Application

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ePrep | Application Note 2025

Automated Sample Preparation for PFAS in Soils with analysis on Agilent LC/TQ 6495D

vortex extraction and no blowdown

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Figure 1 ePrep ONE Sample Preparation Workstation and Agilent LC/TQ 8495D

SUMMARY

This application note presents a fully automated workflow for extraction and sample preparation of PFAS from soils down to 100ppt using the ePrep ONE workstation. The Workflow delivers weighed soil samples through sample preparation to output autosampler vials ready for analysis with no manual intervention. Results demonstrate that this Workflow meets the performance requirements specified in EPA Method 1633.

A range of soil samples were tested from zero organic sand to high organic Peat. The ePrep ONE Workflow demonstrated excellent recoveries and reproducibility of PFAS compounds and Extracted Internal Standards across the soils range.

Performance Summary

- Test Range: soils tested LOQ =0.25 ng/g and LOD = 0.08 ng/g using Agilent LC/TQ 6495D
- Extraction: Efficient Vortex Mixing which can achieve 75% reduction in extraction time.
- Direct to output vial: No blowdown
- Precision: Consistent automated handling reduces variability
- Carryover Control: Dedicated syringes and automated washing to eliminate cross-contamination and carryover

- Clean: LC ready sample through Carbo SPE cleanup cartridge
- Reliability: Integrated workflow validation

INTRODUCTION

PER- AND POLYFLUOROALKYL SUBSTANCES (PFAS) "forever chemicals" persist indefinitely in global soil and water systems, requiring sensitive detection methods. Traditional soil analysis uses labor-intensive SPE and filtration processes that consume significant lab resources.

The ePrep ONE automated workstation transforms PFAS workflows through its PFAS-free design with all Teflon removed from syringes, vials, and consumables to eliminate background contamination. High-precision analytical syringes deliver chromatography-grade accuracy throughout sample preparation, while integrated high-energy vortex mixing effectively breaks apart soil aggregates for maximum extraction efficiency. Built-in SPE cleanup streamlines preparation protocols while reducing operational costs.

This Application Note demonstrates how ePrep ONE effectively extracts trace PFAS from soil matrices while simplifying workflows and enhancing laboratory productivity.

SAMPLES

A range of classified soils were used for testing extraction recoveries using the ePrep ONE. These soils represent the typical samples observed in environmental laboratories for PFAS analysis. Soils selected were classified Reference Standards or Materials supplied with a Certificate of Analysis.

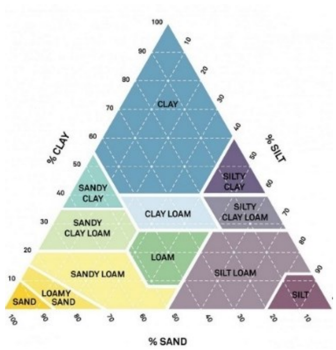

Figure 2 Soil Categorization

Table 1 Soils Types

Type	Supplier No.	Description
Ottawa Sand (20-30 mesh)	MSX00751	Moisture ≤0.1%, Particle 20-40 mesh
Top Soil Mix		1-6% Organic
Clean Clay Loam	CLNSOIL2	10% Clay, 50% Sand, 40% Silt,
Clean Clay	CLNSOIL5	95% Clay, 3.5% Sand, 1.5% Silt, <0.01% Organic
Clean Loam Soil	CLNLOAM6	20% Clay, 40% Sand, 40% Silt ≤4% Organic
Humic Peat		≥90% Organic

EXPERIMENTAL

ePrep ONE Configuration

The ePrep ONE was configured per Figure 3 below. Solvent dispenser, Luer Manifold, Vials and Syringes were all Teflon Free (PFAS-Free) for the entire Workflow.

A full list of accessories and consumables can be found at the end of this document. A method Standard Operating Procedure (SOP) is also available from ePrep on request.

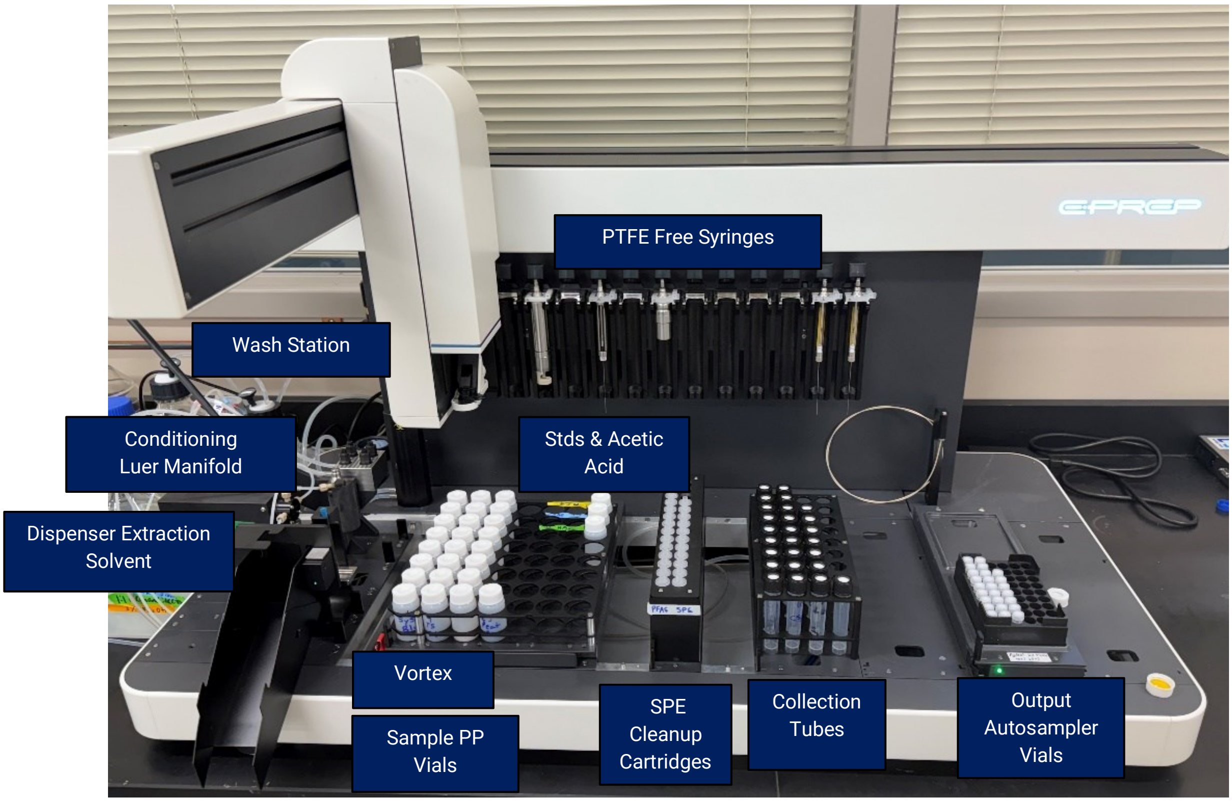

Figure 3 ePrep ONE PFAS Soils Configuration

HPLC Conditions

Table 2 LC Conditions

Instrument: Agilent LC/TQ 6495D with Hybrid Multisampler (DEAG800112)
Column: Poroshell Aq-C18 2.7um, 2.1x150mm
Flowrate: See Below
Injection vol: 10 µL, feed mode
Column temp: 45°C
Mobile phase A: 2mM ammonium acetate
Mobile phase B: 95:5 Acetonitrile: H2O
Gradient: 13.1 minutes total runtime

Table 3 Gradient Conditions

Time (min)	A. Conc %	B. Conc %	Flow mL/min	Max Pressure (bar)
0	100	0	0.4	800
2	70	30	0.4	800
8	40	60	0.4	800
11	0	100	0.4	800
13.1	100	0	0.4	800

TQ Conditions

Table 4 TQ Conditions

Model: 6495D TQ
Polarity: Negative
Gas Flow: 18 L/mini
Sheath Gas Flow: 11 L/min
Nozzle Voltage: 0
Gas Temperature: 200°C
Sheath Gas Temperature: 300°C
Nebulizer: 30psi

Automated ePrep ONE Extraction WORKFLOW

The following describes the workflow for the preparation of all soil types. It offers unattended processing after weighing of 2g soil samples into 20 mL PP (polypropylene) extraction vials with options to go direct to output autosampler vial with acidification. Alternatively, blowdown and reconstitution can be added to the workflow if ultra low sensitivity is required.

Table 5 ePrep PFAS Soils Workflow (without blowdown)

- 2g of Soil set in 20mL PP vials
- Spike with IS and PFAS mix

- Pause for 5 minutes (Equilibration time)
- Add 15mL of 3% NH4OH in 50:50 Methanol/Acetone extraction solvent

- Vortex for 15 minutes at 800 rpm
- Pause for 15 minutes (Settling time)

- Condition SPE Cartridges (5mL MeOH followed by 2.5mL Extraction Solvent)
- Aliquot 6mL of sample extract through carbo cartridges

- Elute carbo cartridges with 4mL of 3% NH4OH Acetonitrile.
- Votex to homogenise.

- Aliquot 150uL to Output Autosampler Vial
- Neutralize with Acetic Acid

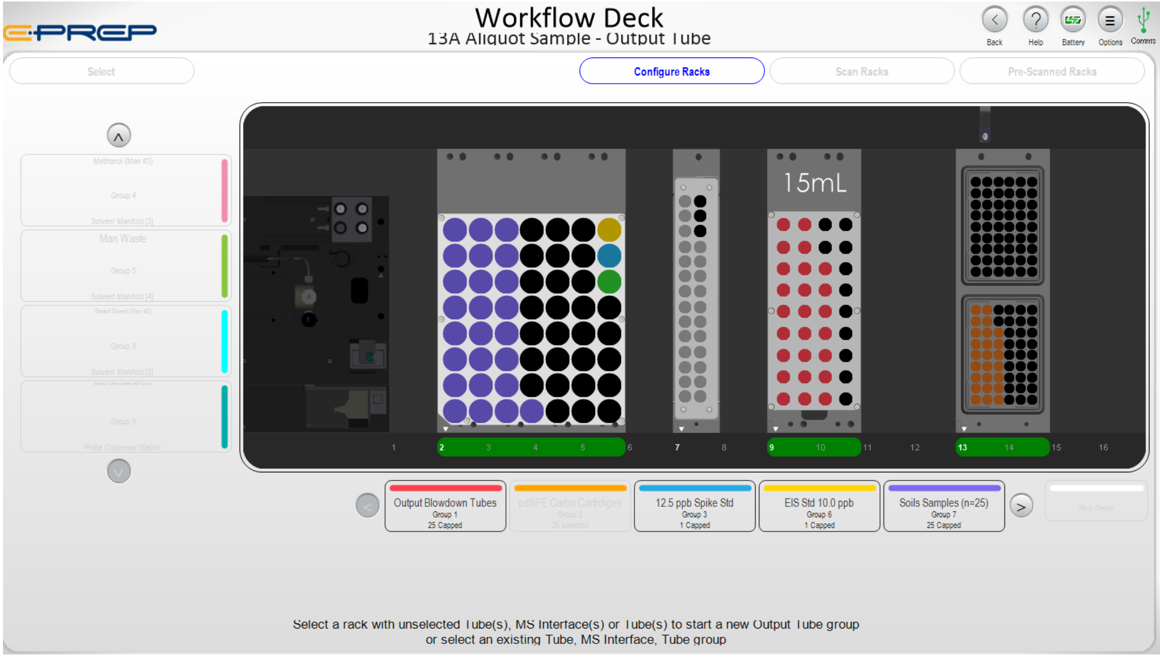

Figure 4 ePrep ONE Software Deck Layout (screen capture)

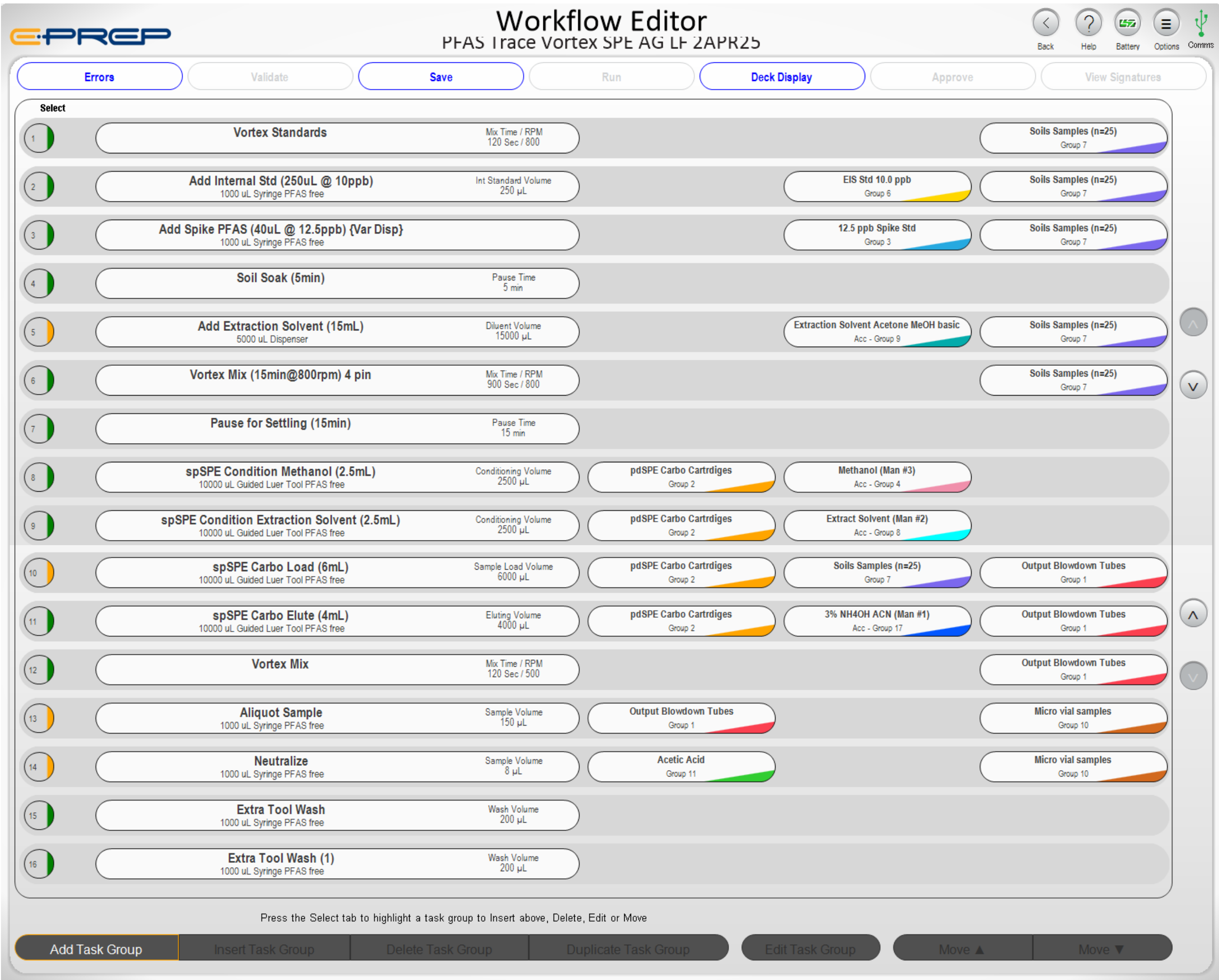

Figure 5 ePrep ONE Software Workflow (screen capture)

FEATURES OF THE WORKFLOW

Syringe Management

The ePrep ONE system utilizes RFID-identified syringes to ensure precise reagent handling and prevent cross-contamination. If necessary, up to 12 different syringes can be integrated into a single workflow, allowing for optimized volume and dedicated syringes for specific solutions

Enhanced Extraction Efficiency

The workflow uses vortex mixing for PFAS extraction from soil matrices, with programmable speed and timing parameters to optimize both extraction efficiency and processing time. The ePrep LA Vortex achieves equivalent extraction performance in 15 minutes compared to traditional 1-hour tumbling methods, representing a 75% reduction in extraction time.

Integrated SPE Cleanup

The system automatically cleans and elutes the extracted sample through a luer-connected Carbo SPE cartridge incorporated into the workflow to remove LC-interfering compounds.

Solvent Management System

The integrated solvent manifold enables direct delivery of multiple solvents to luer syringes, significantly accelerating syringe operation and improving workflow efficiency.

Contamination Prevention

Cross-contamination is prevented through comprehensive automated washing protocols using a positive flow Wash-station and Dual step washing combining Wash Station and manifold ports

Workflow Validation

The software includes built-in solution validation. Before running, it will calculate required solution volumes and compares them against selected reservoirs providing feedback to the user if validation is not achieved.

RESULTS AND DISCUSSION

Results demonstrate excellent performance of the ePrep ONE system for PFAS soil sample preparation across multiple validation parameters, including instrument calibration linearity, extraction efficiency in environmental samples, and matrix-normalized spike recoveries spanning diverse soil types.

Instrument Calibration Curves

The PFAS calibration curves met all acceptance criteria and were within established specifications. Correlation coefficients (r^2) for all PFAS analytes exceeded 0.985, and the relative standard deviation for replicate injections was less than 15% across a 0.1-10ng/mL range. The calibration standards demonstrated linear response within the specified concentration range, with accuracy and precision meeting method requirements.

Figure 6 Calibration Curves for PFAS Analytes 0.01-10ng/mL

%Recovery PFAS Extracted Internal Standard (EIS) in different soil types - 5ppt

Raw 5ppt (8327) PFAS EIS recoveries for all sample types tested were satisfactory and met EPA Method 1633 acceptance criteria. Raw recovery percentages for EIS standard compounds for all soil types were approximately 70-130% . These were consistently within the acceptable EPA 1633 range of 50-150%, demonstrating adequate ePrep Workflow performance across all soil types.

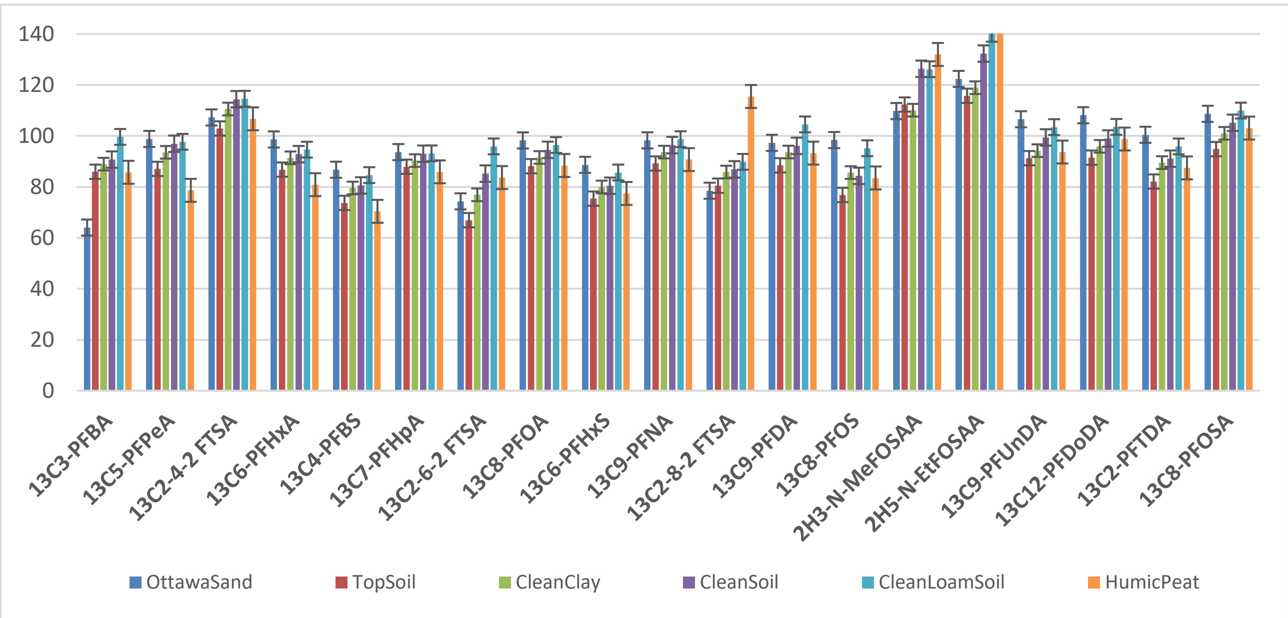

Figure 7 Recoveries from different Soil Types (EIS at 5ppt)

%RSD recovery, PFAS EIS @ 5 ppt in different soil types

Although EPA Method 1633A relies on recovery percentage ranges rather than %RSD criteria for EIS compounds, ePrep validation studies achieved exceptional precision performance. Reproducibility assessments (n=3) across diverse soil matrices yielded %RSD values typically below 10%, including results for the most analytically challenging PFAS EIS compounds and complex soil types.

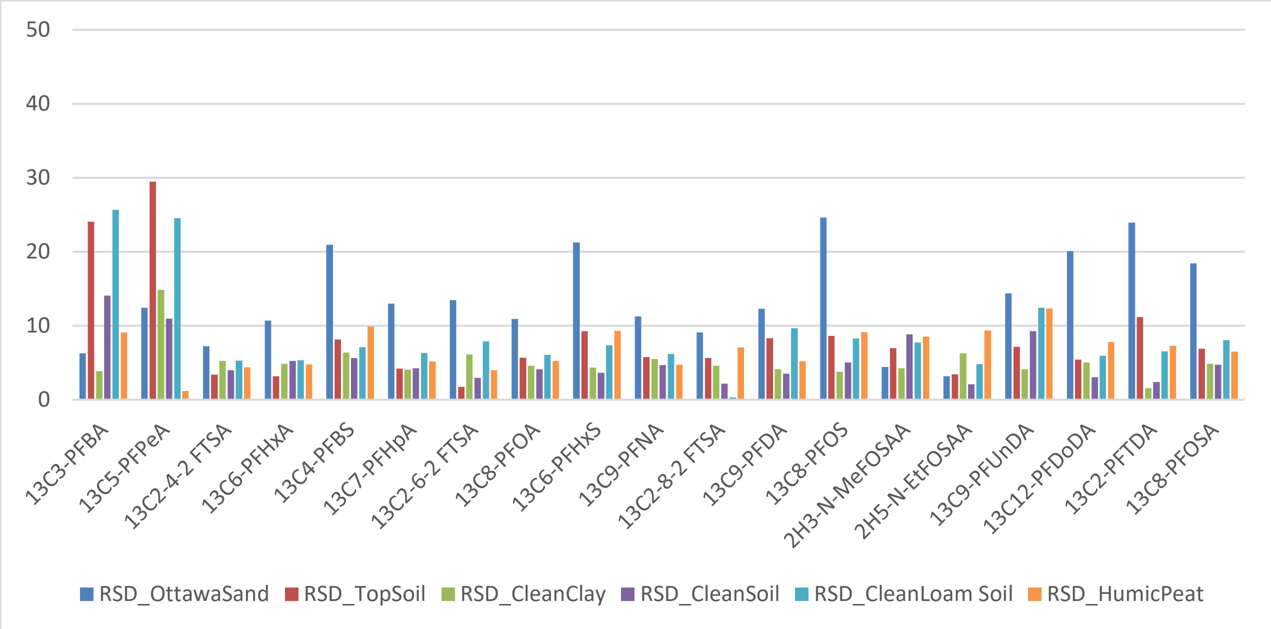

Figure 8 %RSD Recoveries n=3 form different Soil types (EIS at 5ppt)

%Recovery PFAS Spikes in different soil types - 0.25 ng/g spike

PFAS recoveries (EIS normalized) from diverse soil matrices spiked at 0.25 ng/g demonstrated excellent method performance across all soil types tested. Recovery percentages consistently fell within acceptable ranges, with minimal matrix interference observed regardless of soil composition, organic carbon content, or texture. The robust extraction and cleanup procedures effectively overcame potential matrix effects, yielding reliable and reproducible results at this low fortification level. These excellent recoveries confirm the method's suitability for trace-level PFAS analysis in various soil environments, including sandy, clay, and organic-rich matrices.

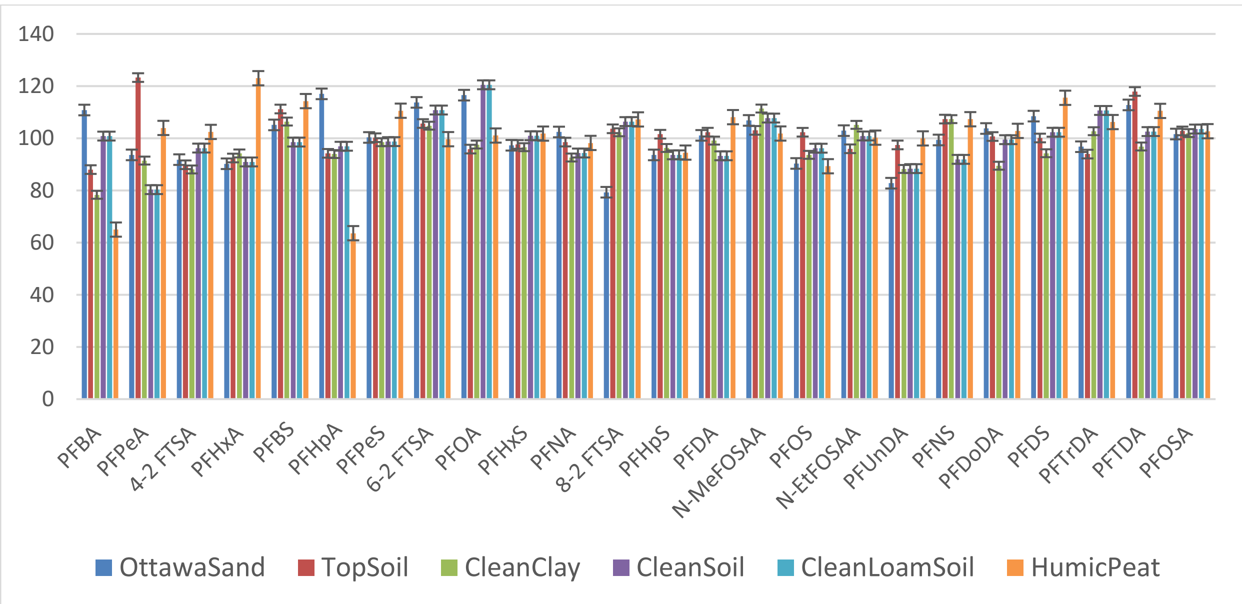

Figure 9 Data showing the percentage recovery of 0.25ng/g PFAS Spikes in different Soil Types

Recovery %RSD of spiked soil matrix

Again, reproducibility at very low concentration for a n=3 sample set across diverse soil matrices yielded EIS normalized %RSD values around 10%. It should be noted that an acetic acid contamination caused high PFBA %RSD on same samples.

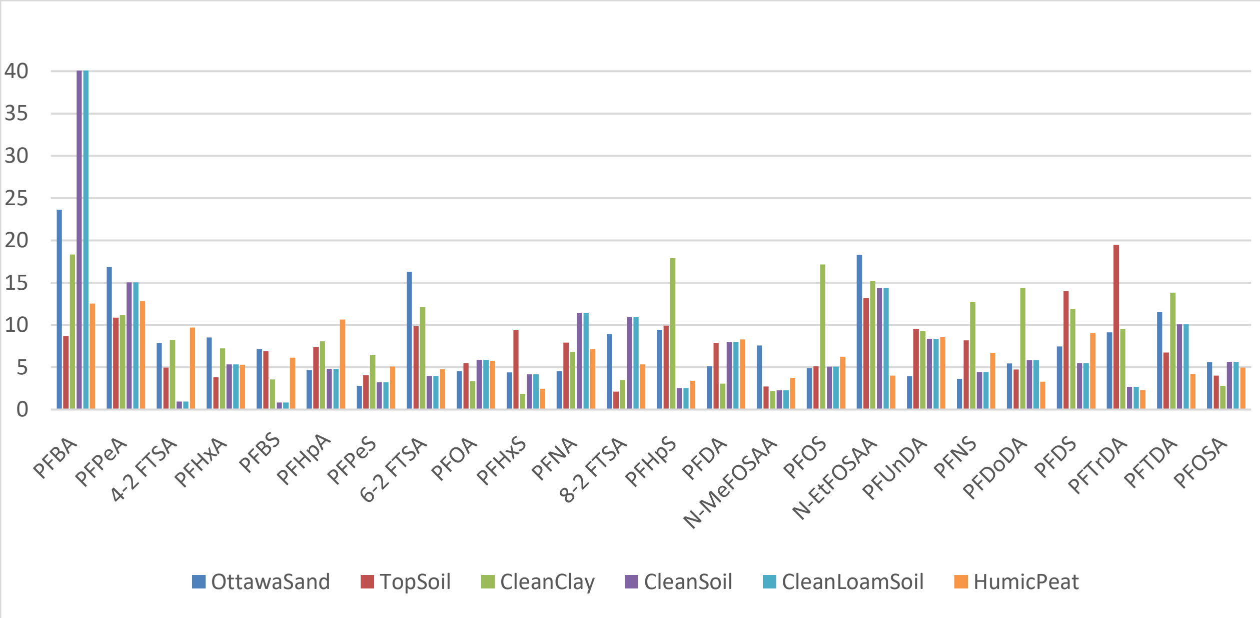

Figure 10 %RSD Recoveries for 0.25ng/g PFAS Spikes in different Soil Types

CONCLUSION

The ePrep ONE workstation provides a fully automated, high-throughput solution for PFAS extraction from soil samples. This integrated workflow processes up to 56 soil samples unattended, handling all steps from weighing through autosampler vial preparation while eliminating post-extraction solvent evaporation requirements.

Validation across diverse soil matrices—ranging from sandy soils to high-organic peat—demonstrates consistent analytical performance. PFAS compound recoveries of 80-120% across different soil types confirm the method delivers reliable, precise results suitable for quantitative analysis.

The validated workflow fully meets EPA Method 1633 performance criteria, providing environmental laboratories with a robust automated solution for regulatory PFAS monitoring. This approach significantly enhances laboratory productivity while maintaining the analytical quality required for compliance testing.

APPLICATION PRODUCT LIST

Description	Part No
ePrep One Sample Preparation Workstation	01-01000
Accessories and Adapter Plates	
4P Vortex Stirrer	01-04105
Probe Dispenser, PTFE Free	01-04304
spSPE Rack	01-04184
Luer Enabling Kit	01-04160
Solvent manifold, Teflon Free	01-04205
4P Vortex Adapter Plate for 20-40 mL Enviro Vials	01-03002
Adapter Plate for 10mL/15mL Centrifuge Tubes	01-03094
Teflon Free Syringes	
100 µL ePrep (PFAS Free) syringe, Cone 80 mm Removable needle	01-09046
1 ml ePrep (PFAS Free) syringe, Cone 80 mm Removable needle	01-09047
10 ml ePrep (PFAS Free) syringe, Guided Luer	01-09091
Consumables	
20mL PP container (PFAS Free)	01-03140
Polyimide lined Caps for 20mL PP container (PFAS Free)	01-18002
10mL V-Base tubes, {Thermo P/N LBS1004 (Box 1000)}	See OEM
Polyimide Lined Caps - 10mL V-Base tubes {ALWSCI, PART No. C0003182 (Pkt 100)}	n/a
SPE cartridge S*Pure, 220mg carbo {S*Pure P/N 8186557 (100pkt}}	n/a
ePrep Needles for SPE cartridge	01-10996
ePrep Aspiration Needles (Spare)	01-10995

APPENDIX 1

dMRM transitions

Compound name	Compound formula	Precursor m/z	Product m/z	RT (min)	RT window (min)	Fragmentor (V)	CAV (V)	CE (V)
2H3-N-MeFOSAA	C11H3D3F17NO4S	573	482.9	8.35	2	166	2	12
2H3-N-MeFOSAA	C11H3D3F17NO4S	573	419	8.35	2	166	2	20
2H5-N-EtFOSAA	C12H3D5F17NO4S	589	531	8.64	2	166	2	20
2H5-N-EtFOSAA	C12H3D5F17NO4S	589	419	8.64	2	166	2	20
4-2 FTSA	C6H5F9O3S	327	307	4.83	2	166	2	19
4-2 FTSA	C6H5F9O3S	327	81	4.83	2	166	2	39
6-2 FTSA	C8H5F13O3S	427	81	6.3	2	166	2	40
6-2 FTSA	C8H5F13O3S	427	80	6.3	2	166	2	48
8-2 FTSA	C10H5F17O3S	527	507	7.75	2	166	2	31
8-2 FTSA	C10H5F17O3S	527	80	7.75	2	166	2	51
13C2D4-4-2 FTSA	C4[13C]2H5F9O3S	333	312	4.83	2	166	2	19
13C2D4-4-2 FTSA	C4[13C]2H5F9O3S	333	81.9	4.83	2	166	2	39
13C2D4-6-2 FTSA	C6[13C]2H5F13O3S	433	412	6.3	2	166	2	27
13C2D4-6-2 FTSA	C6[13C]2H5F13O3S	433	82	6.3	2	166	2	40
13C2D4-8-2 FTSA	C8[13C]2H5F17O3S	533	512	7.75	2	166	2	31
13C2D4-8-2 FTSA	C8[13C]2H5F17O3S	533	82	7.75	2	166	2	43
13C2-PFTDA	C12[13C]2HF27O2	715	670	10.36	2	166	4	12
13C2-PFTDA	C12[13C]2HF27O2	715	169	10.36	2	166	4	36
13C3-PFBA	C[13C]3HF7O2	216	172	3.88	1	166	2	7
13C4-PFBA	[13C]4HF7O2	217	172	3.88	2	166	2	7
13C4-PFBS	[13C]4HF9O3S	303	99	5.19	2	166	2	36
13C4-PFBS	[13C]4HF9O3S	303	80	5.19	2	166	2	43
13C5-PFPeA	[13C]5HF9O2	268	223	4.41	2	166	2	7
13C6-PFHxA	[13C]6HF11O2	319	273.9	5.09	2	166	2	7
13C6-PFHxA	[13C]6HF11O2	319	121	5.09	2	166	2	23
13C6-PFHxS	[13C]6HF13O3S	405	99	6.84	2	166	3	43
13C6-PFHxS	[13C]6HF13O3S	405	80	6.84	2	166	3	48
13C7-PFHpA	[13C]7HF13O2	370	325.4	5.86	2	166	2	7
13C7-PFHpA	[13C]7HF13O2	370	172	5.86	2	166	2	19
13C8-PFOA	[13C]8HF15O2	421	376	6.63	2	166	2	7
13C8-PFOA	[13C]8HF15O2	421	172	6.63	2	166	2	19
13C8-PFOS	[13C]8HF17O3S	507	99	8.36	2	166	4	48
13C8-PFOS	[13C]8HF17O3S	507	80	8.36	2	166	4	52
13C8-PFOSA	[13C]8H2F17NO2S	506	78	10.27	2	166	3	36
13C9-PFDA	C[13C]9HF19O2	521	476	8.09	2	166	2	8
13C9-PFDA	C[13C]9HF19O2	521	222	8.09	2	166	2	16
13C9-PFNA	[13C]9HF17O2	472	427	7.37	2	166	2	8
13C9-PFNA	[13C]9HF17O2	472	223	7.37	2	166	2	16
13C9-PFUnDA	C2[13C]9HF21O2	572	527	8.78	2	166	2	11
13C9-PFUnDA	C2[13C]9HF21O2	572	273	8.78	2	166	2	19
13C12-PFDoDA	[13C]12HF23O2	625	580	9.45	2	166	4	11
13C12-PFDoDA	[13C]12HF23O2	625	274	9.45	2	166	4	20
N-EtFOSAA	C12H8F17NO4S	584	526	8.66	2	166	2	20
N-EtFOSAA	C12H8F17NO4S	584	482.9	8.66	2	166	2	12
N-EtFOSAA	C12H8F17NO4S	584	419	8.66	2	166	2	20
N-MeFOSAA	C11H6F17NO4S	570	512	8.36	2	166	2	19
N-MeFOSAA	C11H6F17NO4S	570	482.9	8.36	2	166	2	12
N-MeFOSAA	C11H6F17NO4S	570	419	8.36	2	166	2	20
PFBA	C4HF7O2	213	169	3.88	2	166	2	7
PFBS	C4HF9O3S	298.9	99	5.19	2	166	2	36
PFBS	C4HF9O3S	298.9	80	5.19	2	166	2	43
PFDA	C10HF19O2	513	469	8.09	2	166	2	8

PFDA	C10HF19O2	513	269	8.09	2	166	2	16
PFDA	C10HF19O2	513	219	8.09	2	166	2	16
PFDoDA	C12HF23O2	613	569	9.45	2	166	4	11
PFDoDA	C12HF23O2	613	319	9.45	2	166	4	20
PFDoDA	C12HF23O2	613	169	9.45	2	166	4	28
PFDS	C10HF21O3S	598.9	99	9.71	2	166	4	55
PFDS	C10HF21O3S	598.9	80	9.71	2	166	4	63
PFHpA	C7HF13O2	363	319	5.86	2	166	2	7
PFHpA	C7HF13O2	363	169	5.86	2	166	2	19
PFHpS	C7HF15O3S	448.9	99	7.62	2	166	3	44
PFHpS	C7HF15O3S	448.9	80	7.62	2	166	3	48
PFHxA	C6HF11O2	313	269	5.09	2	166	2	7
PFHxA	C6HF11O2	313	119	5.09	2	166	2	23
PFHxS	C6HF13O3S	398.9	99	6.84	2	166	3	43
PFHxS	C6HF13O3S	398.9	80	6.84	2	166	3	48
PFNA	C9HF17O2	463	419	7.37	2	166	2	8
PFNA	C9HF17O2	463	219	7.37	2	166	2	16
PFNA	C9HF17O2	463	169	7.37	2	166	2	20
PFNS	C9HF19O3S	548.9	99	9.06	2	166	4	51
PFNS	C9HF19O3S	548.9	80	9.06	2	166	4	59
PFOA	C8HF15O2	413	369	6.63	2	166	2	7
PFOA	C8HF15O2	413	169	6.63	2	166	2	19
PFOS	C8HF17O3S	498.9	99	8.36	2	166	4	48
PFOS	C8HF17O3S	498.9	80	8.36	2	166	4	52
PFOSA	C8H2F17NO2S	497.9	169	10.27	2	166	3	34
PFOSA	C8H2F17NO2S	497.9	78	10.27	2	166	3	36
PFOSA	C8H2F17NO2S	497.9	48	10.27	2	166	3	80
PFPeA	C5HF9O2	263	219	4.41	2	166	2	7
PFPeA	C5HF9O2	263	69	4.41	2	166	2	40
PFPeS	C5HF11O3S	348.9	99	6.03	2	166	2	36
PFPeS	C5HF11O3S	348.9	80	6.03	2	166	2	40
PFTDA	C14HF27O2	712.9	369	10.37	2	166	4	24
PFTDA	C14HF27O2	712.9	219	10.37	2	166	4	28
PFTDA	C14HF27O2	712.9	169	10.37	2	166	4	36
PFTrDA	C13HF25O2	663	619	9.99	2	166	4	11
PFTrDA	C13HF25O2	663	319	9.99	2	166	4	20
PFTrDA	C13HF25O2	663	169	9.99	2	166	4	32
PFUnDA	C11HF21O2	563	519	8.78	2	166	2	11
PFUnDA	C11HF21O2	563	319	8.78	2	166	2	16
PFUnDA	C11HF21O2	563	269	8.78	2	166	2	19

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