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Analysis of per- and polyfluoroalkyl substances in aqueous samples by SPE and LC-MS/MS according to EPA Draft Method 1633

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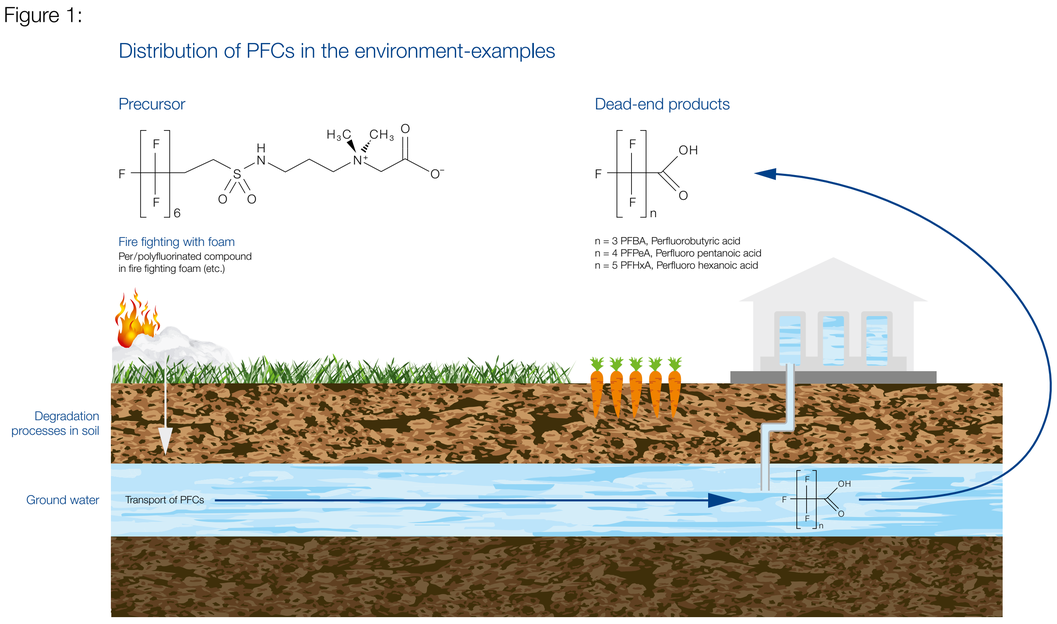
Introduction

In September 2021, the United States Environmental Protection Agency (US EPA) has published a draft method for the analysis of per- and polyfluoroalkyl substances (PFAS) [1]. The draft method is a single laboratory validated method to test for 40 PFAS compounds in a diverse range of environmental matrices including wastewater, surface water, groundwater, soil, biosolids, sediment, landfill leachate, and fish tissue. The guideline can be used in various applications, exemplarily for use in the Clean Water Act (CWA) or the National Pollutant Discharge Elimination System (NPDES) [2]. These bio-accumulative pollutants are characterized by a linear aliphatic backbone, a high degree of fluorination, and often feature a carboxylic- or sulfonic- acid functionality. People can be exposed to PFAS in a variety of ways, including drinking water. But the exposure to PFAS can lead to adverse health effects. Many studies have examined possible connections between the level of per- and polyfluoroalkyl substances in the blood and adverse health effects in humans [3]. The research suggests that high levels of certain PFAS can lead to an increased cholesterol level, a reduced vaccine response in children, changes in liver enzymes, an increased risk of high blood pressure or pre-eclampsia in pregnant women and so on.

A SPE methodology according EPA Draft Method 1633 was developed and optimized on CHROMABOND®(R) WAX SPE column.

Figure 1:



Distribution of PFAS in the environment-examples.

Sample pretreatment

Solid phase extraction according to EPA Draft Method 1633

Sample preparation:

This method is applicable to aqueous samples containing up to 50 mg of suspended solids per sample. The procedure requires the preparation of the entire sample. Subsampling should be avoided whenever possible. Typical sample size is 500 mL.

1. Homogenize the sample by inverting the sample 3 – 4 times and allowing the sample to settle. Do not filter the sample.
2. Check that the pH is 6.5 ± 0.5. If necessary, adjust pH with 50 % formic acid or ammonium hydroxide. The extract is now ready for solid-phase extraction (SPE).
3. Add the spiking solution containing the internal standard substances to the water sample (500 mL) in the sample bottle (adding 2.5 ng of each) and mix thoroughly by shaking.

Column	CHROMABOND® WAX, 6 mL 150 mg (REF 7300011)
Conditioning	With 15 mL of 1 % methanolic ammonium hydroxide, followed by 5 mL of 0.3M formic acid. Do not allow the SPE to run dry. Discard the wash solvents.
Sample application	Add 500 mL water sample with a flow rate of 5 mL/min to the cartridge. (Do not let the sorbent material in the cartridge run dry and ensure it is always immersed in water.)
Bottle Rinse	Rinse the walls of the reservoir with 5 mL reagent water (twice) followed by 5 mL of 1:1 0.1M formic acid/methanol.
Washing step	Pass those rinses through the cartridge using vacuum. Discard the rinse solution.
Drying step	Dry the cartridge by pulling air through for 15 seconds.
Elution	Rinse the inside of the sample bottle and the SPE reservoir with 5 mL of 1 % methanolic ammonium hydroxide. Use vacuum to pull the elution solvent through the cartridge and into the collection tubes.
Clean-up	Add 25 µL of concentrated acetic acid to each sample eluted in the collection tubes and vortex to mix. Add 10 mg of car-bon (GCB) to each sample. Immediately vortex (30 seconds) and centrifuge at 2800 rpm for 10 minutes.
Membrane Filtration	Filter eluate through a syringe filter (25-mm filter, 0.2-µm nylon membrane, REF 729212) into polypropylene vial.

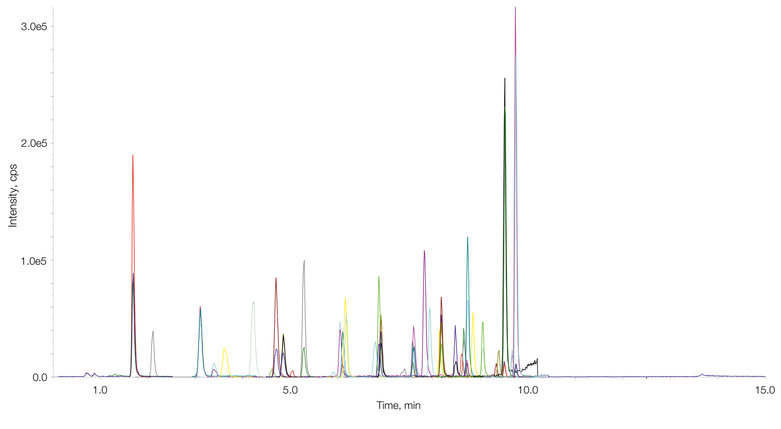
Analysis by HPLC-MS / MS

Chromatographic conditions

DELAY Column	EC 50/2 NUCLEODUR® PFAS Delay (REF 760673.20)	Acquisition mode	SRM
Column	EC 100/2 NUCLEODUR® PFAS, 3 µm (REF 760666.20)	Interface	ESI
Eluent A	5 mM ammonium acetate in water	Polarity	negative
Eluent B	5 mM ammonium acetate in methanol	Curtain Gas	30 psi
Gradient	hold 40 % B for 1 min, in 8 min from 40 % B to 95 % B, hold 95 % B for 3 min, in 0.1 min to 40 % B, hold 40 % B for 2.9 min	Collision Gas	medium
Flow rate	0.3 mL/min	Ionspray Voltage	-4500 V
Temperature	40 °C	Temperature	400 °C
Injection volume	2 µL	Ion Source Gas 1	50 psi
Acquisition mode	SRM	Ion Source Gas 2	60 psi
Interface	ESI	Detection Window	60 sec

Chromatogram

Figure 2:

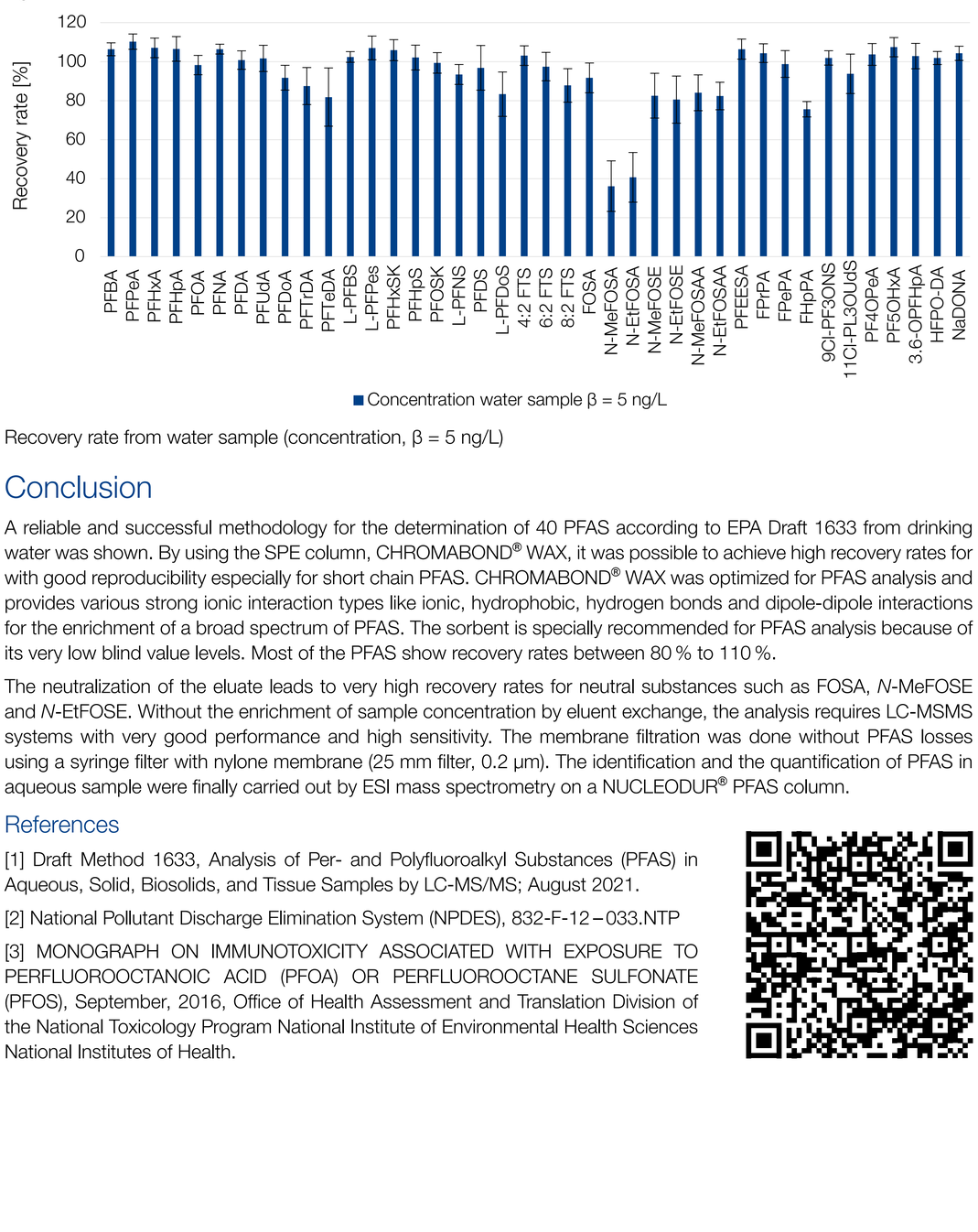


MRM transitions

Analyte	Abbreviation	CAS number	Q1 mass [Da]	Q3 mass [Da]	Retention time [min]
Perfluoro-3-methoxypropanoic acid	PFMPA	377-73-1	229.00	85.00	1.96
Perfluoro-*n*-butanoic acid	PFBA	375-22-4	212.90	168.80	2.01
3-Perfluoropropyl propanoic acid	3:3FTCA	356-02-5	241.00	177.00	3.33
Perfluoro-4-methoxybutanoic acid	PFMBA	863090-89-5	279.00	85.00	3.64
Perfluoro-*n*-pentanoic	PFPeA	2706-90-3	262.88	219.00	3.90
Perflu-oro(2-ethoxyethane)sulfonic acid	PFEESA	113507-82-7	315.00	135.00	4.14
Perfluoro-*n*-butanesulfonic acid	PFBS	375-73-5	298.93	98.90	4.20
Nonaflu-oro-3,6-dioxaheptanoic acid	NFDHA	151772-58-6	295.00	201.00	4.48
1H,1H, 2H, 2H-Perfluorohexane sulfonic acid	4:2FTS	757124-72-4	326.94	306.90	5.27
Perfluoro-*n*-hexanoic acid	PFHxA	307-24-4	312.91	268.80	5.40
Perfluoropentansulfonic acid	PFPeS	2706-91-4	348.85	80.00	5.54
Hexafluoropropylene oxide dimer acid	HFPO-DA	13252-13-6	284.99	168.70	5.77
2H,2H,3H,3H-Perfluorooctanoic acid	5:3FTCA	914637-49-3	341.00	237.00	6.33
Perfluoro-*n*-heptanoic acid	PFHpA	375-85-9	362.93	318.80	6.45
Perfluoro-*n*-hexanesulfonic acid	PFHxS	355-46-4	398.94	79.80	6.49
4,8-Dioxa-3H-perfluorononanoic acid	ADONA	919005-14-4	376.90	250.70	6.58
1H,1H, 2H, 2H-Perfluorooctane sulfonic acid	6:2FTS	27619-97-2	426.93	406.90	7.24
Perfluoro-*n*-heptanesulfonic acid	PFHpS	375-92-8	448.93	79.80	7.26
Perfluoro-*n*-octanoic acid	PFOA	335-67-1	412.91	369.00	7.26
Perfluoro-*n*-octanesulfonic acid	PFOS	1763-23-1	498.84	79.90	7.89
Perfluoro-*n*-nonanoic acid	PFNA	375-95-1	462.89	418.90	7.92
3-Perfluoroheptyl propanoic acid	7:3FTCA	812-70-4	441.00	317.00	8.00
9-Chlorohexadecafluoro-3-oxanonane-1-sulfonic acid	9Cl-PF3ONS	73606-19-6	530.75	350.70	8.25
Perfluorononanesulfonic acid	PFNS	68259-12-1	548.81	79.90	8.45
Perfluoro-*n*-decanoic acid	PFDA	335-76-2	512.84	468.90	8.49
1H,1H, 2H, 2H-Perfluorodecane sulfonic acid	8:2FTS	39108-34-4	526.00	506.80	8.50
N-methyl perfluorooctanesul-fonamidoacetic acid	*N*-MeFOSAA	2355-31-9	569.80	418.90	8.78
Perfluorooctanesulfonamide	FOSA	754-91-6	497.87	77.80	8.84
Perfluoro-*n*-decanesulfonic	PFDS	335-77-3	598.79	79.90	8.90
Perfluoro-*n*-undecanoic acid	PFUnDA	2058-94-8	562.80	518.90	8.95
N-ethyl perfluorooctanesulfo-namidoacetic acid	*N*-EtFOSAA	2991-50-6	583.81	418.80	9.02
11-Chloroeicosafluoro-3-oxaundecane-1-sulfonic acid	11Cl-PF3OUdS	763051-92-9	630.74	451.00	9.15
Perfluoro-*n*-dodecanoic acid	PFDoDA	307-55-1	612.79	568.90	9.33
N-methyl perfluorooctanesul-fonamide	*N*-MeFOSA	31506-32-8	512.00	169.00	9.55
Perfluoro-*n*-tridecanoic acid	PFTrDA	72629-94-8	662.77	618.90	9.66
Perfluorododecanesulfonic acid	PFDoS	79780-39-5	698.72	80.00	9.70
N-ethyl perfluorooctanesulfo-namide	*N*-EtFOSA	4151-50-2	526.00	169.00	9.80
N-methyl perfluorooctanesul-fonamidoethanol	NMeFOSE	24448-09-7	616.00	59.00	9.82
Perfluoro-*n*-tetradecanoic acid	PFTeDA	376-06-7	712.77	668.80	9.94
N-ethyl perfluorooctanesulfo-namidoethanol	NEtFOSE	1961-99-2	630.00	59.00	10.04

Recovery rates

Figure 3:



Recovery rate from water sample (concentration, β = 5 ng/L)

Conclusion

A reliable and successful methodology for the determination of 40 PFAS according to EPA Draft 1633 from drinking water was shown. By using the SPE column, CHROMABOND® WAX, it was possible to achieve high recovery rates for with good reproducibility especially for short chain PFAS. CHROMABOND® WAX was optimized for PFAS analysis and provides various strong ionic interaction types like ionic, hydrophobic, hydrogen bonds and dipole-dipole interactions for the enrichment of a broad spectrum of PFAS. The sorbent is specially recommended for PFAS analysis because of its very low blind value levels. Most of the PFAS show recovery rates between 80 % to 110 %.

The neutralization of the eluate leads to very high recovery rates for neutral substances such as FOSA, *N*-MeFOSE and *N*-EtFOSE. Without the enrichment of sample concentration by eluent exchange, the analysis requires LC-MSMS systems with very good performance and high sensitivity. The membrane filtration was done without PFAS losses using a syringe filter with nylone membrane (25 mm filter, 0.2 µm). The identification and the quantification of PFAS in aqueous sample were finally carried out by ESI mass spectrometry on a NUCLEODUR® PFAS column.

References

[1] Draft Method 1633, Analysis of Per- and Polyfluoroalkyl Substances (PFAS) in Aqueous, Solid, Biosolids, and Tissue Samples by LC-MS/MS; August 2021.

[2] National Pollutant Discharge Elimination System (NPDES), 832-F-12 – 033.NTP

[3] MONOGRAPH ON IMMUNOTOXICITY ASSOCIATED WITH EXPOSURE TO PERFLUOROOCTANOIC ACID (PFOA) OR PERFLUOROOCTANE SULFONATE (PFOS), September, 2016, Office of Health Assessment and Translation Division of the National Toxicology Program National Institute of Environmental Health Sciences National Institutes of Health.