TN-0151

Analysis of PFAS in Drinking Water by EPA Method 533: A Direct Comparison of the Accuracy and Precision of Manual and Automated SPE Sample Preparation

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Introduction

Per- and polyfluoroalkyl substances (PFAS) are a group of nearly 5000 synthetic compounds that have been used in firefighting foam, chrome-plating, waterproof textiles, and other Teflon products. They have attracted particular attention due to their toxicity, persistence, and prevalence in the environment. Alpha Analytical Labs, a leading environmental lab, has been utilizing PromoChrom's fully automated 8-Channel solid phase extraction (SPE) system, SPE-03, to analyze PFAS in drinking water and other matrices. This technical note presents the results of using the SPE-03 and draws comparisons with manual extraction.

EPA Method 533 is a complement to EPA Method 537.1 for analyzing short-chain and more polar PFAS compounds in drinking water. It covers a list of 25 PFAS compounds using isotope dilution and anion-exchange SPE[1]. The extraction procedure requires meticulous handling, proper control of flow rates, and many different conditioning, elution, and rinsing steps.

This technical note demonstrates the full automation of EPA Method 533 cartridge extraction using SPE-03 and uses manual extraction results as performance reference. Both extractions were performed using the Phenomenex Strata-X-AW 33 µm Polymeric Weak Anion SPE Cartridge. It includes data on background, detection limit, accuracy, reproducibility and matrix effects. Results were taken from field sample extractions performed by Alpha Analytical Labs.

Manual SPE Protocol

SPE Cartridge:		Strata-X-AW 33 µm Polymeric Weak Anion; 500 mg/6 mL (Part No. 8B-S038-HCH)
Cartridge Preconditioning:	**Rinse:**	Cartridge with 2 aliquots of 5 mL Methanol with 2 % Ammonium Hydroxide.
	Rinse:	Cartridge with 10 mL of Methanol.
	Rinse:	Cartridge with 10 mL of 0.1 M Phosphate Buffer.
	Rinse:	Cartridge with 15 mL of DI Water
Sample Extraction:	**Load:**	Sample at a rate of 5-10 mL/min.
	Rinse:	Bottle and cartridge with 10 mL of 1 g/L Ammonium Acetate in reagent water.
	Rinse:	Cartridge with 1 mL Methanol.
	Air Dry:	Cartridge for 5-10 minutes.
Cartridge Elution:	**Rinse:**	Bottle with 2 washes 5 mL of Methanol with 2 % Ammonium Hydroxide.
	Elute:	Sample from cartridge with previous rinses.

Automated SPE Protocol

Instrumentation:	PromoChrom Technologies SPE-03 Automated SPE System with MOD-004 (sample bottle rinsing) and MOD-005 (minimal Teflon option)
Sample Volume:	250 mL
SPE Cartridge:	Strata™-X-AW 33 µm Polymeric Weak Anion; 500 mg/6 mL (Part No. 8B-S038-HCH)
Solvent 1:	Methanol
Solvent 2:	Milli-Q® water
Solvent 3:	0.1 M Phosphate Buffer
Solvent 4:	1 g/L Ammonium Acetate
Solvent 5:	Methanol with 2 % Ammonium Hydroxide
Sample Extraction:	See Table 1

Table 1. PromoChrom SPE-03 Extraction Procedure

Step	Action	Inlet	Flow Rate (mL/min)	Volume (mL)
1	Elute W2	Solvent 1	10	10
2	Elute W1	Solvent 3	10	10
3	Elute W1	Solvent 3	10	3
4	Elute W1	Solvent 5	10	3
5	Add Samp W1	Sample	5	270
6	Rinse W1	Solvent 4	5	10
7	Shake	Time Based	-	30 sec
8	Clean	Solvent 1	5	1
9	Add Samp W2	Sample	5	9
10	Blow N2	Time Based	-	5 min
11	Rinse 1	Solvent 5	2	5
12	Rinse 1	Solvent 5	2	5
13	Shake	Time Based	-	10 sec
14	Collect 1	Sample	2	4.5

Results and Discussion

Data was collected from field sample extraction batches between March and April 2021. As required by EPA Method 533, each field sample batch must be extracted alongside QC samples consisting of a Lab Reagent Blank (LRB), Lab Fortified Blank (LFB), Laboratory Fortified Sample Matrix (LFSM), Laboratory Fortified Sample Matrix Duplicate (LFSMD) or Field Duplicate (FD). All QC samples are rotated between different channels of the SPE-03 and vacuum manifold for each extraction.

EPA Method 533 requires background to be <1/3 MRL, which corresponds to 0.67 ng/L for Alpha Analytical Labs. Analyzing the LRBs from 7 separate extraction batches, both manual and SPE-03 setup demonstrated clean Background (**Figure 1**). Any detected compounds >0.1 ng/L were seen on both setups, suggesting contamination from other sources. **Figure 2** shows the corresponding isotope dilution analog (labeled compound) recoveries of the blanks showing good extraction performance. The higher MPFDoA recoveries is attributed to the method quantifying the 12- carbon compound with an 8-carbon M2PFOA performance standard. A more similar internal standard should produce better accuracy.

The SPE-03 demonstrated average recoveries between 95 % to 110 %, compared to manual extraction recoveries between 90 % to 120 %. Method requirements are 70 % to 130 % (**Figure 3**). NFDHA is known to behave differently than its corresponding M5PFHxA isotope but was seen to have better accuracy on the SPE-03. Since the analyte recoveries are corrected through isotope dilution, the actual extraction performance can be determined by looking at the recoveries of the labeled compounds. All labeled compounds recovered above 90 % on the SPE-03 and follow a similar trend as manual extraction (**Figure 4**). Method limits are 50 % to 200 %.

The response of the 3 fluorotelomers were enhanced, with M2-4:2FtS having a significant increase of about 30 %. This is quite commonly seen in field samples, especially for non-potable water applications. In Wellington Labs' reference guide[2], it mentions that matrix effects can have a considerable impact on the ionization of fluorotelomer sulfonic acids. This phenomenon is speculated to be independent of the extraction. The only other compound that recovered higher was M2PFOA and is interesting to note that these are the only 4 isotopes that have 2 C13 atoms. Later eluting compounds like MPFDoA can stick to the sorbent and particles in dirtier samples, resulting in lower recoveries. The effects again are more significant on non-drinking water applications, but the corresponding methods have the flexibility to increase elution volume. To confirm whether the matrix effects are independent of the SPE-03, **Figure 5** compares the labeled recoveries of 20 fields samples extracted on the manifold vs SPE-03. Both systems had similar trends with higher perceived recoveries for the fluorotelomers and M2PFOA, as well as reduced recoveries for MPFDoA. The %RSD of labeled compounds in field samples also show strong agreement between both setups (**Figure 6**). As discussed, fluorotelomers are more susceptible to matrix enhancement which explains the higher variation.

The primary barrier to the automation of sample preparation is the high capital cost of the instrumentation which runs in the tens of thousands of dollars, nearly 10X higher than a manual SPE manifold and vacuum pump. At a certain level of sample throughput, the total cost of the automated method (including capital depreciation) becomes lower than that of the manual method on a total $/sample basis. (Note that the consumable consumption costs of the two approaches are virtually identical on a $/sample basis). Therefore, high throughput laboratories tend to migrate to the automated solution and lower throughput laboratories are comfortable staying with the manual method. Each laboratory must make an analysis and buying decision based upon their specific circumstances. However, in either case, the laboratory can be assured that, whether manual or automated, the Strata™-X-AW SPE cartridges will provide accurate and precise analytical data.

Figure 1. Background Levels of Manual vs SPE-03 Extraction

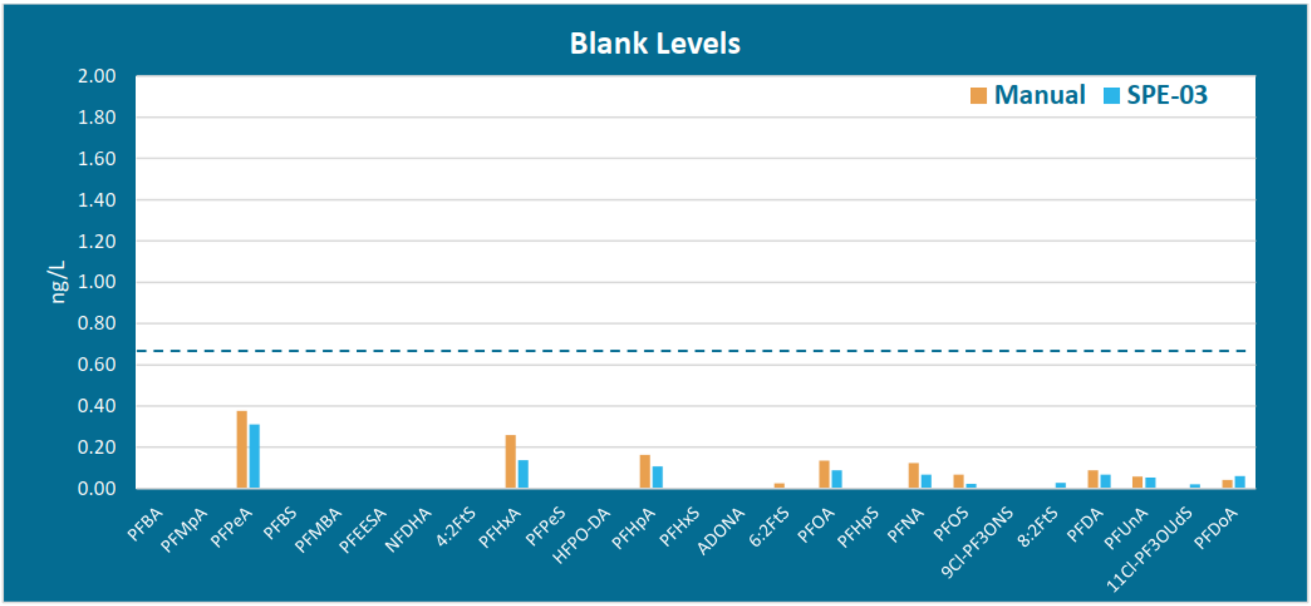

Figure 2. Isotope Dilution Analog Recoveries of the Blanks

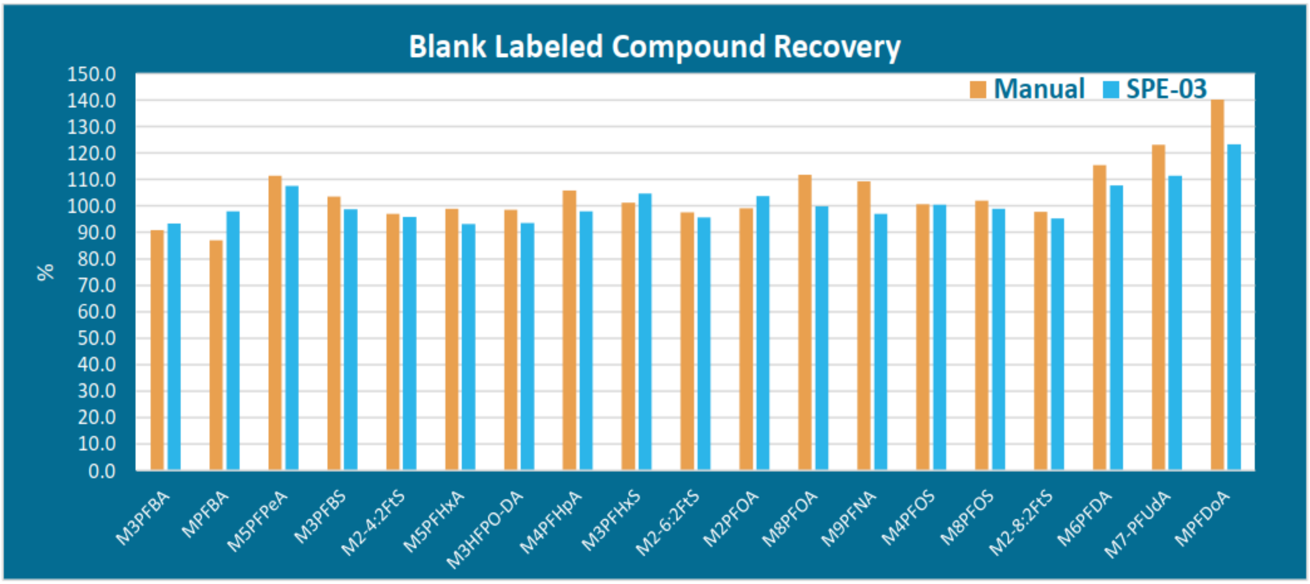

Figure 3. Average Recoveries of Manual vs SPE-03 Extraction

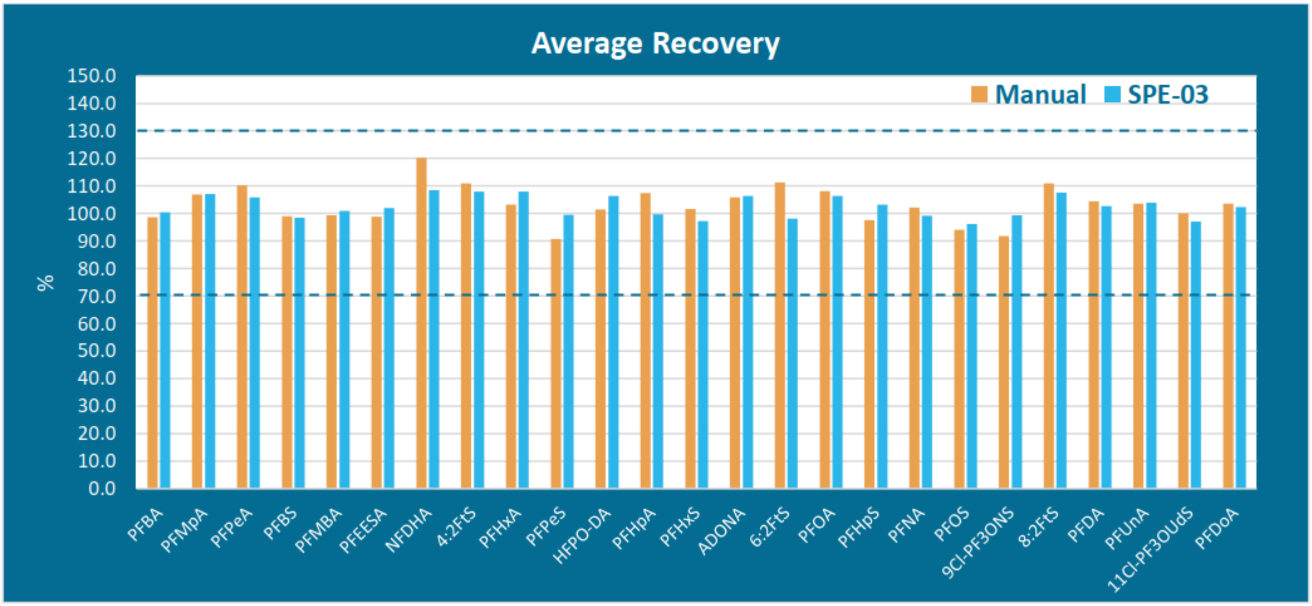

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Figure 4. Average Labeled Compound Recoveries of Manual vs SPE-03 Extraction

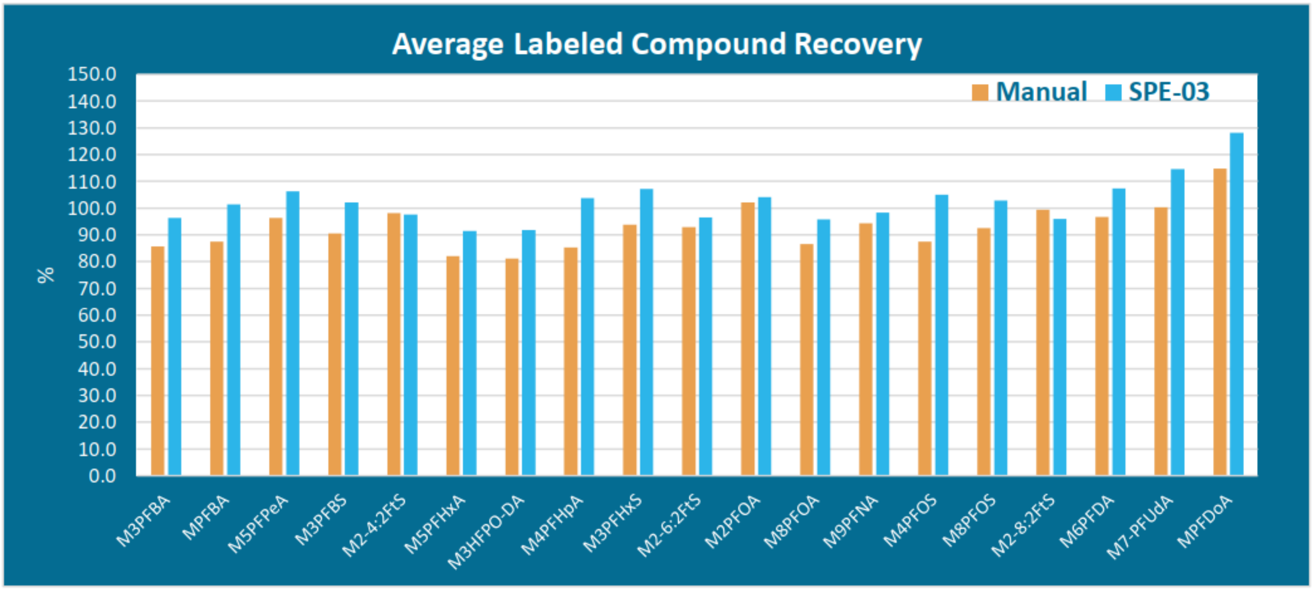

Figure 5. Average Labeled Compound Recoveries from Field Samples of Manual vs SPE-03 Extraction

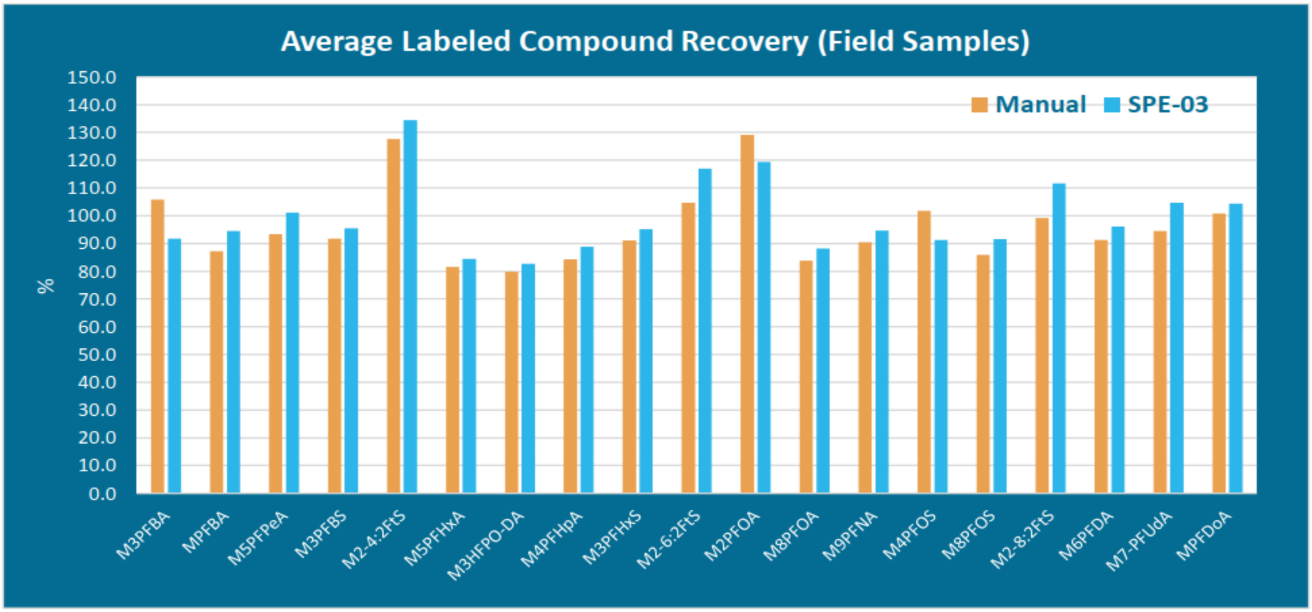

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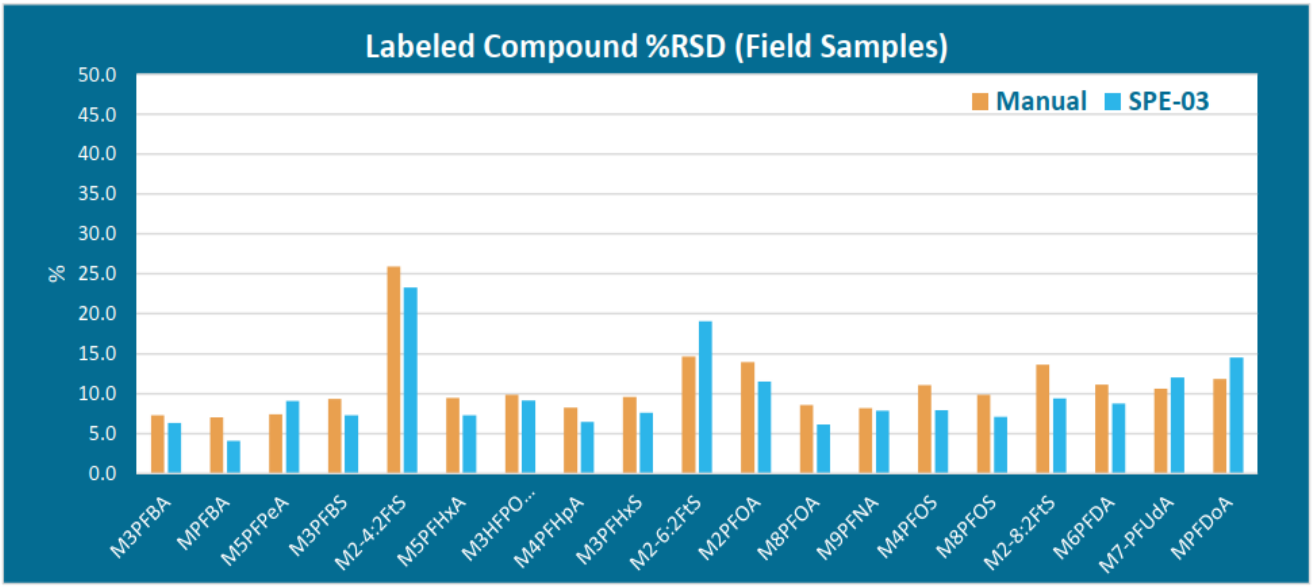
Figure 6. Labeled Compound %RSD from Field Samples of Manual vs SPE-03 Extraction



Conclusions

Both the manual and the automated sample preparation techniques, when diligently applied, are shown to be acceptable, analytically-equivalent approaches to the performance of EPA method 533. The decision of which technique to apply is primarily an economic choice which balances an individual laboratory's need for high sample throughput (with higher capital cost, but lower per sample labor cost) against lower sample throughput (with lower capital cost, but higher per sample labor cost). Whichever approach is chosen, Strata™-X-AW cartridges are the logical choice for EPA method 533. Phenomenex SPE cartridges have proven themselves to be the reliable "work horse" of the PFAS testing industry as demonstrated by the successful analysis of hundreds of thousands of environmental PFAS samples.

References

1. EPA Method 533
   https://www.epa.gov/sites/production/files/201912/documents/method-533-815b19020.pdf
2. Wellington Laboratories
   https://welllabs.com/docs/pfc_reference_handling_guide.pdf

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Strata™-X-AW Ordering Information

Strata			
Format	**Sorbent Mass**	**Part Number**	**Unit**
Tube			
	30 mg	8B-S038-TAK**	1 mL (100/box)
	30 mg	8B-S038-TBJ	3 mL (50/box)
	60 mg	8B-S038-UBJ	3 mL (50/box)
	100 mg	8B-S038-EBJ	3 mL (50/box)
	100 mg	8B-S038-ECH	6 mL (30/box)
	200 mg	8B-S038-FBJ	3 mL (50/box)
	200 mg	8B-S038-FCH	6 mL (30/box)
	500 mg	8B-S038-HBJ	3 mL (50/box)
	500 mg	8B-S038-HCH	6 mL (30/box)
Giga Tube			
	500 mg	8B-S038-HDG	12 mL (20/box)
	1 g	8B-S038-JDG	12 mL (20/box)
	1 g	8B-S038-JEG	20 mL (20/box)
	5 g	8B-S038-LFF	60 mL (16/box)
96-Well Plate			
	10 mg	8E-S038-AGB	2 Plates/Box
	30 mg	8E-S038-TGB	2 Plates/Box
	60 mg	8E-S038-UGB	2 Plates/Box
96-Well Microelution Plate			
	2 mg	8M-S038-4GA	ea

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Product	Part	Volume	Concentration
EPA 533 mix	AL0-101838	1 mL	2 µg/mL in Methanol
EPA 537.1 mix	AL0-101839	1mL	2 µg/mL in Methanol
EPA 533 + 537.1 mix	AL0-101840	1 mL	2 µg/mL in Methanol

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Luna™ Omega Column 3 µm PS C18 50 x 3 mm	00B-4758-Y0
Kinetex™ EVO Column 5 µm C18 100 x 2.1 mm	00D-4633-AN
Strata PFAS (WAX/GCB) SPE 200 mg, /50 mg, /6mL tubes, 30/pk	CS0-9207
Strata SDB-L 500 mg/6mL tubes, 30/pk	8B-S014-HCH
Verex™ Vial, 9 mm Screw, PP, 1.7 mL, 1000/pk	AR0-39P0-13
Verex Vial, 9 mm Screw, PP, 300 µL, 1000/pk	AR0-39P2-13
Verex Vial, 9 mm Screw, PP, 700 µL, 1000/pk	AR0-39P1-13
Vial Cap Verex Cert+ Cap (one piece), 9 mm, PE w/ Starburst pre-Slit, 2mL, 1000/pk	AR0-89P6-13-C

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