426.9324,	427.9392	0.9392	4.1770005	53884	54257	55288

187	(481.9764,	482.9838	0.9838	2.5090003	53579	53960	55335
188	(268.9651,	539.9362	0.9362	2.1520002	52932	49766	56730
189	(345.1378,	346.1453	0.1453	4.4010005	43269	44253	46319
190	(532.9878,	533.9951	0.9951	4.5100007	46849	40818	52196
191	(366.9319,	367.9389	0.9389	3.466	53453	51572	52694
192	(458.9573,	459.9638	0.9638	3.581	49592	49615	47250
193	(718.9429,	719.9506	0.9506	3.8060002	39141	38946	39283
194	(438.9845,	439.9916	0.9916	3.416	42220	41634	41879
195	(283.1209,	284.128	0.128	2.556	34576	39221	42156
196	(772.9196,	773.9265	0.9265	3.9739995	36415	36110	36760
197	(614.9601,	615.9671	0.9671	5.2200003	34014	34926	39021
198	(456.9557,	457.963	0.963	4.625	36778	37931	39319
199	(574.9455,	575.9538	0.9538	3.959	31954	28918	35940
200	(628.9159,	629.923	0.923	2.7830002	33422	34801	32944
201	(662.9183,	663.9251	0.9251	3.256	29661	32564	29975
202	(383.9789,	384.9866	0.9866	0.952	29484	29293	31047

Downstrea	Downstrea	Upstream_	Upstream_	Upstream_	Upstream_	Upstream_	Downstrea	Upstream_
2.65E+07	2.64E+07	520539	477813	482186	563236	545931	2.62E+07	517941
1635378	1770845	25630	25638	21399	25278	28722	1.66E+06	25333.4
1279007	1293478	783815	793149	663133	788996	797295	1.28E+06	765277.6
561533	563580	641697	635385	700347	667259	687477	5.47E+05	666433
697731	787690	25630	25638	21399	25278	28722	6.85E+05	25333.4
393646	426460	519217	546346	531451	531267	574499	4.10E+05	540556
468437	486001	468634	554489	544608	544737	553989	4.71E+05	533291.4
498432	511659	108693	110716	114212	112410	112848	4.97E+05	111775.8
232805	239665	412191	370165	405546	443862	387065	2.47E+05	403765.8
233461	235513	280048	291946	309761	317308	312734	2.25E+05	302359.4
113313	81201	230684	201255	127562	147672	156114	1.71E+05	172657.4
144699	86873	137692	122765	269097	224343	142121	9.54E+04	179203.6
199418	209238	47174	48072	47351	47363	51331	1.96E+05	48258.2
64226	49624	55444	53804	154204	79155	51524	5.26E+04	78826.2
102645	103679	150156	148443	149044	156468	160657	1.01E+05	152953.6
160927	163733	144741	150241	148000	151870	163080	1.60E+05	151586.4
68818	64684	132627	106875	107420	87591	78825	7.56E+04	102667.6
1	1	162799	170216	170619	163538	164789	1.00E+00	166392.2
97157	100501	120964	122226	111786	129108	126261	9.64E+04	122069
1	1	129331	131243	133452	134456	132827	1.00E+00	132261.8
46958	51970	128050	125870	126906	131751	132781	4.87E+04	129071.6
1	1	154931	169956	166543	157195	164548	1.00E+00	162634.6
88929	86099	94132	97381	103881	100266	101874	8.53E+04	99506.8
66014	45312	23530	23100	18201	25142	26267	8.64E+04	23248
86443	90980	108935	109938	113534	115383	115603	8.76E+04	112678.6
1	1	125131	128664	120300	111017	112614	1.00E+00	119545.2
112293	109650	20898	21063	22384	21799	20833	1.07E+05	21395.4
62601	65026	80440	84174	93299	87377	92648	6.17E+04	87587.6
32026	65023	154457	156889	121767	160090	162525	4.36E+04	151145.6
130243	133036	83350	85293	87653	91102	89275	1.30E+05	87334.6
1	1	85595	84806	81286	84071	86718	1.00E+00	84495.2
18349	21971	65709	63477	63288	66144	66601	2.72E+04	65043.8
47698	41579	63499	64152	66008	65031	66683	4.05E+04	65074.6
1	1	68205	75518	73682	75788	80265	1.00E+00	74691.6
66915	44120	103649	95007	101643	104416	92604	5.51E+04	99463.8
545593	565607	66164	633352	16790	632842	664784	3.61E+05	402786.4
546515	547050	623697	62269	679835	648105	667694	3.48E+05	536320
22165	24705	61735	63881	36875	64151	67629	2.27E+04	58854.2
1	1	47619	50098	49481	50724	53093	1.00E+00	50203
1	1	51673	53516	49935	56274	56217	1.00E+00	53523
45638	43465	37332	37875	37663	38812	40544	4.35E+04	38445.2
1	1	33918	40196	34827	40624	47004	1.00E+00	39313.8
16139	16629	27562	29245	38762	30397	29148	1.55E+04	31022.8
33393	31835	29319	31238	31278	29093	32193	3.17E+04	30624.2
16241	14759	32121	33169	30334	33117	34898	1.59E+04	32727.8

4.97E+07	4.95E+07	1	1	1	1	1	4.86E+07	1
2.42E+07	2.45E+07	1	1	1	1	1	2.44E+07	1
1.17E+07	1.17E+07	1	1	1	1	1	1.15E+07	1
9076449	8929166	1	1	1	1	1	8.91E+06	1
1	8388609	1	1	1	1	1	4.97E+06	1
7179518	7207891	10679	1	10957	1	1	7.09E+06	4327.8
7163942	7272219	1	1	1	1	1	7.00E+06	1
6501250	6561621	1	1	1	1	1	6.49E+06	1
7002966	7118705	1	1	1	1	1	6.90E+06	1
4676024	4712333	10679	1	10957	1	1	4.62E+06	4327.8
2055930	2090638	1	1	1	1	1	2.02E+06	1
1985861	2027813	1	1	1	1	1	1.99E+06	1
1681405	1731695	1	1	1	1	1	1.69E+06	1
1806335	1849481	1	1	1	1	1	1.81E+06	1
1534029	1582304	1	1	1	1	1	1.51E+06	1
1432511	1429296	1	1	1	1	1	1.40E+06	1
1395651	1415624	1	1	1	1	1	1.38E+06	1
1390432	1467451	1	1	1	1	1	1.43E+06	1
1397163	1442534	1	1	1	1	1	1.28E+06	1
1378656	1375585	1	1	1	1	1	1.36E+06	1
1140203	1220524	1	1	1	1	1	1.20E+06	1
1359401	1381333	1	1	1	1	1	1.35E+06	1
1191697	1178561	1	1	1	1	1	1.19E+06	1
1152165	1161509	1	1	1	1	1	1.15E+06	1
1127956	1152956	1	1	1	1	1	1.12E+06	1
1103347	1120945	1	1	1	1	1	1.10E+06	1
914403	940643	1	1	1	1	1	8.82E+05	1
868370	875733	1	1	1	1	1	8.50E+05	1
857528	854204	1	1	1	1	1	8.52E+05	1
663788	691132	1	1	1	1	1	6.55E+05	1
726536	705331	1	1	1	1	1	7.03E+05	1
534951	350256	1	1	1	1	1	6.31E+05	1
727241	743330	1	1	1	1	1	7.14E+05	1
791359	811680	1	1	1	1	1	7.90E+05	1
734974	758066	1	1	1	1	1	7.41E+05	1
759622	775234	1	1	1	1	1	7.47E+05	1
739873	732108	1	1	1	1	1	7.23E+05	1
439013	446724	1	1	1	1	1	4.33E+05	1
491295	503496	1	1	1	1	1	4.89E+05	1
369283	333988	1	1	1	1	1	3.55E+05	1
342955	347098	1	1	1	1	1	3.34E+05	1
420359	378974	1	1	1	1	1	3.82E+05	1
486502	474168	1	1	1	1	1	4.82E+05	1
431140	448995	1	1	1	1	1	4.27E+05	1
432929	435007	1	1	1	1	1	4.17E+05	1
263757	273169	1	1	1	1	1	2.62E+05	1
399396	394653	1	1	1	1	1	3.92E+05	1

256156	154465	1	1	1	1	1	2.80E+05	1
414519	418181	1	1	1	1	1	4.06E+05	1
353037	361205	1	1	1	1	1	3.40E+05	1
367866	362368	1	1	1	1	1	3.52E+05	1
305989	305944	1	1	1	1	1	2.99E+05	1
300389	303642	1	1	1	1	1	2.95E+05	1
339590	339800	1	1	1	1	1	3.15E+05	1
358044	308495	1	1	1	1	1	3.31E+05	1
301081	298546	1	1	1	1	1	2.94E+05	1
260443	261185	1	1	1	1	1	2.61E+05	1
299352	296468	1	1	1	1	1	2.97E+05	1
292661	295512	1	1	1	1	1	2.92E+05	1
333123	340218	1	1	1	1	1	3.31E+05	1
269331	268678	1	1	1	1	1	2.61E+05	1
286781	290572	1	1	1	1	1	2.85E+05	1
263790	243315	1	1	1	1	1	2.45E+05	1
235430	220491	1	1	1	1	1	2.32E+05	1
266263	269568	1	1	1	1	1	2.65E+05	1
205467	213066	1	1	1	1	1	2.01E+05	1
246100	253109	1	1	1	1	1	2.42E+05	1
244144	229551	1	1	1	1	1	2.34E+05	1
241739	231668	1	1	1	1	1	2.31E+05	1
247077	243219	1	1	1	1	1	2.45E+05	1
242488	245803	1	1	1	1	1	2.38E+05	1
203963	192915	1	1	1	1	1	1.96E+05	1
214697	211025	1	1	1	1	1	2.08E+05	1
170212	184806	1	1	1	1	1	1.72E+05	1
196918	225382	1	1	1	1	1	1.97E+05	1
192482	194130	1	1	1	1	1	1.86E+05	1
212928	208022	1	1	1	1	1	2.10E+05	1
208528	210524	1	1	1	1	1	2.04E+05	1
179994	194313	1	1	1	1	1	1.75E+05	1
164984	168290	1	1	1	1	1	1.61E+05	1
160343	156591	1	1	1	1	1	1.54E+05	1
186054	186170	1	1	1	1	1	1.80E+05	1
161739	160264	1	1	1	1	1	1.57E+05	1
157028	125620	1	1	1	1	1	1.32E+05	1
161926	166113	1	1	1	1	1	1.58E+05	1
136333	123111	1	1	1	1	1	1.28E+05	1
146186	150891	1	1	1	1	1	1.43E+05	1
144465	146519	1	1	1	1	1	1.43E+05	1
138486	138917	1	1	1	1	1	1.34E+05	1
117470	120301	1	1	1	1	1	1.10E+05	1
145711	145291	1	1	1	1	1	1.43E+05	1
183440	184293	1	1	1	1	1	1.77E+05	1
111329	117516	1	1	1	1	1	1.10E+05	1
182137	208178	1	1	1	1	1	1.80E+05	1

147178	141166	1	1	1	1	1	1.43E+05	1
110351	131128	1	1	1	1	1	1.10E+05	1
116917	121860	1	1	1	1	1	1.18E+05	1
135625	132833	1	1	1	1	1	1.32E+05	1
107103	122971	1	1	1	1	1	1.05E+05	1
76869	79614	1	1	1	1	1	7.66E+04	1
120734	121182	1	1	1	1	1	1.30E+05	1
107246	105998	1	1	1	1	1	1.07E+05	1
111005	121062	1	1	1	1	1	1.09E+05	1
122904	133389	1	1	1	1	1	1.19E+05	1
105251	103183	1	1	1	1	1	1.02E+05	1
120838	121535	1	1	1	1	1	1.18E+05	1
119748	117463	1	1	1	1	1	1.17E+05	1
90500	85683	1	1	1	1	1	8.67E+04	1
105247	117581	1	1	1	1	1	1.09E+05	1
108288	108089	1	1	1	1	1	1.06E+05	1
82587	95383	1	1	1	1	1	8.39E+04	1
1	14432	67737	79701	64095	74908	1	8.47E+03	57288.4
1	107984	1	1	1	1	1	3.91E+04	1
93562	92984	10679	1	10957	1	1	9.33E+04	4327.8
106667	101413	1	1	1	1	1	9.82E+04	1
87065	90821	1	1	1	1	1	8.61E+04	1
83222	91511	1	1	1	1	1	8.64E+04	1
99603	90348	1	1	1	1	1	7.52E+04	1
86165	89955	1	1	1	1	1	8.30E+04	1
92030	87581	1	1	1	1	1	8.89E+04	1
84591	88621	1	1	1	1	1	8.62E+04	1
80519	81210	1	1	1	1	1	7.67E+04	1
74447	72536	1	1	1	1	1	7.33E+04	1
84869	76286	1	1	1	1	1	8.04E+04	1
68617	68615	55502	1	72943	1	1	6.29E+04	25689.6
72007	76949	1	1	1	1	1	7.15E+04	1
76062	74553	1	1	1	1	1	7.50E+04	1
67727	54774	1	1	1	1	1	5.66E+04	1
63763	60486	55502	1	75106	1	1	5.97E+04	26122.2
71931	76762	1	1	1	1	1	7.12E+04	1
59612	58282	1	1	1	1	1	5.34E+04	1
75770	56694	1	1	1	1	1	6.64E+04	1
64063	63867	1	1	1	1	1	6.25E+04	1
67519	68871	1	1	1	1	1	6.69E+04	1
69652	67508	1	1	1	1	1	6.71E+04	1
66140	64143	1	1	1	1	1	6.46E+04	1
69199	66411	1	1	1	1	1	6.78E+04	1
64088	59953	1	1	1	1	1	6.24E+04	1
65335	61910	1	1	1	1	1	6.31E+04	1
52457	54648	55502	1	75106	1	1	5.06E+04	26122.2
56289	58150	1	1	1	1	1	5.56E+04	1

54947	55238	1	1	1	1	1	5.46E+04	1
57526	53188	1	1	1	1	1	5.40E+04	1
44218	44563	1	1	1	1	1	4.45E+04	1
46517	45184	1	1	1	1	1	4.63E+04	1
52369	53867	1	1	1	1	1	5.28E+04	1
49952	47878	1	1	1	1	1	4.89E+04	1
39393	38627	1	1	1	1	1	3.91E+04	1
41331	44116	1	1	1	1	1	4.22E+04	1
33842	39035	1	1	1	1	1	3.78E+04	1
38322	37707	1	1	1	1	1	3.71E+04	1
38183	34053	1	1	1	1	1	3.60E+04	1
38010	37605	1	1	1	1	1	3.79E+04	1
36127	34623	1	1	1	1	1	3.35E+04	1
36977	33243	1	1	1	1	1	3.43E+04	1
30127	32390	1	1	1	1	1	3.09E+04	1
33812	31443	1	1	1	1	1	3.10E+04	1

log2 Fold Change	p-val (unadjusted)	Signif? (Bonferroni)
5.66	9.7646E-14	TRUE
6.04	1.77084E-11	TRUE
0.74	5.75955E-08	TRUE
-0.28	3.21691E-05	TRUE
4.76	2.66099E-08	TRUE
-0.40	2.37831E-06	TRUE
-0.18	0.006505222	FALSE
2.15	6.73064E-11	TRUE
-0.71	2.62598E-06	TRUE
-0.43	1.30328E-05	TRUE
-0.01	0.978260126	FALSE
-0.91	0.027738455	FALSE
2.02	4.0186E-10	TRUE
-0.58	0.22060371	FALSE
-0.61	5.87386E-08	TRUE
0.07	0.061741543	FALSE
-0.44	0.039962115	FALSE
-17.34	1.17789E-13	TRUE
-0.34	6.11101E-05	TRUE
-17.01	5.11421E-15	TRUE
-1.40	2.83793E-11	TRUE
-17.31	9.63865E-12	TRUE
-0.22	0.000144639	TRUE
1.89	0.00422241	TRUE
-0.36	3.21582E-07	TRUE
-16.87	5.05815E-10	TRUE
2.32	9.13732E-11	TRUE
-0.51	1.03803E-05	TRUE
-1.79	1.19639E-05	TRUE
0.57	9.43295E-09	TRUE
-16.37	2.10173E-13	TRUE
-1.26	0.000429434	TRUE
-0.68	2.20225E-06	TRUE
-16.19	2.36978E-10	TRUE
-0.85	0.000157349	TRUE
-0.16	0.832694016	FALSE
-0.62	0.293496372	FALSE
-1.37	0.000199023	TRUE
-15.62	1.08056E-11	TRUE
-15.71	9.6012E-11	TRUE
0.18	0.000242894	TRUE
-15.26	1.67492E-07	TRUE
-1.00	5.85779E-05	TRUE
0.05	0.221514972	FALSE
-1.05	2.93155E-08	TRUE

25.54	1.5697E-12	TRUE
24.54	2.57654E-16	TRUE
23.46	1.10578E-12	TRUE
23.09	2.11148E-13	TRUE
22.24	0.039992361	FALSE
10.68	3.82694E-14	TRUE
22.74	7.10267E-12	TRUE
22.63	3.02789E-17	TRUE
22.72	1.9255E-12	TRUE
10.06	1.40763E-13	TRUE
20.95	4.5986E-12	TRUE
20.93	2.20115E-14	TRUE
20.69	1.44498E-14	TRUE
20.79	4.02211E-15	TRUE
20.53	2.53264E-12	TRUE
20.42	1.97103E-12	TRUE
20.40	1.29149E-13	TRUE
20.45	4.54489E-13	TRUE
20.29	8.31692E-07	TRUE
20.38	3.51627E-15	TRUE
20.19	1.22904E-12	TRUE
20.37	4.18721E-13	TRUE
20.18	5.62687E-16	TRUE
20.13	8.96699E-17	TRUE
20.09	5.4491E-14	TRUE
20.07	1.09144E-14	TRUE
19.75	3.24761E-10	TRUE
19.70	3.60215E-12	TRUE
19.70	2.1641E-10	TRUE
19.32	3.96035E-11	TRUE
19.42	1.04221E-11	TRUE
19.27	0.000256543	TRUE
19.45	7.79395E-12	TRUE
19.59	4.99803E-14	TRUE
19.50	1.71912E-15	TRUE
19.51	5.48272E-12	TRUE
19.46	3.44606E-12	TRUE
18.72	1.25774E-13	TRUE
18.90	1.60459E-13	TRUE
18.44	1.79923E-08	TRUE
18.35	4.09258E-10	TRUE
18.54	2.04747E-09	TRUE
18.88	1.33094E-12	TRUE
18.70	7.51762E-12	TRUE
18.67	2.11675E-11	TRUE
18.00	4.57148E-11	TRUE
18.58	1.40038E-14	TRUE

18.09	0.000131813	TRUE
18.63	5.58517E-13	TRUE
18.37	1.21453E-10	TRUE
18.43	7.24511E-11	TRUE
18.19	5.27983E-12	TRUE
18.17	4.69041E-13	TRUE
18.26	2.16638E-09	TRUE
18.34	1.69217E-09	TRUE
18.17	4.05074E-13	TRUE
17.99	2.49525E-11	TRUE
18.18	6.4791E-15	TRUE
18.16	6.21904E-13	TRUE
18.34	5.46981E-13	TRUE
17.99	4.67176E-11	TRUE
18.12	9.08674E-14	TRUE
17.91	2.15805E-11	TRUE
17.82	3.59668E-11	TRUE
18.02	7.23071E-16	TRUE
17.62	3.81708E-10	TRUE
17.88	3.03066E-12	TRUE
17.83	2.74784E-12	TRUE
17.82	3.08857E-11	TRUE
17.90	2.09938E-13	TRUE
17.86	5.14979E-13	TRUE
17.58	7.07804E-13	TRUE
17.66	4.44378E-10	TRUE
17.39	2.85844E-11	TRUE
17.59	9.01137E-09	TRUE
17.51	3.20829E-11	TRUE
17.68	9.63491E-11	TRUE
17.64	5.82136E-12	TRUE
17.42	1.85155E-09	TRUE
17.30	1.36462E-12	TRUE
17.23	2.26497E-10	TRUE
17.46	1.29089E-11	TRUE
17.26	1.14469E-13	TRUE
17.01	4.6002E-07	TRUE
17.27	7.2582E-12	TRUE
16.97	3.59842E-09	TRUE
17.12	5.47321E-11	TRUE
17.12	3.68419E-14	TRUE
17.03	1.28579E-11	TRUE
16.74	3.6211E-09	TRUE
17.13	5.45724E-14	TRUE
17.44	6.82515E-12	TRUE
16.75	1.74464E-11	TRUE
17.46	7.70856E-08	TRUE

17.12	2.61794E-13	TRUE
16.74	5.55692E-08	TRUE
16.85	9.0635E-14	TRUE
17.01	1.20454E-13	TRUE
16.68	2.82572E-08	TRUE
16.22	6.77317E-11	TRUE
16.99	5.59995E-10	TRUE
16.71	8.16449E-15	TRUE
16.74	7.56098E-10	TRUE
16.86	4.40692E-09	TRUE
16.64	4.7378E-10	TRUE
16.84	7.24703E-13	TRUE
16.83	5.53958E-13	TRUE
16.40	5.40543E-11	TRUE
16.73	6.1719E-11	TRUE
16.69	3.16732E-13	TRUE
16.36	2.10753E-08	TRUE
-2.76	0.011874065	FALSE
15.25	0.144325943	FALSE
4.43	8.23986E-09	TRUE
16.58	2.13085E-09	TRUE
16.39	7.56441E-12	TRUE
16.40	5.83227E-11	TRUE
16.20	2.00813E-05	TRUE
16.34	1.29245E-09	TRUE
16.44	1.74019E-12	TRUE
16.39	1.30305E-13	TRUE
16.23	4.10966E-10	TRUE
16.16	1.33977E-13	TRUE
16.30	2.82253E-09	TRUE
1.29	0.05012723	FALSE
16.13	2.88069E-10	TRUE
16.19	4.4199E-13	TRUE
15.79	5.89254E-08	TRUE
1.19	0.073947338	FALSE
16.12	6.79153E-11	TRUE
15.71	1.05893E-07	TRUE
16.02	1.35786E-07	TRUE
15.93	2.4927E-13	TRUE
16.03	9.30195E-12	TRUE
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15.98	6.93504E-11	TRUE
16.05	1.60092E-13	TRUE
15.93	5.07218E-12	TRUE
15.95	4.07076E-12	TRUE
0.95	0.173123706	FALSE
15.76	1.49348E-12	TRUE

15.74	3.55852E-15	TRUE
15.72	2.32737E-10	TRUE
15.44	2.74618E-13	TRUE
15.50	6.1547E-09	TRUE
15.69	1.32392E-14	TRUE
15.58	2.52472E-13	TRUE
15.25	2.31053E-17	TRUE
15.37	3.79027E-13	TRUE
15.20	8.9705E-09	TRUE
15.18	2.67979E-13	TRUE
15.14	6.45209E-10	TRUE
15.21	2.09753E-13	TRUE
15.03	8.29721E-09	TRUE
15.06	5.58488E-11	TRUE
14.92	3.31769E-11	TRUE
14.92	2.52054E-10	TRUE

Compound	MS1 Comp Mass		Mass Defect	Retention Time	Formula	Score
8	(526.9574,	263.983	0.983	2.1119998	C5 H F9 O2	87.87
13	(218.9862,	219.9933	0.9933	2.1119998	C4 H F9	87.94
20	(626.9553,	313.9803	0.9803	3.129	C6 H F11 O2	87.72
24	(506.9234,	507.9302	0.9302	1.691	C8 H F11 N3 O8 S	99.3
27	(426.9569,	213.9871	0.9871	1.014	C4 H F7 O2	84.6
38	(498.9295,	499.9366	0.9366	4.7119994	C8 H F17 O3 S	98.28
40	(268.9826,	269.9902	0.9902	3.132	C5 H F11	87.46
46	(622.9279,	623.9349	0.9349	2.857	C13 H F17 O9	95.59
47	(296.946, 1	595.9069	0.9069	1.047	C9 H F19 N O3 S2	98.47
48	(754.9113,	755.9186	0.9186	3.993	C15 H F21 O11	95.92
49	(658.9419,	659.9491	0.9491	3.301	C15 H F21 O5	94.97
51	(926.8723,	463.9386	0.9386	3.9220004	C10 H F13 O4 S	95.88
52	(644.9111,	645.9188	0.9188	2.8729997	C13 H F19 O6 S	89.89
53	(611.9272,	612.9344	0.9344	1.045	C10 H9 F12 O12 S2	97.59
54	(310.9611,	311.9686	0.9686	2.8619998	C5 H F9 O5	99.47
55	(926.8723,	463.9391	0.9391	4.035	C7 H2 F14 O5 S	98.12
56	(886.8955,	887.9034	0.9034	4.7309995	C14 H2 F26 O14	99.74
57	(396.9402,	397.9477	0.9477	3.852	C6 H2 F12 O4 S	98.34
58	(474.9521,	475.9597	0.9597	3.4030004	C8 H5 F13 O6 S	98.99
59	(346.9438,	347.9512	0.9512	2.507	C4 H F9 O8	77.91
60	(266.9574,	535.9269	0.9269	2.045	C9 H5 F13 O7 S2	99.18
61	(680.9254,	681.9324	0.9324	3.301	C11 H F19 N2 O10	99.13
62	(474.9524,	475.9598	0.9598	3.2280002	C8 H5 F13 O6 S	99.44
64	(150.9825,	151.9897	0.9897	2.8699996	C5 H3 F3 S	71.25
65	(184.9843,	185.9917	0.9917	5.028	C3 H F7 O	99.93
67	(168.9893,	169.9965	0.9965	3.301	C3 H F7	88.05
68	(298.9446,	299.952	0.952	1.047	C6 H2 F6 O5 S	87.76
69	(250.9764,	251.984	0.984	3.6630006	C4 H F9 O2	99.34
70	(284.9781,	285.9853	0.9853	3.2970004	C5 H F11 O	87.86
71	(364.9348,	365.9421	0.9421	3.468	C5 H F11 O4 S	78.04
73	(269.9588,	541.9316	0.9316	2.1380002	C9 H2 F16 O6 S	99.06
74	(776.8949,	777.9031	0.9031	3.993	C15 H F23 O8 S	90.83
75	(590.9386,	295.9738	0.9738	2.7630002	C5 H F9 O4	99.19
76	(266.9568,	535.9267	0.9267	1.964	C9 H5 F13 O7 S2	98.81
80	(261.9462,	525.9063	0.9063	0.992	C7 H3 F13 O8 S2	99.33
82	(328.9679,	329.9755	0.9755	3.301	C6 H F11 O3	87.61
83	(943.8996,	944.9076	0.9076	3.9220004	C12 H F30 N5 O8 S	96.18
84	(476.9283,	477.9354	0.9354	4.5429997	C8 H F15 O4 S	99.49
85	(926.91, 57	927.9177	0.9177	1.0430001	C12 H3 F27 N10 O2 S3	97.32
86	(458.9551,	229.9818	0.9818	0.97599995	C4 H F7 O3	98.05
88	(234.981, 8	235.9887	0.9887	2.3419998	C4 H F9 O	88
91	(690.9324,	345.9701	0.9701	3.6630006	C6 H F11 O4	99.79
92	(492.9233,	493.9307	0.9307	4.5159993	C8 H F15 O5 S	99.55
94	(526.9285,	527.9358	0.9358	3.5149999	C8 H3 F15 O7 S	99.33
96	(396.9411,	397.9484	0.9484	3.25	C6 H2 F12 O4 S	99.75
97	(854.9375,	427.9719	0.9719	4.5120006	C4 H F13 N6 O S	96.58

98	(943.8994,	944.9063	0.9063	4.035	C13 H F28 N8 O5 S2	98.23
102	(506.9216,	507.9284	0.9284	1.0720001	C8 H2 F14 O7 S	97.67
103	(412.9358,	413.9428	0.9428	3.492	C6 H2 F12 O5 S	99.67
104	(558.9493,	559.9562	0.9562	2.347	C10 H2 F18 O6	99.24
105	(564.9041,	565.9121	0.9121	1.111	C9 H F19 O2 S2	94.94
106	(494.9523,	991.9217	0.9217	5.029	C15 H2 F28 N5 O13	99.27
107	(360.9736,	361.9812	0.9812	3.6810002	C7 H2 F12 O3	99.01
108	(476.9282,	477.9354	0.9354	4.6559997	C8 H F15 O4 S	99.34
109	(640.9359,	641.9431	0.9431	3.258	C11 H2 F20 O8	99.06
110	(316.9677,	317.975	0.975	4.4179997	C5 H F11 O3	87.79
111	(562.9114,	563.919	0.919	2.1340003	C8 H7 F11 N2 O8 S3	61.08
112	(854.9373,	427.9734	0.9734	3.881	C8 H2 F14 O4	99.5
113	(606.9341,	607.9414	0.9414	2.7800002	C10 H2 F18 O9	98.64
114	(278.9709,	279.9784	0.9784	2.3419998	C5 H F9 O3	87.83
116	(350.9696,	351.9771	0.9771	4.5100007	C6 H F13 O2	87.79
117	(281.961, 8	565.936	0.936	2.0639997	C10 H4 F14 O9 S	95.88
118	(442.9443,	443.9504	0.9504	4.749	C7 H F13 O7	86.68
119	(722.9221,	361.9649	0.9649	3.881	C6 H F11 O5	87.42
122	(184.9842,	185.9918	0.9918	0.974	C3 H F7 O	87.97
124	(672.9075,	673.9152	0.9152	3.322	C11 H2 F20 O8 S	99.56
127	(422.9399,	423.9469	0.9469	0.908		
128	(458.9375,	459.9446	0.9446	4.3649993	C8 H2 F14 O4 S	97.98
129	(672.9078,	673.9148	0.9148	3.426	C11 H2 F20 O8 S	98.85
130	(370.9555,	371.9634	0.9634	3.2970004	C7 H3 F5 N O11	84.58
132	(360.9434,	361.9508	0.9508	0.967	C5 H3 F9 O6 S	98.02
133	(442.9261,	443.9332	0.9332	4.2519994	C7 H F13 O5 S	98.74
134	(426.9652,	427.9725	0.9725	3.9509997	C8 H2 F14 O4	86.91
136	(294.9819,	295.99	0.99	2.316	C6 H2 F10 O2	87.37
140	(380.9052,	381.9126	0.9126	3.5919995	C5 H Cl F10 O4 S	98.34
141	(506.9217,	507.9293	0.9293	0.98599994	C8 H2 F14 O7 S	98.06
143	(314.9381,	315.9449	0.9449	2.712	C4 H F9 O4 S	78.05
145	(1018.8791	509.943	0.943	5.223	C9 H2 F16 O4 S	74.75
146	(310.9433,	311.9502	0.9502	2.5739996	C5 H F9 O3 S	78.31
148	(456.9248,	457.9321	0.9321	0.9610001	C10 H F11 O6 S	75.19
151	(346.9442,	347.9511	0.9511	2.7910001	C4 H F9 O8	80.11
152	(346.9439,	347.9515	0.9515	2.667	C5 H2 F10 O4 S	77.82
153	(822.916, 3	411.9615	0.9615	4.4119997	C7 H F13 O5	87.04
155	(358.9447,	359.9517	0.9517	2.5839999	C6 H2 F10 O4 S	98.96
156	(281.9629,	565.9369	0.9369	1.9750001	C8 H5 F13 N3 O7 S2	97.14
159	(462.9322,	463.9397	0.9397	4.389	C7 H2 F14 O5 S	97.48
161	(246.9507,	247.9579	0.9579	0.923	C3 H2 F6 O4 S	93.28
163	(548.9687,	549.9751	0.9751	4.7729993	C10 H10 F14 N3 O S3	97.94
165	(310.9771,	311.9845	0.9845	2.3250003	C6 H2 F10 O3	87.6
166	(348.9414,	349.949	0.949	2.507	C7 H2 F8 O5 S	72.57
167	(718.9433,	719.9504	0.9504	3.7280002	C11 H3 F17 N5 O12	83.52
168	(772.919, 1	773.9261	0.9261	4.154	C14 H2 F22 N3 O7 S	74.55
171	(574.9461,	575.9538	0.9538	3.8629994	C10 H5 F17 O6 S	98.91

172	(528.9545,	529.9615	0.9615	5.243	C7 H F13 N5 O8	82.56
173	(640.9348,	641.9426	0.9426	2.7940001	C14 H F19 O7	84.7
175	(412.9358,	413.943	0.943	3.6810002	C6 H2 F12 O5 S	99.54
176	(482.9766,	483.9821	0.9821	4.146	C7 H7 F9 N9 S3	97.54
177	(821.9021,	822.9107	0.9107	1.0720001	C14 H9 F18 O15 S2	98.58
178	(481.9769,	482.9844	0.9844	2.7059999	C9 H8 F11 N O7 S	97.85
180	(525.9488,	526.9561	0.9561	2.229	C10 H8 F11 N O7 S2	95.16
181	(458.9571,	459.9646	0.9646	3.7280002	C8 H5 F13 O5 S	98.95
182	(328.9677,	329.9746	0.9746	4.2459993	C6 H F11 O3	86.97
183	(428.9613,	429.9682	0.9682	4.563	C8 H F15 O3	84.38
184	(508.9523,	509.9592	0.9592	2.0989997	C9 H2 F16 O6	86.24
186	(426.9324,	427.9392	0.9392	4.1770005	C7 H F13 O4 S	95.81
187	(481.9764,	482.9838	0.9838	2.5090003	C11 H10 F7 O11 S	98.2
188	(268.9651,	539.9362	0.9362	2.1520002	C9 H6 F10 N2 O9 S2	62.58
190	(532.9878,	533.9951	0.9951	4.5100007	C11 H11 F11 N5 O S3	96.9
191	(366.9319,	367.9389	0.9389	3.466	C7 H F9 O5 S	73.9
192	(458.9573,	459.9638	0.9638	3.581	C8 H5 F13 O5 S	98.5
193	(718.9429,	719.9506	0.9506	3.8060002	C10 H5 F17 N8 O6 S2	65.89
194	(438.9845,	439.9916	0.9916	3.416	C7 H3 F13 N3 O4	87.1
196	(772.9196,	773.9265	0.9265	3.9739995	C11 H6 F18 N5 O10 S2	65.13
197	(614.9601,	615.9671	0.9671	5.2200003	C11 H13 F11 N5 O4 S4	95.85
198	(456.9557,	457.963	0.963	4.625	C9 H F15 O4	86.15
199	(574.9455,	575.9538	0.9538	3.959	C8 H2 F14 N6 O6 S	98.58
200	(628.9159,	629.923	0.923	2.7830002	C12 H7 F11 N O12 S2	63.75
201	(662.9183,	663.9251	0.9251	3.256	C10 H F17 N2 O12	82.94
202	(383.9789,	384.9866	0.9866	0.952	C7 H7 F8 N O6 S	91.03

Downstrea	Downstrea	Downstrea	Downstrea	Downstrea	Upstream_	Upstream_	Upstream_	Upstream_
475443	479333	518468	498432	511659	108693	110716	114212	112410
188550	186792	197391	199418	209238	47174	48072	47351	47363
1	1	1	1	1	129331	131243	133452	134456
123401	123966	73094	66014	45312	23530	23100	18201	25142
101023	104576	106834	112293	109650	20898	21063	22384	21799
21383	21966	23333	22165	24705	61735	63881	36875	64151
1	1	1	1	1	51673	53516	49935	56274
4.67E+07	4.74E+07	4.99E+07	4.97E+07	4.95E+07	1	1	1	1
2.41E+07	2.44E+07	2.47E+07	2.42E+07	2.45E+07	1	1	1	1
1.10E+07	1.14E+07	1.19E+07	1.17E+07	1.17E+07	1	1	1	1
8623775	8758179	9140374	9076449	8929166	1	1	1	1
6893698	7000858	7181519	7179518	7207891	10679	1	10957	1
6629912	6830686	7102419	7163942	7272219	1	1	1	1
6427296	6449398	6493521	6501250	6561621	1	1	1	1
6586903	6762269	7037287	7002966	7118705	1	1	1	1
4468358	4562471	4705168	4676024	4712333	10679	1	10957	1
1920297	1980926	2076225	2055930	2090638	1	1	1	1
1943535	1981885	2031345	1985861	2027813	1	1	1	1
1650174	1681813	1686338	1681405	1731695	1	1	1	1
1795315	1784648	1832358	1806335	1849481	1	1	1	1
1456760	1473889	1521578	1534029	1582304	1	1	1	1
1333354	1374786	1432478	1432511	1429296	1	1	1	1
1341068	1355810	1398775	1395651	1415624	1	1	1	1
1306963	1341051	914935	1397163	1442534	1	1	1	1
1339702	1345596	1380999	1378656	1375585	1	1	1	1
1371779	1353170	1290535	1359401	1381333	1	1	1	1
1172497	1188579	1208202	1191697	1178561	1	1	1	1
1134946	1144691	1157293	1152165	1161509	1	1	1	1
1094947	1103242	1113333	1127956	1152956	1	1	1	1
1074486	1086525	1108636	1103347	1120945	1	1	1	1
820848	815566	870332	868370	875733	1	1	1	1
771929	874099	904415	857528	854204	1	1	1	1
619486	626790	672007	663788	691132	1	1	1	1
681725	669121	733315	726536	705331	1	1	1	1
740591	735992	735187	734974	758066	1	1	1	1
680824	720039	740594	739873	732108	1	1	1	1
421986	428906	428137	439013	446724	1	1	1	1
475877	480076	496652	491295	503496	1	1	1	1
329462	330632	411828	369283	333988	1	1	1	1
309185	314678	356458	342955	347098	1	1	1	1
475582	469013	505949	486502	474168	1	1	1	1
244932	253912	273575	263757	273169	1	1	1	1
383435	386893	396699	399396	394653	1	1	1	1
400418	390514	407403	414519	418181	1	1	1	1
335494	330211	364166	367866	362368	1	1	1	1
287088	287504	308930	305989	305944	1	1	1	1

283675	290881	294321	300389	303642	1	1	1	1
261396	244891	277373	260443	261185	1	1	1	1
290960	294002	302953	299352	296468	1	1	1	1
284054	284738	303561	292661	295512	1	1	1	1
327944	336666	317030	333123	340218	1	1	1	1
241902	254362	269941	269331	268678	1	1	1	1
274793	283972	288849	286781	290572	1	1	1	1
237470	238864	243948	263790	243315	1	1	1	1
242563	239257	219839	235430	220491	1	1	1	1
261060	263991	266419	266263	269568	1	1	1	1
188810	186736	211174	205467	213066	1	1	1	1
231327	238411	241203	246100	253109	1	1	1	1
232207	223845	237900	244144	229551	1	1	1	1
220939	220121	241064	241739	231668	1	1	1	1
231188	231920	236622	242488	245803	1	1	1	1
190525	193474	199582	203963	192915	1	1	1	1
189338	199774	222789	214697	211025	1	1	1	1
164834	167752	172991	170212	184806	1	1	1	1
203613	198207	226939	212928	208022	1	1	1	1
161147	176817	164487	179994	194313	1	1	1	1
169989	175142	183897	186054	186170	1	1	1	1
153613	154475	157301	161739	160264	1	1	1	1
112643	114877	148771	157028	125620	1	1	1	1
150870	156255	155342	161926	166113	1	1	1	1
134967	135959	145462	146186	150891	1	1	1	1
141795	140772	139765	144465	146519	1	1	1	1
127666	128252	134681	138486	138917	1	1	1	1
139777	140503	145384	145711	145291	1	1	1	1
145358	138102	142509	147178	141166	1	1	1	1
98799	106023	102028	110351	131128	1	1	1	1
129183	128285	132512	135625	132833	1	1	1	1
70966	75034	80478	76869	79614	1	1	1	1
135773	134045	138538	120734	121182	1	1	1	1
102021	105231	106682	111005	121062	1	1	1	1
115398	114390	115855	120838	121535	1	1	1	1
113798	112958	119594	119748	117463	1	1	1	1
82090	83595	91633	90500	85683	1	1	1	1
101675	105680	105036	108288	108089	1	1	1	1
73274	78999	89089	82587	95383	1	1	1	1
85995	92407	101617	93562	92984	10679	1	10957	1
81933	84727	85791	87065	90821	1	1	1	1
59596	57365	68925	99603	90348	1	1	1	1
87472	85734	91698	92030	87581	1	1	1	1
86032	83979	87624	84591	88621	1	1	1	1
69601	74367	77863	80519	81210	1	1	1	1
71211	75508	72951	74447	72536	1	1	1	1
67660	66862	74153	72007	76949	1	1	1	1

71869	75460	77130	76062	74553	1	1	1	1
50694	52870	57166	67727	54774	1	1	1	1
67704	68647	70724	71931	76762	1	1	1	1
44302	48276	56663	59612	58282	1	1	1	1
69364	72459	57882	75770	56694	1	1	1	1
61752	60337	62478	64063	63867	1	1	1	1
65699	64404	68354	69652	67508	1	1	1	1
60938	69229	62513	66140	64143	1	1	1	1
66561	67160	69861	69199	66411	1	1	1	1
60914	61741	65361	64088	59953	1	1	1	1
64093	64444	59874	65335	61910	1	1	1	1
53884	54257	55288	56289	58150	1	1	1	1
53579	53960	55335	54947	55238	1	1	1	1
52932	49766	56730	57526	53188	1	1	1	1
46849	40818	52196	46517	45184	1	1	1	1
53453	51572	52694	52369	53867	1	1	1	1
49592	49615	47250	49952	47878	1	1	1	1
39141	38946	39283	39393	38627	1	1	1	1
42220	41634	41879	41331	44116	1	1	1	1
36415	36110	36760	38322	37707	1	1	1	1
34014	34926	39021	38183	34053	1	1	1	1
36778	37931	39319	38010	37605	1	1	1	1
31954	28918	35940	36127	34623	1	1	1	1
33422	34801	32944	36977	33243	1	1	1	1
29661	32564	29975	30127	32390	1	1	1	1
29484	29293	31047	33812	31443	1	1	1	1

Upstream_	Downstrea	Upstream_	log2 Fold Change	p-val (unadjusted)	Signif? (Bonferroni)
112848	4.97E+05	111775.8	2.15	6.73064E-11	TRUE
51331	1.96E+05	48258.2	2.02	4.0186E-10	TRUE
132827	1.00E+00	132261.8	-17.01	5.11421E-15	TRUE
26267	8.64E+04	23248	1.89	0.00422241	TRUE
20833	1.07E+05	21395.4	2.32	9.13732E-11	TRUE
67629	2.27E+04	58854.2	-1.37	0.000199023	TRUE
56217	1.00E+00	53523	-15.71	9.6012E-11	TRUE
1	4.86E+07	1	25.54	1.5697E-12	TRUE
1	2.44E+07	1	24.54	2.57654E-16	TRUE
1	1.15E+07	1	23.46	1.10578E-12	TRUE
1	8.91E+06	1	23.09	2.11148E-13	TRUE
1	7.09E+06	4327.8	10.68	3.82694E-14	TRUE
1	7.00E+06	1	22.74	7.10267E-12	TRUE
1	6.49E+06	1	22.63	3.02789E-17	TRUE
1	6.90E+06	1	22.72	1.9255E-12	TRUE
1	4.62E+06	4327.8	10.06	1.40763E-13	TRUE
1	2.02E+06	1	20.95	4.5986E-12	TRUE
1	1.99E+06	1	20.93	2.20115E-14	TRUE
1	1.69E+06	1	20.69	1.44498E-14	TRUE
1	1.81E+06	1	20.79	4.02211E-15	TRUE
1	1.51E+06	1	20.53	2.53264E-12	TRUE
1	1.40E+06	1	20.42	1.97103E-12	TRUE
1	1.38E+06	1	20.40	1.29149E-13	TRUE
1	1.28E+06	1	20.29	8.31692E-07	TRUE
1	1.36E+06	1	20.38	3.51627E-15	TRUE
1	1.35E+06	1	20.37	4.18721E-13	TRUE
1	1.19E+06	1	20.18	5.62687E-16	TRUE
1	1.15E+06	1	20.13	8.96699E-17	TRUE
1	1.12E+06	1	20.09	5.4491E-14	TRUE
1	1.10E+06	1	20.07	1.09144E-14	TRUE
1	8.50E+05	1	19.70	3.60215E-12	TRUE
1	8.52E+05	1	19.70	2.1641E-10	TRUE
1	6.55E+05	1	19.32	3.96035E-11	TRUE
1	7.03E+05	1	19.42	1.04221E-11	TRUE
1	7.41E+05	1	19.50	1.71912E-15	TRUE
1	7.23E+05	1	19.46	3.44606E-12	TRUE
1	4.33E+05	1	18.72	1.25774E-13	TRUE
1	4.89E+05	1	18.90	1.60459E-13	TRUE
1	3.55E+05	1	18.44	1.79923E-08	TRUE
1	3.34E+05	1	18.35	4.09258E-10	TRUE
1	4.82E+05	1	18.88	1.33094E-12	TRUE
1	2.62E+05	1	18.00	4.57148E-11	TRUE
1	3.92E+05	1	18.58	1.40038E-14	TRUE
1	4.06E+05	1	18.63	5.58517E-13	TRUE
1	3.52E+05	1	18.43	7.24511E-11	TRUE
1	2.99E+05	1	18.19	5.27983E-12	TRUE

1	2.95E+05	1	18.17	4.69041E-13	TRUE
1	2.61E+05	1	17.99	2.49525E-11	TRUE
1	2.97E+05	1	18.18	6.4791E-15	TRUE
1	2.92E+05	1	18.16	6.21904E-13	TRUE
1	3.31E+05	1	18.34	5.46981E-13	TRUE
1	2.61E+05	1	17.99	4.67176E-11	TRUE
1	2.85E+05	1	18.12	9.08674E-14	TRUE
1	2.45E+05	1	17.91	2.15805E-11	TRUE
1	2.32E+05	1	17.82	3.59668E-11	TRUE
1	2.65E+05	1	18.02	7.23071E-16	TRUE
1	2.01E+05	1	17.62	3.81708E-10	TRUE
1	2.42E+05	1	17.88	3.03066E-12	TRUE
1	2.34E+05	1	17.83	2.74784E-12	TRUE
1	2.31E+05	1	17.82	3.08857E-11	TRUE
1	2.38E+05	1	17.86	5.14979E-13	TRUE
1	1.96E+05	1	17.58	7.07804E-13	TRUE
1	2.08E+05	1	17.66	4.44378E-10	TRUE
1	1.72E+05	1	17.39	2.85844E-11	TRUE
1	2.10E+05	1	17.68	9.63491E-11	TRUE
1	1.75E+05	1	17.42	1.85155E-09	TRUE
1	1.80E+05	1	17.46	1.29089E-11	TRUE
1	1.57E+05	1	17.26	1.14469E-13	TRUE
1	1.32E+05	1	17.01	4.6002E-07	TRUE
1	1.58E+05	1	17.27	7.2582E-12	TRUE
1	1.43E+05	1	17.12	5.47321E-11	TRUE
1	1.43E+05	1	17.12	3.68419E-14	TRUE
1	1.34E+05	1	17.03	1.28579E-11	TRUE
1	1.43E+05	1	17.13	5.45724E-14	TRUE
1	1.43E+05	1	17.12	2.61794E-13	TRUE
1	1.10E+05	1	16.74	5.55692E-08	TRUE
1	1.32E+05	1	17.01	1.20454E-13	TRUE
1	7.66E+04	1	16.22	6.77317E-11	TRUE
1	1.30E+05	1	16.99	5.59995E-10	TRUE
1	1.09E+05	1	16.74	7.56098E-10	TRUE
1	1.18E+05	1	16.84	7.24703E-13	TRUE
1	1.17E+05	1	16.83	5.53958E-13	TRUE
1	8.67E+04	1	16.40	5.40543E-11	TRUE
1	1.06E+05	1	16.69	3.16732E-13	TRUE
1	8.39E+04	1	16.36	2.10753E-08	TRUE
1	9.33E+04	4327.8	4.43	8.23986E-09	TRUE
1	8.61E+04	1	16.39	7.56441E-12	TRUE
1	7.52E+04	1	16.20	2.00813E-05	TRUE
1	8.89E+04	1	16.44	1.74019E-12	TRUE
1	8.62E+04	1	16.39	1.30305E-13	TRUE
1	7.67E+04	1	16.23	4.10966E-10	TRUE
1	7.33E+04	1	16.16	1.33977E-13	TRUE
1	7.15E+04	1	16.13	2.88069E-10	TRUE

1	7.50E+04	1	16.19	4.4199E-13	TRUE
1	5.66E+04	1	15.79	5.89254E-08	TRUE
1	7.12E+04	1	16.12	6.79153E-11	TRUE
1	5.34E+04	1	15.71	1.05893E-07	TRUE
1	6.64E+04	1	16.02	1.35786E-07	TRUE
1	6.25E+04	1	15.93	2.4927E-13	TRUE
1	6.71E+04	1	16.03	1.5773E-12	TRUE
1	6.46E+04	1	15.98	6.93504E-11	TRUE
1	6.78E+04	1	16.05	1.60092E-13	TRUE
1	6.24E+04	1	15.93	5.07218E-12	TRUE
1	6.31E+04	1	15.95	4.07076E-12	TRUE
1	5.56E+04	1	15.76	1.49348E-12	TRUE
1	5.46E+04	1	15.74	3.55852E-15	TRUE
1	5.40E+04	1	15.72	2.32737E-10	TRUE
1	4.63E+04	1	15.50	6.1547E-09	TRUE
1	5.28E+04	1	15.69	1.32392E-14	TRUE
1	4.89E+04	1	15.58	2.52472E-13	TRUE
1	3.91E+04	1	15.25	2.31053E-17	TRUE
1	4.22E+04	1	15.37	3.79027E-13	TRUE
1	3.71E+04	1	15.18	2.67979E-13	TRUE
1	3.60E+04	1	15.14	6.45209E-10	TRUE
1	3.79E+04	1	15.21	2.09753E-13	TRUE
1	3.35E+04	1	15.03	8.29721E-09	TRUE
1	3.43E+04	1	15.06	5.58488E-11	TRUE
1	3.09E+04	1	14.92	3.31769E-11	TRUE
1	3.10E+04	1	14.92	2.52054E-10	TRUE

INPUT	FOUND_BY	DTXCID_IN	FORMULA	SMILES_IN	DTXSID	PREFERRI	CASRN
C5HF9O2	MS Ready Formula	DTXCID90	C5HF9O2	OC(=O)C(F)	DTXSID60	Perfluorop	2706-90-3
C5HF9O2	MS Ready Formula	DTXCID90	C5HF9O2	OC(=O)C(F)	DTXSID40	Sodium pe	2706-89-0
C5HF9O2	MS Ready Formula	DTXCID90	C5HF9O2	OC(=O)C(F)	DTXSID10	Ammoniun	68259-11-0
C5HF9O2	MS Ready Formula	DTXCID00	C5HF9O2	FC(F)(F)C(C	DTXSID80	1,1,1,3,3,3-	42031-15-2
C5HF9O2	MS Ready Formula	DTXCID90	C5HF9O2	OC(=O)C(F)	DTXSID00	Perfluorop	45167-47-3
C5HF9O2	MS Ready Formula	DTXCID70	C5HF9O2	OC(=O)C(C	DTXSID00	3,3,3-Triflu	917951-63-
C5HF9O2	MS Ready Formula	DTXCID40	C5HF9O2	FC(OC(F)(F)	DTXSID00	1,1,1,3,3-P	88457-11-8
C5HF9O2	MS Ready Formula	DTXCID90	C5HF9O2	OC(=O)C(F)	DTXSID90	Nonafluorc	64808-55-5
C5HF9O2	MS Ready Formula	DTXCID90	C5HF9O2	OC(=O)C(F)	DTXSID80	Perfluoroal	NOCAS_89
C4HF9	MS Ready Formula	DTXCID30	C4HF9	FC(F)C(F)(F	DTXSID20	1H-Nonaflu	375-17-7
C4HF9	MS Ready Formula	DTXCID80	C4HF9	FC(F)(F)C(C	DTXSID40	2-(Trifluorc	382-24-1
C4HF9	MS Ready Formula	DTXCID30	C4HF9	FC(F)C(F)(F	DTXSID60	Cadmium b	93367-04-5
C4HF9	MS Ready Formula	DTXCID30	C4HF9	FC(F)C(F)(F	DTXSID30	Magnesiun	82416-69-1
C4HF9	MS Ready Formula	DTXCID30	C4HF9	FC(F)C(F)(F	DTXSID00	Copper(1+)	124521-58-
C6HF11O2	MS Ready Formula	DTXCID10	C6HF11O2	OC(=O)C(F)	DTXSID30	Perfluoroh	307-24-4
C6HF11O2	MS Ready Formula	DTXCID10	C6HF11O2	OC(=O)C(F)	DTXSID30	Sodium pe	2923-26-4
C6HF11O2	MS Ready Formula	DTXCID10	C6HF11O2	OC(=O)C(F)	DTXSID90	Ammoniun	21615-47-4
C6HF11O2	MS Ready Formula	DTXCID10	C6HF11O2	OC(=O)C(F)	DTXSID20	Undecafluc	423-47-2
C6HF11O2	MS Ready Formula	DTXCID10	C6HF11O2	OC(=O)C(F)	DTXSID20	1-phenylpi	985-60-4
C6HF11O2	MS Ready Formula	DTXCID10	C6HF11O2	OC(=O)C(F)	DTXSID20	Perfluoroh	92612-52-7
C6HF11O2	MS Ready Formula	DTXCID50	C6HF11O2	OC(=O)C(F)	DTXSID00	2,2,3,4,4,5,	64139-66-8
C6HF11O2	MS Ready Formula	DTXCID10	C6HF11O2	OC(=O)C(F)	DTXSID30	Undecafluc	565225-91-
C6HF11O2	MS Ready Formula	DTXCID10	C6HF11O2	OC(=O)C(F)	DTXSID80	Perfluoroal	NOCAS_89
C8HF11N3(	NO_MATCH	-	-	-	-	-	-
C4HF7O2	MS Ready Formula	DTXCID10	C4HF7O2	OC(=O)C(F)	DTXSID40	Perfluorob	375-22-4
C4HF7O2	MS Ready Formula	DTXCID10	C4HF7O2	OC(=O)C(F)	DTXSID70	Silver perfl	3794-64-7
C4HF7O2	MS Ready Formula	DTXCID10	C4HF7O2	OC(=O)C(F)	DTXSID70	Sodium pe	2218-54-4
C4HF7O2	MS Ready Formula	DTXCID70	C4HF7O2	OC(=O)C(F)	DTXSID10	Perfluoris	335-10-4
C4HF7O2	MS Ready Formula	DTXCID10	C4HF7O2	OC(=O)C(F)	DTXSID60	Heptafluor	375-04-2
C4HF7O2	MS Ready Formula	DTXCID10	C4HF7O2	OC(=O)C(F)	DTXSID80	heptafluor	2263-11-8
C4HF7O2	MS Ready Formula	DTXCID10	C4HF7O2	OC(=O)C(F)	DTXSID80	Perfluorob	45048-62-2
C4HF7O2	MS Ready Formula	DTXCID50	C4HF7O2	FC(F)C(F)(F	DTXSID40	Difluoro(1,	89076-47-1
C4HF7O2	MS Ready Formula	DTXCID10	C4HF7O2	OC(=O)C(F)	DTXSID20	N~5~-(5-Hy	936233-19-
C4HF7O2	MS Ready Formula	DTXCID10	C4HF7O2	OC(=O)C(F)	DTXSID80	Perfluoroal	NOCAS_89
C8HF17O3S	MS Ready Formula	DTXCID10	C8HF17O3S	OS(=O)(=O)	DTXSID30	Perfluoroo	1763-23-1
C8HF17O3S	MS Ready Formula	DTXCID10	C8HF17O3S	OS(=O)(=O)	DTXSID80	Potassium	2795-39-3
C8HF17O3S	MS Ready Formula	DTXCID10	C8HF17O3S	OS(=O)(=O)	DTXSID20	Lithium pe	29457-72-5
C8HF17O3S	MS Ready Formula	DTXCID10	C8HF17O3S	OS(=O)(=O)	DTXSID50	Tetraethyla	56773-42-3
C8HF17O3S	MS Ready Formula	DTXCID10	C8HF17O3S	OS(=O)(=O)	DTXSID90	Ammoniun	29081-56-9
C8HF17O3S	MS Ready Formula	DTXCID10	C8HF17O3S	OS(=O)(=O)	DTXSID80	Perfluoroo	45298-90-6
C8HF17O3S	MS Ready Formula	DTXCID10	C8HF17O3S	OS(=O)(=O)	DTXSID20	Bis(2-hydrc	70225-14-8
C8HF17O3S	MS Ready Formula	DTXCID10	C8HF17O3S	OS(=O)(=O)	DTXSID00	Piperidiniu	71463-74-6
C8HF17O3S	MS Ready Formula	DTXCID10	C8HF17O3S	OS(=O)(=O)	DTXSID00	N-Decyl-N,	251099-16-
C8HF17O3S	MS Ready Formula	DTXCID10	C8HF17O3S	OS(=O)(=O)	DTXSID50	Sodium pe	4021-47-0
C8HF17O3S	MS Ready Formula	DTXCID40	C8HF17O3S	OS(=O)(=O)	DTXSID30	Magnesiun	93894-73-6
C8HF17O3S	MS Ready Formula	DTXCID40	C8HF17O3S	OS(=O)(=O)	DTXSID50	Tetraethyla	93894-70-3

C8HF17O3S	MS Ready Formula	DTXCID10	C8HF17O3S	OS(=O)(=O)	DTXSID40	Tetrabutyla	111873-33
C8HF17O3S	MS Ready Formula	DTXCID10	C8HF17O3S	OS(=O)(=O)	DTXSID70	Perfluoroal	NOCAS_89
C5HF11	MS Ready Formula	DTXCID00	C5HF11	FC(C(F)(F)C	DTXSID90	1,1,1,2,2,3,	95576-22-0
C13HF17O	NO_MATCH	-	-	-	-	-	-
C9HF19NO	NO_MATCH	-	-	-	-	-	-
C15HF21O	NO_MATCH	-	-	-	-	-	-
C15HF21O	NO_MATCH	-	-	-	-	-	-
C10HF13O	NO_MATCH	-	-	-	-	-	-
C13HF19O	NO_MATCH	-	-	-	-	-	-
C10H9F12	NO_MATCH	-	-	-	-	-	-
C5HF9O5	MS Ready Formula	DTXCID80	C5HF9O5	OC(=O)C(F)	DTXSID20	Perfluoro-3	39492-89-2
C7H2F14O	MS Ready Formula	DTXCID60	C7H2F14O	OS(=O)(=O)	DTXSID10	Perfluoro-2	749836-20
C7H2F14O	MS Ready Formula	DTXCID70	C7H2F14O	OS(=O)(=O)	DTXSID40	1,1,2,2-Tet	NOCAS_89
C14H2F26	NO_MATCH	-	-	-	-	-	-
C6H2F12O	NO_MATCH	-	-	-	-	-	-
C8H5F13O	NO_MATCH	-	-	-	-	-	-
C4HF9O8	NO_MATCH	-	-	-	-	-	-
C9H5F13O	NO_MATCH	-	-	-	-	-	-
C11HF19N	NO_MATCH	-	-	-	-	-	-
C8H5F13O	NO_MATCH	-	-	-	-	-	-
C5H3F3S	MS Ready Formula	DTXCID40	C5H3F3S	FC(F)(F)C1=	DTXSID20	2-(Trifluorc	86093-76-7
C5H3F3S	MS Ready Formula	DTXCID10	C5H3F3S	FC(F)(F)C1=	DTXSID50	3-(trifluoro	86093-77-8
C3HF7O	MS Ready Formula	DTXCID40	C3HF7O	OC(F)(F)C(F	DTXSID60	Perfluorop	72301-80-5
C3HF7O	MS Ready Formula	DTXCID80	C3HF7O	OC(F)(C(F)(	DTXSID80	Heptafluor	24427-67-6
C3HF7	MS Ready Formula	DTXCID20	C3HF7	FC(C(F)(F)F	DTXSID40	2H-Perfluo	431-89-0
C3HF7	MS Ready Formula	DTXCID50	C3HF7	FC(F)C(F)(F	DTXSID40	1H-Heptafl	2252-84-8
C3HF7	MS Ready Formula	DTXCID50	C3HF7	FC(F)C(F)(F	DTXSID90	Propyl,hep	3170-79-4
C3HF7	MS Ready Formula	DTXCID50	C3HF7	FC(F)C(F)(F	DTXSID30	PUBCHEM_	7223-98-5
C3HF7	MS Ready Formula	DTXCID20	C3HF7	FC(C(F)(F)F	DTXSID50	Silver(1+) 1	22407-25-6
C3HF7	MS Ready Formula	DTXCID20	C3HF7	FC(C(F)(F)F	DTXSID90	Iodozinc(1+	62656-71-7
C6H2F6O5S	NO_MATCH	-	-	-	-	-	-
C4HF9O2	NO_MATCH	-	-	-	-	-	-
C5HF11O	MS Ready Formula	DTXCID50	C5HF11O	FC(OC(F)(F)	DTXSID80	Perfluoro-3	3330-15-2
C5HF11O	MS Ready Formula	DTXCID00	C5HF11O	OC(C(F)(F)F	DTXSID70	1,1,1,3,3,4,	6188-98-3
C5HF11O4S	NO_MATCH	-	-	-	-	-	-
C9H2F16O	NO_MATCH	-	-	-	-	-	-
C15HF23O	NO_MATCH	-	-	-	-	-	-
C5HF9O4	MS Ready Formula	DTXCID60	C5HF9O4	OC(=O)C(F)	DTXSID30	Perfluoro-3	151772-58
C9H5F13O	NO_MATCH	-	-	-	-	-	-
C7H3F13O	NO_MATCH	-	-	-	-	-	-
C6HF11O3	MS Ready Formula	DTXCID10	C6HF11O3	OC(=O)C(F)	DTXSID40	Ammonium	62037-80-3
C6HF11O3	MS Ready Formula	DTXCID10	C6HF11O3	OC(=O)C(F)	DTXSID70	Perfluoro-2	13252-13-6
C6HF11O3	MS Ready Formula	DTXCID30	C6HF11O3	OC(=O)C(F)	DTXSID20	2,2,3,3-Tet	378-03-0
C6HF11O3	MS Ready Formula	DTXCID30	C6HF11O3	OC(=O)C(F)	DTXSID50	Potassium	84100-11-8
C6HF11O3	MS Ready Formula	DTXCID30	C6HF11O3	OC(=O)C(F)	DTXSID60	2,2,3,3,4,4-	919005-54
C12HF30N	NO_MATCH	-	-	-	-	-	-
C8HF15O4S	MS Ready Formula	DTXCID90	C8HF15O4S	OS(=O)(=O)	DTXSID70	6-Oxoperfl	NOCAS_88

C12H3F27N	NO_MATCH	-	-	-	-	-	-
C4HF7O3	MS Ready Formula	DTXCID80	C4HF7O3	OC(=O)C(F)	DTXSID70	Perfluoro-3	377-73-1
C4HF7O3	MS Ready Formula	DTXCID20	C4HF7O3	OC(=O)C(F)	DTXSID80	Perfluoro-2	13140-29-9
C4HF9O	MS Ready Formula	DTXCID90	C4HF9O	OC(C(F)(F)F	DTXSID50	Nonafluorc	2378-02-1
C4HF9O	MS Ready Formula	DTXCID10	C4HF9O	FC(F)C(F)(F	DTXSID00	1,1,2,2,3,3-	67490-36-2
C4HF9O	MS Ready Formula	DTXCID40	C4HF9O	OC(F)(F)C(F	DTXSID80	Perfluorob	3056-01-7
C4HF9O	MS Ready Formula	DTXCID90	C4HF9O	OC(C(F)(F)F	DTXSID20	Potassium	29646-16-0
C6HF11O4	MS Ready Formula	DTXCID70	C6HF11O4	OC(=O)C(F)	DTXSID20	Perfluoro-3	80153-82-8
C8HF15O5	NO_MATCH	-	-	-	-	-	-
C8H3F15O	NO_MATCH	-	-	-	-	-	-
C6H2F12O	NO_MATCH	-	-	-	-	-	-
C4HF13N6	NO_MATCH	-	-	-	-	-	-
C13HF28N	NO_MATCH	-	-	-	-	-	-
C8H2F14O	NO_MATCH	-	-	-	-	-	-
C6H2F12O	NO_MATCH	-	-	-	-	-	-
C10H2F18	NO_MATCH	-	-	-	-	-	-
C9HF19O2	NO_MATCH	-	-	-	-	-	-
C15H2F28	NO_MATCH	-	-	-	-	-	-
C7H2F12O	MS Ready Formula	DTXCID60	C7H2F12O	OC(=O)C(O	DTXSID30	2,3,3,4,4,5,	93682-62-3
C8HF15O4	MS Ready Formula	DTXCID90	C8HF15O4	OS(=O)(=O)	DTXSID70	6-Oxoperfl	NOCAS_88
C11H2F20	NO_MATCH	-	-	-	-	-	-
C5HF11O3	NO_MATCH	-	-	-	-	-	-
C8H7F11N	NO_MATCH	-	-	-	-	-	-
C8H2F14O	NO_MATCH	-	-	-	-	-	-
C10H2F18	NO_MATCH	-	-	-	-	-	-
C5HF9O3	MS Ready Formula	DTXCID50	C5HF9O3	OC(=O)C(F)	DTXSID60	Perfluoro(4	863090-89
C5HF9O3	MS Ready Formula	DTXCID90	C5HF9O3	OC(=O)C(F)	DTXSID60	Perfluoro-3	377-76-4
C5HF9O3	MS Ready Formula	DTXCID50	C5HF9O3	OC(=O)C(F)	DTXSID30	Difluoro(pe	919005-50
C6HF13O2	NO_MATCH	-	-	-	-	-	-
C10H4F14	NO_MATCH	-	-	-	-	-	-
C7HF13O7	MS Ready Formula	DTXCID90	C7HF13O7	OC(=O)C(F)	DTXSID50	Perfluoro-3	39492-91-6
C6HF11O5	NO_MATCH	-	-	-	-	-	-
C3HF7O	MS Ready Formula	DTXCID40	C3HF7O	OC(F)(F)C(F	DTXSID60	Perfluorop	72301-80-5
C3HF7O	MS Ready Formula	DTXCID80	C3HF7O	OC(F)(C(F)(	DTXSID80	Heptafluor	24427-67-6
C11H2F20	NO_MATCH	-	-	-	-	-	-
C8H2F14O	NO_MATCH	-	-	-	-	-	-
C11H2F20	NO_MATCH	-	-	-	-	-	-
C7H3F5NO	NO_MATCH	-	-	-	-	-	-
C5H3F9O6	NO_MATCH	-	-	-	-	-	-
C7HF13O5	MS Ready Formula	DTXCID60	C7HF13O5	OS(=O)(=O)	DTXSID30	Perfluoro-3	29311-67-9
C8H2F14O	NO_MATCH	-	-	-	-	-	-
C6H2F10O	MS Ready Formula	DTXCID50	C6H2F10O	OC(=O)C(F)	DTXSID20	6-H-Perfluc	1726-50-7
C6H2F10O	MS Ready Formula	DTXCID60	C6H2F10O	OC(O)(C(F)	DTXSID10	1,1,1,3,5,5,	89810-81-1
C6H2F10O	MS Ready Formula	DTXCID10	C6H2F10O	FC(F)(F)CO	DTXSID20	2,2,2-Triflu	336-63-0
C5HClF10O	NO_MATCH	-	-	-	-	-	-
C8H2F14O	NO_MATCH	-	-	-	-	-	-
C4HF9O4S	MS Ready Formula	DTXCID10	C4HF9O4S	OS(=O)(=O)	DTXSID50	Perfluoro(2	113507-82

C4HF9O4S	MS Ready Formula	DTXCID10	C4HF9O4S	OS(=O)(=O)	DTXSID40	Potassium	117205-07
C4HF9O4S	MS Ready Formula	DTXCID10	C4HF9O4S	OS(=O)(=O)	DTXSID40	Sodium 1,1	113507-87
C9H2F16O	NO_MATCH	-	-	-	-	-	-
C5HF9O3S	NO_MATCH	-	-	-	-	-	-
C10HF11O	NO_MATCH	-	-	-	-	-	-
C4HF9O8	NO_MATCH	-	-	-	-	-	-
C5H2F10O	NO_MATCH	-	-	-	-	-	-
C7HF13O5	MS Ready Formula	DTXCID60	C7HF13O5	OC(=O)C(F)	DTXSID80	Perfluoro-3	151772-59
C6H2F10O	NO_MATCH	-	-	-	-	-	-
C8H5F13N	NO_MATCH	-	-	-	-	-	-
C7H2F14O	MS Ready Formula	DTXCID60	C7H2F14O	OS(=O)(=O)	DTXSID10	Perfluoro-2	749836-20
C7H2F14O	MS Ready Formula	DTXCID70	C7H2F14O	OS(=O)(=O)	DTXSID40	1,1,2,2-Tet	NOCAS_89
C3H2F6O4	NO_MATCH	-	-	-	-	-	-
C10H10F1	NO_MATCH	-	-	-	-	-	-
C6H2F10O	MS Ready Formula	DTXCID90	C6H2F10O	OC(=O)C(F)	DTXSID30	2,2,3-Triflu	824393-42
C6H2F10O	MS Ready Formula	DTXCID90	C6H2F10O	OC(=O)C(F)	DTXSID70	2,2,3,3,4,4-	919005-52
C6H2F10O	MS Ready Formula	DTXCID70	C6H2F10O	OC(=O)C(F)	DTXSID90	2,2,3,4,4-P	919005-25
C6H2F10O	MS Ready Formula	DTXCID90	C6H2F10O	OC(=O)C(F)	DTXSID10	2,3,3-Triflu	919005-19
C6H2F10O	MS Ready Formula	DTXCID70	C6H2F10O	OC(=O)C(O	DTXSID30	3,3,3-Triflu	326495-77
C7H2F8O5	NO_MATCH	-	-	-	-	-	-
C11H3F17	NO_MATCH	-	-	-	-	-	-
C14H2F22	NO_MATCH	-	-	-	-	-	-
C10H5F17	NO_MATCH	-	-	-	-	-	-
C7HF13N5	NO_MATCH	-	-	-	-	-	-
C14HF19O	NO_MATCH	-	-	-	-	-	-
C6H2F12O	NO_MATCH	-	-	-	-	-	-
C7H7F9N9	NO_MATCH	-	-	-	-	-	-
C14H9F18	NO_MATCH	-	-	-	-	-	-
C9H8F11N	NO_MATCH	-	-	-	-	-	-
C10H8F11	NO_MATCH	-	-	-	-	-	-
C8H5F13O	NO_MATCH	-	-	-	-	-	-
C6HF11O3	MS Ready Formula	DTXCID10	C6HF11O3	OC(=O)C(F)	DTXSID40	Ammonium	62037-80-3
C6HF11O3	MS Ready Formula	DTXCID10	C6HF11O3	OC(=O)C(F)	DTXSID70	Perfluoro-2	13252-13-6
C6HF11O3	MS Ready Formula	DTXCID30	C6HF11O3	OC(=O)C(F)	DTXSID20	2,2,3,3-Tet	378-03-0
C6HF11O3	MS Ready Formula	DTXCID30	C6HF11O3	OC(=O)C(F)	DTXSID50	Potassium	84100-11-8
C6HF11O3	MS Ready Formula	DTXCID30	C6HF11O3	OC(=O)C(F)	DTXSID60	2,2,3,3,4,4-	919005-54
C8HF15O3	MS Ready Formula	DTXCID50	C8HF15O3	OC(=O)C(F)	DTXSID20	Perfluoro-2	504435-11
C8HF15O3	MS Ready Formula	DTXCID20	C8HF15O3	OC(=O)C(O	DTXSID80	Perfluoro-2	NOCAS_89
C8HF15O3	MS Ready Formula	DTXCID30	C8HF15O3	OC(=O)C(F)	DTXSID90	2,2,3,3,4,4-	174767-06
C9H2F16O	NO_MATCH	-	-	-	-	-	-
C7HF13O4	NO_MATCH	-	-	-	-	-	-
C11H10F7	NO_MATCH	-	-	-	-	-	-
C9H6F10N	NO_MATCH	-	-	-	-	-	-
C11H11F11	NO_MATCH	-	-	-	-	-	-
C7HF9O5S	NO_MATCH	-	-	-	-	-	-
C8H5F13O	NO_MATCH	-	-	-	-	-	-
C10H5F17	NO_MATCH	-	-	-	-	-	-

C7H3F13N	NO_MATCH	-	-	-	-	-	-
C11H6F18	NO_MATCH	-	-	-	-	-	-
C11H13F11	NO_MATCH	-	-	-	-	-	-
C9HF15O4	NO_MATCH	-	-	-	-	-	-
C8H2F14N	NO_MATCH	-	-	-	-	-	-
C12H7F11	NO_MATCH	-	-	-	-	-	-
C10HF17N	NO_MATCH	-	-	-	-	-	-
C7H7F8NO	NO_MATCH	-	-	-	-	-	-

MOLECUL	MONOISO	DATA_SOURCES
C5HF9O2	263.9833	34
C5F9NaO2	285.9652	8
C5H4F9NO	281.0098	8
C5HF9O2	263.9833	4
C5F9O2	262.976	2
C5HF9O2	263.9833	1
C5HF9O2	263.9833	1
C13H11F9N	430.0398	1
(CF2)nCHF(	-	1
C4HF9	219.9935	9
C4HF9	219.9935	3
C8CdF18	551.8746	3
C4BrF9Mg	321.889	1
C4CuF9	281.9152	1
C6HF11O2	313.9801	61
C6F11NaO2	335.962	18
C6H4F11N(	331.0066	9
C10H11F11	400.0645	3
C16H15F11	476.0958	2
C6F11O2	312.9728	2
C6HF11O2	313.9801	1
C12H16F11	415.1005	1
(CF2)nCHF(	-	1
-	-	-
C4HF7O2	213.9865	46
C4AgF7O2	319.8837	11
C4F7NaO2	235.9684	8
C4HF7O2	213.9865	5
C8H11F7N2	300.0709	3
C14H15F7N	376.1022	2
C4F7O2	212.9792	2
C4HF7O2	213.9865	1
C19H20F14	682.1108	1
(CF2)nCHF(	-	1
C8HF17O3S	499.9375	84
C8F17KO3S	537.8934	47
C8F17LiO3S	505.9457	36
C16H20F17	629.0892	26
C8H4F17N(	516.964	17
C8F17O3S	498.9302	16
C12H12F17	605.0165	13
C13H12F17	585.0266	9
C30H48F17	825.3083	9
C8F17NaO3	521.9194	7
C16F34Mg(	1021.844	5
C16H20F17	629.0892	4

C24H36F17	741.2144	3
(CF2)nFHO	-	1
C5HF11	269.9903	1
-	-	-
-	-	-
-	-	-
-	-	-
-	-	-
-	-	-
-	-	-
C5HF9O5	311.968	2
C7H2F14O	463.9399	2
C7H2F14O	463.9399	1
-	-	-
-	-	-
-	-	-
-	-	-
-	-	-
-	-	-
-	-	-
C5H3F3S	151.9908	2
C5H3F3S	151.9908	2
C3HF7O	185.9916	1
C3HF7O	185.9916	1
C3HF7	169.9966	39
C3HF7	169.9966	9
C3F7	168.9888	3
C16H31F7F	490.1085	2
C3AgF7	275.8939	1
C3F7IZn	359.8224	1
-	-	-
-	-	-
C5HF11O	285.9852	18
C5HF11O	285.9852	7
-	-	-
-	-	-
-	-	-
C5HF9O4	295.9731	6
-	-	-
-	-	-
C6H4F11N(	347.0016	21
C6HF11O3	329.975	14
C6HF11O3	329.975	5
C6F11KO3	367.9309	3
C6HF11O3	329.975	1
-	-	-
C8HF15O4	477.9356	2

-	-	-
C4HF7O3	229.9814	8
C4HF7O3	229.9814	4
C4HF9O	235.9884	13
C4HF9O	235.9884	3
C4HF9O	235.9884	2
C4F9KO	273.9442	1
C6HF11O4	345.9699	4
-	-	-
-	-	-
-	-	-
-	-	-
-	-	-
-	-	-
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C7H2F12O3	361.9812	1
C8HF15O4S	477.9356	2
-	-	-
-	-	-
-	-	-
-	-	-
-	-	-
C5HF9O3	279.9782	5
C5HF9O3	279.9782	4
C5HF9O3	279.9782	2
-	-	-
-	-	-
C7HF13O7	443.9515	2
-	-	-
C3HF7O	185.9916	1
C3HF7O	185.9916	1
-	-	-
-	-	-
-	-	-
-	-	-
-	-	-
C7HF13O5S	443.9337	2
-	-	-
C6H2F10O2	295.9895	4
C6H2F10O2	295.9895	1
C6H2F10O2	295.9895	1
-	-	-
-	-	-
C4HF9O4S	315.9452	5

C4F9KO4S	353.9011	3
C4F9NaO4S	337.9271	3
-	-	-
-	-	-
-	-	-
-	-	-
-	-	-
C7HF13O5	411.9616	6
-	-	-
-	-	-
C7H2F14O5	463.9399	2
C7H2F14O5	463.9399	1
-	-	-
-	-	-
C6H2F10O3	311.9844	1
C6H2F10O3	311.9844	1
C6H2F10O3	311.9844	1
C6H2F10O3	311.9844	1
C6H2F10O3	311.9844	1
-	-	-
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-	-	-
-	-	-
-	-	-
-	-	-
-	-	-
-	-	-
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C6H4F11NO	347.0016	21
C6HF11O3	329.975	14
C6HF11O3	329.975	5
C6F11KO3	367.9309	3
C6HF11O3	329.975	1
C8HF15O3	429.9686	4
C8HF15O3	429.9686	2
C8HF15O3	429.9686	1
-	-	-
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Compound	MS1 Comp	Mass	Mass Defe	Retention T	Formula	Score	Downstrea	Downstrea
8	(526.9574,	263.983	0.983	2.112	C5HF9O2	87.87	475443	479333
13	(218.9862,	219.9933	0.9933	2.112	C4HF9	87.94	188550	186792
20	(626.9553,	313.9803	0.9803	3.129	C6HF11O2	87.72	1	1
24	(506.9234,	507.9302	0.9302	1.691	C8HF11N3(	99.3	123401	123966
27	(426.9569,	213.9871	0.9871	1.014	C4HF7O2	84.6	101023	104576
38	(498.9295,	499.9366	0.9366	4.711999	C8HF17O3S	98.28	21383	21966
40	(268.9826,	269.9902	0.9902	3.132	C5HF11	87.46	1	1
46	(622.9279,	623.9349	0.9349	2.857	C13HF17O9	95.59	4.67E+07	4.74E+07
47	(296.946, 1	595.9069	0.9069	1.047	C9HF19NO	98.47	2.41E+07	2.44E+07
48	(754.9113,	755.9186	0.9186	3.993	C15HF21O1	95.92	1.10E+07	1.14E+07
49	(658.9419,	659.9491	0.9491	3.301	C15HF21O5	94.97	8623775	8758179
51	(926.8723,	463.9386	0.9386	3.922	C10HF13O4	95.88	6893698	7000858
52	(644.9111,	645.9188	0.9188	2.873	C13HF19O6	89.89	6629912	6830686
53	(611.9272,	612.9344	0.9344	1.045	C10H9F12C	97.59	6427296	6449398
54	(310.9611,	311.9686	0.9686	2.862	C5HF9O5	99.47	6586903	6762269
55	(926.8723,	463.9391	0.9391	4.035	C7H2F14O5	98.12	4468358	4562471
56	(886.8955,	887.9034	0.9034	4.731	C14H2F26C	99.74	1920297	1980926
57	(396.9402,	397.9477	0.9477	3.852	C6H2F12O4	98.34	1943535	1981885
58	(474.9521,	475.9597	0.9597	3.403	C8H5F13O6	98.99	1650174	1681813
59	(346.9438,	347.9512	0.9512	2.507	C4HF9O8	77.91	1795315	1784648
60	(266.9574,	535.9269	0.9269	2.045	C9H5F13O1	99.18	1456760	1473889
61	(680.9254,	681.9324	0.9324	3.301	C11HF19N2	99.13	1333354	1374786
62	(474.9524,	475.9598	0.9598	3.228	C8H5F13O6	99.44	1341068	1355810
64	(150.9825,	151.9897	0.9897	2.87	C5H3F3S	71.25	1306963	1341051
65	(184.9843,	185.9917	0.9917	5.028	C3HF7O	99.93	1339702	1345596
67	(168.9893,	169.9965	0.9965	3.301	C3HF7	88.05	1371779	1353170
68	(298.9446,	299.952	0.952	1.047	C6H2F6O5S	87.76	1172497	1188579
69	(250.9764,	251.984	0.984	3.663001	C4HF9O2	99.34	1134946	1144691
70	(284.9781,	285.9853	0.9853	3.297	C5HF11O	87.86	1094947	1103242
71	(364.9348,	365.9421	0.9421	3.468	C5HF11O4S	78.04	1074486	1086525
73	(269.9588,	541.9316	0.9316	2.138	C9H2F16O6	99.06	820848	815566
74	(776.8949,	777.9031	0.9031	3.993	C15HF23O8	90.83	771929	874099
75	(590.9386,	295.9738	0.9738	2.763	C5HF9O4	99.19	619486	626790
76	(266.9568,	535.9267	0.9267	1.964	C9H5F13O1	98.81	681725	669121
80	(261.9462,	525.9063	0.9063	0.992	C7H3F13O8	99.33	740591	735992
82	(328.9679,	329.9755	0.9755	3.301	C6HF11O3	87.61	680824	720039
83	(943.8996,	944.9076	0.9076	3.922	C12HF30N5	96.18	421986	428906
84	(476.9283,	477.9354	0.9354	4.543	C8HF15O4S	99.49	475877	480076
85	(926.91, 57	927.9177	0.9177	1.043	C12H3F27N	97.32	329462	330632
86	(458.9551,	229.9818	0.9818	0.976	C4HF7O3	98.05	309185	314678
88	(234.981, 8	235.9887	0.9887	2.342	C4HF9O	88	475582	469013
91	(690.9324,	345.9701	0.9701	3.663001	C6HF11O4	99.79	244932	253912
92	(492.9233,	493.9307	0.9307	4.515999	C8HF15O5S	99.55	383435	386893
94	(526.9285,	527.9358	0.9358	3.515	C8H3F15O1	99.33	400418	390514
96	(396.9411,	397.9484	0.9484	3.25	C6H2F12O4	99.75	335494	330211
97	(854.9375,	427.9719	0.9719	4.512001	C4HF13N6(	96.58	287088	287504

98	(943.8994,	944.9063	0.9063	4.035	C13HF28N8	98.23	283675	290881
102	(506.9216,	507.9284	0.9284	1.072	C8H2F14O7	97.67	261396	244891
103	(412.9358,	413.9428	0.9428	3.492	C6H2F12O5	99.67	290960	294002
104	(558.9493,	559.9562	0.9562	2.347	C10H2F18O	99.24	284054	284738
105	(564.9041,	565.9121	0.9121	1.111	C9HF19O2S	94.94	327944	336666
106	(494.9523,	991.9217	0.9217	5.029	C15H2F28N	99.27	241902	254362
107	(360.9736,	361.9812	0.9812	3.681	C7H2F12O3	99.01	274793	283972
108	(476.9282,	477.9354	0.9354	4.656	C8HF15O4S	99.34	237470	238864
109	(640.9359,	641.9431	0.9431	3.258	C11H2F20O	99.06	242563	239257
110	(316.9677,	317.975	0.975	4.418	C5HF11O3	87.79	261060	263991
111	(562.9114,	563.919	0.919	2.134	C8H7F11N2	61.08	188810	186736
112	(854.9373,	427.9734	0.9734	3.881	C8H2F14O4	99.5	231327	238411
113	(606.9341,	607.9414	0.9414	2.78	C10H2F18O	98.64	232207	223845
114	(278.9709,	279.9784	0.9784	2.342	C5HF9O3	87.83	220939	220121
116	(350.9696,	351.9771	0.9771	4.510001	C6HF13O2	87.79	231188	231920
117	(281.961, 8	565.936	0.936	2.064	C10H4F14O	95.88	190525	193474
118	(442.9443,	443.9504	0.9504	4.749	C7HF13O7	86.68	189338	199774
119	(722.9221,	361.9649	0.9649	3.881	C6HF11O5	87.42	164834	167752
122	(184.9842,	185.9918	0.9918	0.974	C3HF7O	87.97	203613	198207
124	(672.9075,	673.9152	0.9152	3.322	C11H2F20O	99.56	161147	176817
127	(422.9399,	423.9469	0.9469	0.908			169989	175142
128	(458.9375,	459.9446	0.9446	4.364999	C8H2F14O4	97.98	153613	154475
129	(672.9078,	673.9148	0.9148	3.426	C11H2F20O	98.85	112643	114877
130	(370.9555,	371.9634	0.9634	3.297	C7H3F5NO	84.58	150870	156255
132	(360.9434,	361.9508	0.9508	0.967	C5H3F9O6S	98.02	134967	135959
133	(442.9261,	443.9332	0.9332	4.251999	C7HF13O5S	98.74	141795	140772
134	(426.9652,	427.9725	0.9725	3.951	C8H2F14O4	86.91	127666	128252
136	(294.9819,	295.99	0.99	2.316	C6H2F10O2	87.37	139777	140503
140	(380.9052,	381.9126	0.9126	3.592	C5HClF10O	98.34	145358	138102
141	(506.9217,	507.9293	0.9293	0.986	C8H2F14O7	98.06	98799	106023
143	(314.9381,	315.9449	0.9449	2.712	C4HF9O4S	78.05	129183	128285
145	(1018.8791	509.943	0.943	5.223	C9H2F16O4	74.75	70966	75034
146	(310.9433,	311.9502	0.9502	2.574	C5HF9O3S	78.31	135773	134045
148	(456.9248,	457.9321	0.9321	0.961	C10HF11O6	75.19	102021	105231
151	(346.9442,	347.9511	0.9511	2.791	C4HF9O8	80.11	115398	114390
152	(346.9439,	347.9515	0.9515	2.667	C5H2F10O4	77.82	113798	112958
153	(822.916, 3	411.9615	0.9615	4.412	C7HF13O5	87.04	82090	83595
155	(358.9447,	359.9517	0.9517	2.584	C6H2F10O4	98.96	101675	105680
156	(281.9629,	565.9369	0.9369	1.975	C8H5F13N2	97.14	73274	78999
159	(462.9322,	463.9397	0.9397	4.389	C7H2F14O5	97.48	85995	92407
161	(246.9507,	247.9579	0.9579	0.923	C3H2F6O4S	93.28	81933	84727
163	(548.9687,	549.9751	0.9751	4.772999	C10H10F14	97.94	59596	57365
165	(310.9771,	311.9845	0.9845	2.325	C6H2F10O3	87.6	87472	85734
166	(348.9414,	349.949	0.949	2.507	C7H2F8O5S	72.57	86032	83979
167	(718.9433,	719.9504	0.9504	3.728	C11H3F17N	83.52	69601	74367
168	(772.919, 1	773.9261	0.9261	4.154	C14H2F22N	74.55	71211	75508
171	(574.9461,	575.9538	0.9538	3.862999	C10H5F17O	98.91	67660	66862

172	(528.9545,	529.9615	0.9615	5.243	C7HF13N5(	82.56	71869	75460
173	(640.9348,	641.9426	0.9426	2.794	C14HF19O1	84.7	50694	52870
175	(412.9358,	413.943	0.943	3.681	C6H2F12O5	99.54	67704	68647
176	(482.9766,	483.9821	0.9821	4.146	C7H7F9N9S	97.54	44302	48276
177	(821.9021,	822.9107	0.9107	1.072	C14H9F18(	98.58	69364	72459
178	(481.9769,	482.9844	0.9844	2.706	C9H8F11N(	97.85	61752	60337
180	(525.9488,	526.9561	0.9561	2.229	C10H8F11N	95.16	65699	64404
181	(458.9571,	459.9646	0.9646	3.728	C8H5F13O5	98.95	60938	69229
182	(328.9677,	329.9746	0.9746	4.245999	C6HF11O3	86.97	66561	67160
183	(428.9613,	429.9682	0.9682	4.563	C8HF15O3	84.38	60914	61741
184	(508.9523,	509.9592	0.9592	2.099	C9H2F16O(	86.24	64093	64444
186	(426.9324,	427.9392	0.9392	4.177001	C7HF13O4S	95.81	53884	54257
187	(481.9764,	482.9838	0.9838	2.509	C11H10F7(	98.2	53579	53960
188	(268.9651,	539.9362	0.9362	2.152	C9H6F10N2	62.58	52932	49766
190	(532.9878,	533.9951	0.9951	4.510001	C11H11F11	96.9	46849	40818
191	(366.9319,	367.9389	0.9389	3.466	C7HF9O5S	73.9	53453	51572
192	(458.9573,	459.9638	0.9638	3.581	C8H5F13O5	98.5	49592	49615
193	(718.9429,	719.9506	0.9506	3.806	C10H5F17N	65.89	39141	38946
194	(438.9845,	439.9916	0.9916	3.416	C7H3F13N3	87.1	42220	41634
196	(772.9196,	773.9265	0.9265	3.974	C11H6F18N	65.13	36415	36110
197	(614.9601,	615.9671	0.9671	5.22	C11H13F11	95.85	34014	34926
198	(456.9557,	457.963	0.963	4.625	C9HF15O4	86.15	36778	37931
199	(574.9455,	575.9538	0.9538	3.959	C8H2F14N6	98.58	31954	28918
200	(628.9159,	629.923	0.923	2.783	C12H7F11N	63.75	33422	34801
201	(662.9183,	663.9251	0.9251	3.256	C10HF17N2	82.94	29661	32564
202	(383.9789,	384.9866	0.9866	0.952	C7H7F8NO	91.03	29484	29293

Downstrea	Downstrea	Downstrea	Upstream_	Upstream_	Upstream_	Upstream_	Upstream_	Downstrea
518468	498432	511659	108693	110716	114212	112410	112848	4.97E+05
197391	199418	209238	47174	48072	47351	47363	51331	1.96E+05
1	1	1	129331	131243	133452	134456	132827	1.00E+00
73094	66014	45312	23530	23100	18201	25142	26267	8.64E+04
106834	112293	109650	20898	21063	22384	21799	20833	1.07E+05
23333	22165	24705	61735	63881	36875	64151	67629	2.27E+04
1	1	1	51673	53516	49935	56274	56217	1.00E+00
4.99E+07	4.97E+07	4.95E+07	1	1	1	1	1	4.86E+07
2.47E+07	2.42E+07	2.45E+07	1	1	1	1	1	2.44E+07
1.19E+07	1.17E+07	1.17E+07	1	1	1	1	1	1.15E+07
9140374	9076449	8929166	1	1	1	1	1	8.91E+06
7181519	7179518	7207891	10679	1	10957	1	1	7.09E+06
7102419	7163942	7272219	1	1	1	1	1	7.00E+06
6493521	6501250	6561621	1	1	1	1	1	6.49E+06
7037287	7002966	7118705	1	1	1	1	1	6.90E+06
4705168	4676024	4712333	10679	1	10957	1	1	4.62E+06
2076225	2055930	2090638	1	1	1	1	1	2.02E+06
2031345	1985861	2027813	1	1	1	1	1	1.99E+06
1686338	1681405	1731695	1	1	1	1	1	1.69E+06
1832358	1806335	1849481	1	1	1	1	1	1.81E+06
1521578	1534029	1582304	1	1	1	1	1	1.51E+06
1432478	1432511	1429296	1	1	1	1	1	1.40E+06
1398775	1395651	1415624	1	1	1	1	1	1.38E+06
914935	1397163	1442534	1	1	1	1	1	1.28E+06
1380999	1378656	1375585	1	1	1	1	1	1.36E+06
1290535	1359401	1381333	1	1	1	1	1	1.35E+06
1208202	1191697	1178561	1	1	1	1	1	1.19E+06
1157293	1152165	1161509	1	1	1	1	1	1.15E+06
1113333	1127956	1152956	1	1	1	1	1	1.12E+06
1108636	1103347	1120945	1	1	1	1	1	1.10E+06
870332	868370	875733	1	1	1	1	1	8.50E+05
904415	857528	854204	1	1	1	1	1	8.52E+05
672007	663788	691132	1	1	1	1	1	6.55E+05
733315	726536	705331	1	1	1	1	1	7.03E+05
735187	734974	758066	1	1	1	1	1	7.41E+05
740594	739873	732108	1	1	1	1	1	7.23E+05
428137	439013	446724	1	1	1	1	1	4.33E+05
496652	491295	503496	1	1	1	1	1	4.89E+05
411828	369283	333988	1	1	1	1	1	3.55E+05
356458	342955	347098	1	1	1	1	1	3.34E+05
505949	486502	474168	1	1	1	1	1	4.82E+05
273575	263757	273169	1	1	1	1	1	2.62E+05
396699	399396	394653	1	1	1	1	1	3.92E+05
407403	414519	418181	1	1	1	1	1	4.06E+05
364166	367866	362368	1	1	1	1	1	3.52E+05
308930	305989	305944	1	1	1	1	1	2.99E+05

294321	300389	303642	1	1	1	1	1	2.95E+05
277373	260443	261185	1	1	1	1	1	2.61E+05
302953	299352	296468	1	1	1	1	1	2.97E+05
303561	292661	295512	1	1	1	1	1	2.92E+05
317030	333123	340218	1	1	1	1	1	3.31E+05
269941	269331	268678	1	1	1	1	1	2.61E+05
288849	286781	290572	1	1	1	1	1	2.85E+05
243948	263790	243315	1	1	1	1	1	2.45E+05
219839	235430	220491	1	1	1	1	1	2.32E+05
266419	266263	269568	1	1	1	1	1	2.65E+05
211174	205467	213066	1	1	1	1	1	2.01E+05
241203	246100	253109	1	1	1	1	1	2.42E+05
237900	244144	229551	1	1	1	1	1	2.34E+05
241064	241739	231668	1	1	1	1	1	2.31E+05
236622	242488	245803	1	1	1	1	1	2.38E+05
199582	203963	192915	1	1	1	1	1	1.96E+05
222789	214697	211025	1	1	1	1	1	2.08E+05
172991	170212	184806	1	1	1	1	1	1.72E+05
226939	212928	208022	1	1	1	1	1	2.10E+05
164487	179994	194313	1	1	1	1	1	1.75E+05
183897	186054	186170	1	1	1	1	1	1.80E+05
157301	161739	160264	1	1	1	1	1	1.57E+05
148771	157028	125620	1	1	1	1	1	1.32E+05
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145462	146186	150891	1	1	1	1	1	1.43E+05
139765	144465	146519	1	1	1	1	1	1.43E+05
134681	138486	138917	1	1	1	1	1	1.34E+05
145384	145711	145291	1	1	1	1	1	1.43E+05
142509	147178	141166	1	1	1	1	1	1.43E+05
102028	110351	131128	1	1	1	1	1	1.10E+05
132512	135625	132833	1	1	1	1	1	1.32E+05
80478	76869	79614	1	1	1	1	1	7.66E+04
138538	120734	121182	1	1	1	1	1	1.30E+05
106682	111005	121062	1	1	1	1	1	1.09E+05
115855	120838	121535	1	1	1	1	1	1.18E+05
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91633	90500	85683	1	1	1	1	1	8.67E+04
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101617	93562	92984	10679	1	10957	1	1	9.33E+04
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68925	99603	90348	1	1	1	1	1	7.52E+04
91698	92030	87581	1	1	1	1	1	8.89E+04
87624	84591	88621	1	1	1	1	1	8.62E+04
77863	80519	81210	1	1	1	1	1	7.67E+04
72951	74447	72536	1	1	1	1	1	7.33E+04
74153	72007	76949	1	1	1	1	1	7.15E+04

77130	76062	74553	1	1	1	1	1	7.50E+04
57166	67727	54774	1	1	1	1	1	5.66E+04
70724	71931	76762	1	1	1	1	1	7.12E+04
56663	59612	58282	1	1	1	1	1	5.34E+04
57882	75770	56694	1	1	1	1	1	6.64E+04
62478	64063	63867	1	1	1	1	1	6.25E+04
68354	69652	67508	1	1	1	1	1	6.71E+04
62513	66140	64143	1	1	1	1	1	6.46E+04
69861	69199	66411	1	1	1	1	1	6.78E+04
65361	64088	59953	1	1	1	1	1	6.24E+04
59874	65335	61910	1	1	1	1	1	6.31E+04
55288	56289	58150	1	1	1	1	1	5.56E+04
55335	54947	55238	1	1	1	1	1	5.46E+04
56730	57526	53188	1	1	1	1	1	5.40E+04
52196	46517	45184	1	1	1	1	1	4.63E+04
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39283	39393	38627	1	1	1	1	1	3.91E+04
41879	41331	44116	1	1	1	1	1	4.22E+04
36760	38322	37707	1	1	1	1	1	3.71E+04
39021	38183	34053	1	1	1	1	1	3.60E+04
39319	38010	37605	1	1	1	1	1	3.79E+04
35940	36127	34623	1	1	1	1	1	3.35E+04
32944	36977	33243	1	1	1	1	1	3.43E+04
29975	30127	32390	1	1	1	1	1	3.09E+04
31047	33812	31443	1	1	1	1	1	3.10E+04

Upstream_	log2 Fold C	p-val (unad	Signif? (Bo
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48258.2	2.02	4.02E-10	TRUE
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1	14.92	2.52E-10	TRUE

Best Database Match
Perfluoropentanoic acid
1H-Nonafluorobutane
Perfluorohexanoic acid
-
Perfluorobutanoic acid
Perfluorooctanesulfonic acid
1,1,1,2,2,3,4,4,5,5,5-Undecafluoropentane
-
-
-
-
-
-
-
Perfluoro-3,5,7-trioxaoctanoic acid
Perfluoro-2-{[perfluoro-3-(perfluoroethoxy)-2-propanyl]oxy}ethanesulfonic acid
-
-
-
-
-
-
-
2-(Trifluoromethyl)thiophene
Perfluoropropanol
2H-Perfluoropropane
-
-
Perfluoro-3-(1H-perfluoroethoxy)propane
-
-
-
Perfluoro-3,6-dioxaheptanoic acid
-
-
Ammonium perfluoro-2-methyl-3-oxahexanoate
-
6-Oxoperfluorooctanesulfonic acid
-
Perfluoro-3-methoxypropanoic acid
Nonafluoro-tert-butanol
Perfluoro-3,6-dioxaoctanoic acid
-
-
-
-

-
-
-
-
-
-
2,3,3,4,4,5,5,6,6,7,7,7-Dodecafluoro-2-hydroxyheptanoic acid
6-Oxoperfluorooctanesulfonic acid
-
-
-
-
-
Perfluoro(4-methoxybutanoic) acid
-
-
Perfluoro-3,5,7,9,11-pentaoxadodecanoic acid
-
Perfluoropropanol
-
#N/A
-
-
-
-
Perfluoro-3,6-dioxa-4-methyl-7-octene-1-sulfonic acid
-
6-H-Perfluorohexanoic acid
-
-
Perfluoro(2-ethoxyethane)sulfonic acid
-
-
-
-
-
Perfluoro-3,6,9-trioxadecanoic acid
-
-
Perfluoro-2-{[perfluoro-3-(perfluoroethoxy)-2-propanyl]oxy}ethanesulfonic acid
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2,2,3-Trifluoro-3-(heptafluoropropoxy)propanoic acid
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Ammonium perfluoro-2-methyl-3-oxahexanoate
Perfluoro-2-[(perfluoropentyl)oxy]propanoic acid
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