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AWWA WATER SCIENCE

An interlaboratory study on EPA methods 537.1 and 533 for per- and polyfluoroalkyl substance analyses

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Abstract

The US Environmental Protection Agency (EPA) Methods 537.1 and 533 were developed for analysis of per- and polyfluoroalkyl substances (PFAS) in drinking water. They have been also widely used for source water assessments. However, there are few studies reportedly supporting such applications. The main purpose of this interlaboratory study was to evaluate the performance of these two methods for use with both potable and nonpotable waters. The obtained matrix spike recoveries indicate that both methods are generally applicable for analysis of PFAS in pristine nonpotable water matrices, however, with a notable challenge for effectively extracting long-chain PFAS from some nonpotable water matrices. Another challenge associated with EPA Method 533 is the impacts likely caused by co-extracted common inorganic anions on those PFAS that do not have their own isotopically labeled analogues available. The experimental results indicate that these challenges can be successfully resolved or reduced by enhancing postextraction bottle rinsing and elution procedures.

KEYWORDS
EPA 533, EPA 537.1, PFAS interlaboratory study, potable and nonpotable water

1 | INTRODUCTION

Human exposure to per- and polyfluoroalkyl substances (PFAS) is a worldwide public health issue (Lindstrom, Strynar, & Libelo, 2011; Paul et al., 2009). Reports have estimated that over 3,000 PFAS exist on the global market and have been used in a wide variety of industrial and consumer applications (Wang et al., 2017). Concerns over PFAS contamination have increased substantially through the years due to increased occurrence assessments and studies reporting PFAS detected in global water sources and finished drinking water (DW) (Ahrens, 2011; Crone et al., 2019; Gebbink et al., 2017; Mak et al., 2009). Recent discoveries of legacy and emerging PFAS identified in water sources related to discharges of PFAS manufacturing facilities have greatly increased concerns about the use of fluoroalkyl ether substances as alternatives for long-chain legacy PFAS (Newton et al., 2017; Strynar et al., 2015; Sun et al., 2016; Wang et al., 2013).

In the United States, a national DW occurrence assessment of six PFAS was conducted under the Third Unregulated Contaminant Monitoring Rule (UCMR 3) (USEPA, 2017a), 29 PFAS have been included in the recently proposed fifth round of UCMR (USEPA, 2019a), and the US Environmental Protection Agency (EPA) established the DW lifetime health advisory levels at 70 ng/L for perfluorooctanoic acid (PFOA) and perfluorooctanesulfonic acid (PFOS) (USEPA, 2016). At the state levels, the pace of action on PFAS issues varies greatly from state to state. Several states have established,

recommended, or proposed DW compliance standards (e.g., maximum contaminant levels, notification levels, human health–based values, trigger levels, or action levels) for individual PFAS or as sums of selected PFAS. A few other states have also developed statewide action plans. Source water evaluation for PFAS is also a current topic (AWWA, 2020; Nakayama et al., 2010; Schultz et al., 2004). DW supplies have become increasingly vulnerable to PFAS contamination from industrial discharges (Hu et al., 2016; Nakayama et al., 2007), firefighting activities (Backe et al., 2013; Place & Field, 2012), wastewater (WW) discharges (Schultz et al., 2006; Zareitalabad et al., 2013), landfill leachates (Huset et al., 2011), biosolids used as fertilizer (Lindstrom, Strynar, Delinsky, et al., 2011; Washington et al., 2010), or air emissions (Stoiber et al., 2020).

Liquid chromatography/tandem mass spectrometry (LC/MS/MS) is the primary analytical technique used for PFAS analyses (Amin et al., 2020; de Voogt & Saez, 2006; Munoz et al., 2019). Direct injection LC/MS/MS is applicable for a wide range of PFAS at part-per trillion concentrations (USEPA, 2019b; Zintek, 2017). Lower limits of quantitation (LOQ) can be achieved by combining LC/MS/MS with solid phase extraction (SPE). However, reversed-phase sorbents are generally less effective in retaining more acidic short-chain PFAS (Shoemaker et al., 2009; Shoemaker & Tettenhorst, 2018; Shoemaker & Tettenhorst, 2020). On the contrary, weak anion exchange (WAX) sorbents are relatively less effective in retaining less acidic long-chain and nonionic PFAS (Rosenblum & Wendelken, 2019).

EPA Methods 537.1 and 533 are two well-established DW methods using LC/MS/MS combined with reversed-phase and WAX SPE, respectively. These methods are also often used to analyze PFAS in nonpotable waters, including surface water (SW), groundwater (GW), and treated WW. However, there are few studies supporting such applications. Therefore, there is a need for investigating the performance and challenges of these two DW methods for use with various nonpotable water matrices. In this work, four Eurofins laboratories conducted a round-robin study on EPA Methods 537.1 and 533 for PFAS in both potable and nonpotable water matrices. The performance of these two methods was evaluated by measuring the native concentrations of PFAS in the selected water matrices and the matrix spike (MS) recoveries. The results indicate that both methods are generally applicable for pristine SW, GW, and treated WW. However, specific nonpotable water matrices may pose a challenge for meeting the quality control acceptance criteria as described in the methods. This may be successfully resolved or reduced simply by enhancing the postextraction bottle rinsing and elution procedures.

Article Impact Statement

This study indicates that Environmental Protection Agency Methods 537.1 and 533 are applicable for pristine nonpotable water per- and polyfluoroalkyl substance analysis or source water assessment.

2 | MATERIALS AND METHODS

2.1 | Chemicals and standards

The PFASs standards used for the water matrix fortification were purchased from Wellington Laboratories (Guelph, ON, Canada) and Absolute Standards (Hamden, CT). All isotopically labeled PFAS analogues were purchased from Wellington Laboratories. The target PFAS, their associated abbreviations or acronyms, and minimum reporting levels (MRLs) from the four participating laboratories are included in Table 1. Purge-and-trap-grade methanol used for the PFAS standard dilution was purchased from Fisher Scientific (St. Louis, MO).

2.2 | Water matrices and sample collection

The water matrices used in this study included laboratory reagent water (RW), DW, GW, SW, and treated WW. The RW (18.2 MΩ-cm resistance) was collected from a Millipore Milli-Q Academic system (Bedford, MA) of Lab A. 250 mL RW and DW samples were directly collected into 8-oz high-density polyethylene (HDPE) bottles containing the appropriate amounts of preservatives, which were obtained from Environmental Sampling Supply (San Leandro, CA). DW1 was a local city tap water. DW2 was prepared from DW1 by adjusting the hardness with an appropriate amount of magnesium chloride. GW, SW, and WW samples were initially collected in 10-L low-density polyethylene cubitainers or 1-gal HDPE jugs, which contained no detectable PFAS of interest. GW1 was collected from a local GW well. GW2 was prepared from GW1 by adjusting the alkalinity with an appropriate amount of sodium bicarbonate. SW1 was collected from St. Joseph River (South Bend, IN). SW2 was collected from Pleasant Lake (Edwardsburg, MI). Both WW1 and WW2 were collected from two local WW treatment plant effluents, which were chlorinated and then dechlorinated before discharge. No prefiltration was performed for

TABLE 1 A summary of analytes and minimum reporting levels

Analyte	Abbreviation or Acronym	EPA Method 537.1/533 MRL (ng/L)			
		Lab A	Lab B	Lab C	Lab D
Perfluorobutanoic acid	PFBA	NA/2.0	NA/2.0	NA/2.0	NA/5.0
Perfluoropentanoic acid	PFPeA	NA/2.0	NA/2.0	NA/2.0	NA/2.0
Perfluorohexanoic acid	PFHxA	2.0/2.0	2.0/2.0	2.0/2.0	2.0/2.0
Perfluoroheptanoic acid	PFHpA	2.0/2.0	2.0/2.0	2.0/4.0	2.0/2.0
Perfluorooctanoic acid	PFOA	2.0/2.0	2.0/2.0	2.0/2.0	2.0/2.0
Perfluorononanoic acid	PFNA	2.0/2.0	2.0/2.0	2.0/2.0	2.0/2.0
Perfluorodecanoic acid	PFDA	2.0/2.0	2.0/2.0	2.0/2.0	2.0/2.0
Perfluoroundecanoic acid	PFUnA	2.0/2.0	2.0/2.0	2.0/2.0	2.0/2.0
Perfluorododecanoic acid	PFDoA	2.0/2.0	2.0/2.0	2.0/2.0	2.0/2.0
Perfluorotridecanoic acid	PFTrDA	2.0/NA	2.0/NA	2.0/NA	2.0/NA
Perfluorotetradecanoic acid	PFTeDA	2.0/NA	2.0/NA	2.0/NA	2.0/NA
Perfluorobutanesulfonic acid	PFBS	2.0/2.0	2.0/2.0	2.0/2.0	2.0/2.0
Perfluoropentanesulfonic acid	PFPeS	2.0/2.0	2.0/2.0	2.0/2.0	2.0/2.0
Perfluorohexanesulfonic acid	PFHxS	2.0/2.0	2.0/2.0	2.0/2.0	2.0/2.0
Perfluoroheptanesulfonic acid	PFHpS	2.0/2.0	2.0/2.0	2.0/2.0	2.0/2.0
Perfluorooctanesulfonic acid	PFOS	2.0/2.0	2.0/2.0	2.0/2.0	2.0/2.0
N-ethyl perfluorooctanesulfonamidoacetic acid	NEtFOSAA	2.0/NA	2.0/NA	2.0/NA	2.0/NA
N-methyl perfluorooctanesulfonamidoacetic acid	NMeFOSAA	2.0/NA	2.0/NA	2.0/NA	2.0/NA
1H,1H,2H,2H- Perfluorohexanesulfonic acid	4:2 FTS	NA/2.0	NA/2.0	NA/2.0	NA/2.0
1H,1H,2H,2H- Perfluorooctanesulfonic acid	6:2 FTS	NA/2.0	NA/2.0	NA/2.0	NA/10
1H,1H,2H,2H- Perfluorodecanesulfonic acid	8:2 FTS	NA/2.0	NA/2.0	NA/2.0	NA/2.0
Perfluoro-3-methoxypropanoic acid	PFMPA	NA/2.0	NA/2.0	NA/2.0	NA/2.0
Perfluoro-4-methoxybutanoic acid	PFMBA	NA/2.0	NA/2.0	NA/2.0	NA/2.0
Perfluoro(2-ethoxyethanesulfonic acid	PFEESA	NA/2.0	NA/2.0	NA/2.0	NA/2.0
Nonafluoro-3,6-dioxaheptanoic acid	NFDHA	NA/2.0	NA/2.0	NA/2.0	NA/2.0
Hexafluoropropylene oxide dimer acid	HFPO-DA	2.0/2.0	2.0/2.0	2.0/2.0	2.0/2.0
4,8-Dioxa-3H-perfluorononanoic acid	ADONA	2.0/2.0	2.0/2.0	2.0/2.0	2.0/2.0
9-chlorohexadecafluoro-3-oxanonane-1-sulfonic acid	9Cl-PF3ONS	2.0/2.0	2.0/2.0	2.0/2.0	2.0/2.0
11-chloroeicosafluoro-3-oxanonane-1-sulfonic acid	11Cl-PF3OUdS	2.0/2.0	2.0/2.0	2.0/2.0	2.0/2.0

Abbreviation: NA, not applicable.

these nonpotable water samples. 250 mL GW, SW, and WW samples were then poured into the 8-oz HDPE bottles. The unfortified bottles were used to measure the native concentrations of PFAS. The fortified bottles were used to measure the recoveries of PFAS. DW1, DW2, GW1, GW2, SW1, and SW2 were fortified with all PFAS at 10 ng/L. WW1 and WW2 were fortified with all PFAS at 50 ng/L. RW1 fortified with all PFAS at 2 ng/L was used to evaluate the performance of quality control samples close to the LOQ. RW2 fortified with all PFAS at 50 ng/L was used as a reference to compare with the other water matrices. All water samples were shipped,

received, and stored as described in EPA Methods 537.1 and 533.

As shown in Table 2, the pH, free chlorine, total organic carbon (TOC), total alkalinity, hardness, nitrate, and heterotrophic plate count (HPC) of these samples were measured by using EPA Method 150.1, SM 4500-Cl G, SM 5310 C, SM 2320 B, SM 2340 B, EPA Method 353.2, and SM 9215 E (SimPlate), respectively. The water hardness was calculated by SM 2340 B based on the concentrations of Ca and Mg cations measured by using EPA Method 200.7. Both chloride and sulfate were measured by using EPA Method 300.0.

TABLE 2 A summary of water quality parameters of studied water matrices

Matrix	DW1	DW2	GW1	GW2	SW1	SW2	WW1	WW2
pH	6.8	7.2	7.3	7.4	7.3	6.7	7.3	7.2
Free chlorine (mg/L)	0.86	0.86	<0.05	<0.05	<0.05	<0.05	<0.05	<0.05
TOC (mg/L)	0.516	0.516	0.858	0.427	3.13	5.84	2.44	3.71
Total hardness as $CaCO_3$ (mg/L)	401	563	257	256	278	122	412	324
Total alkalinity as $CaCO_3$ (mg/L)	276	275	208	320	214	109	276	213
Chloride (mg/L)	147	289	37.8	37.4	33.2	12.9	239	182
Sulfate (mg/L)	59.7	59.6	27.6	26.7	39.2	4.6	93.8	53.5
Nitrate as nitrogen (mg/L)	<1.0	<1.0	1.69	2.12	1.84	<1.0	11.0	13.2
HPC (MPN/mL)	NA	NA	311	372	650	1000	440	623

Abbreviations: DW, drinking water; GW, groundwater; HPC, heterotrophic plate count; NA, not available; SW, surface water; TOC, total organic carbon; WW, wastewater.

2.3 | Sample extraction and analysis

EPA Methods 537.1 and 533 were performed for all 29 PFAS without modification. All participating laboratories fully complied with the requirements for calibration standards and curve fits, quality control types and acceptance criteria, SPE procedures, and extraction/analysis batches as specified in the two reference methods. For EPA Method 533, all laboratories used 33 μm Strata X-AW mixed-mode polymeric WAX cartridges (500 mg/6 ml) obtained from Phenomenex (Torrance, CA). For EPA Method 537.1, Labs A, B, and C used 100 μm Strata SDBL polymeric styrenedivinylbenzene cartridges (500 mg/6 ml) obtained from Phenomenex. Lab D used 45 μm Mega Bond Elut Plexa cartridges (500 mg/6 ml) obtained from Agilent Technologies Inc. (Lake Forest, CA). Upon receipt, 250 mL properly preserved samples were fortified with surrogate standards (SS) for EPA Method 537.1 or isotope dilution analogues (IDAs) for EPA Method 533 and then extracted using the SPE cartridges without performing prefiltration. Prior to the LC/MS/MS analysis, the extracts were evaporated to dryness, reconstituted with proper solvents to 1.0 ml, and then fortified with proper internal standards (IS) for EPA Method 537.1 or isotope performance standards (IPS) for EPA Method 533. The PFAS were separated on the C18 columns using a gradient mobile phase of 20 mM ammonium acetate and methanol and then detected by negative electrospray ionization LC/MS/MS in the multiple-reaction monitoring mode.

The PFAS were quantitated using a minimum of five calibration standards for a nonprocedural linear curve or six calibration standards for a nonprocedural quadratic curve. EPA Method 537.1 calibration curves using the IS were in concentration ranges of 2.0–250 ng/L, 2.0–80 ng/L, 1.0–400 ng/L, and 2.0–80 ng/L for Labs A, B, C, and D, respectively. EPA Method 533 calibration curves using the IDAs were in concentration ranges of 2.0–500 ng/L, 2.0–80 ng/L, 1.0–400 ng/L, and 2.0–100 ng/L for Labs A, B, C, and D, respectively. The extracted SS and IDAs were quantitated using multiple calibration points at the same concentration level against the IS and IPS, respectively. The IS were evaluated by comparing with their average peak areas in the initial calibration and the peak areas from the most recent continuing calibration check. The IPS were evaluated by comparing with their average peak areas in the initial calibration. The percent recoveries of the PFAS, SS, and IDAs were calculated by comparing with the true fortified values. The initial method detection limits (MDLs) were determined using the latest 40 CFR Part 136 Appendix B protocol (USEPA, 2017b). The ongoing MDLs were determined following the 2016 TNI Standard (NELAC Institute, 2016). As shown in Table S1 in Appendix S1, the MDLs of less than 0.67 ng/L (1/3 the MRL of 2.0 ng/L) were obtained for all PFAS for EPA Methods 537.1 and 533 with the following exceptions: Lab C had EPA Method 533 MDLs of 0.91, 0.68, 1.0, and 0.74 ng/L for PFBA, PFPeA, PFHpA, and PFOA, respectively. Lab D had nominal EPA Method 533 MDLs of 2.0 ng/L for both PFBA and 6:2 FTS.

3 | RESULTS AND DISCUSSION

3.1 | Native concentrations of PFAS

As shown in Table S2 in Appendix S1, Labs A and B reported that all PFAS in the RW samples were detected at concentrations less than one-third the MRL of 2.0 ng/L. Labs C and D reported all PFAS as not detected, based on their data processing settings. Table 3 indicates the mean native concentrations with standard deviations

TABLE 3 Native concentrations of PFAS resulting from EPA Methods 537.1 and 533

Analyte	Mean concentration ± SD (ng/L)							
	DW1	DW2	GW1	GW2	SW1	SW2	WW1	WW2
EPA 537.1 results								
PFBS	2.48 ± 0.17	2.53 ± 0.19	5.53 ± 0.25	5.53 ± 0.35	1.4 ± 0.8[a]	0.9 ± 0.5[a]	3.53 ± 0.71	3.99 ± 1.54
PFHxA	1.6 ± 0.9[a]	2.10 ± 0.24	0.7 ± 0.4[a]	0.8 ± 0.5[a]	1.0 ± 0.6[a]	1.1 ± 0.7[a]	14.7 ± 0.8	15.5 ± 1.1
PFHpA	1.5 ± 0.9[a]	1.5 ± 0.9[a]	ND	ND	ND	1.1 ± 0.6[a]	1.98 ± 0.17	1.5 ± 0.3[a]
PFHxS	ND	1.0 ± 0.6[a]	0.8 ± 0.5[a]	0.7 ± 0.5[a]	0.9 ± 0.6[a]	ND	3.19 ± 0.32	5.91 ± 0.66
PFOA	9.70 ± 0.77	10.1 ± 0.6	2.84 ± 0.18	2.85 ± 0.19	1.0 ± 0.6[a]	2.74 ± 0.16	4.55 ± 0.49	3.94 ± 0.33
PFOS	ND	ND	0.7 ± 0.6[a]	0.7 ± 0.6[a]	1.2 ± 0.9[a]	1.1 ± 0.8[a]	3.09 ± 0.86	9.35 ± 1.07
PFNA	ND	ND	ND	ND	ND	ND	0.67 ± 0.30	ND
EPA 533 results								
PFBA	3.2 ± 0.3[b]	3.2 ± 0.3[b]	4.5 ± 0.3[b]	4.6 ± 0.5[b]	2.0 ± 0.3[b]	2.8 ± 0.5[b]	6.89 ± 1.80	7.80 ± 2.92
PFPeA	1.7 ± 0.1[a]	1.7 ± 0.2[a]	0.9 ± 0.1[a]	0.8 ± 0.1[a]	1.4 ± 0.3[a]	1.4 ± 0.3[a]	20.3 ± 1.7	15.4 ± 1.2
PFBS	2.44 ± 0.11	2.41 ± 0.18	5.36 ± 0.35	5.23 ± 0.45	1.3 ± 0.7[a]	0.9 ± 0.5[a]	3.48 ± 0.34	4.04 ± 0.60
PFHxA	2.00 ± 0.12	1.99 ± 0.24	1.0 ± 0.1[a]	0.9 ± 0.1[a]	0.9 ± 0.6[a]	1.0 ± 0.6[a]	14.9 ± 1.7	15.9 ± 1.7
PFHpA	1.8 ± 0.2[a]	1.9 ± 0.2[a]	1.0 ± 0.1[a]	0.8 ± 0.1[a]	ND	1.1 ± 0.2[a]	1.8 ± 0.2[a]	1.5 ± 0.3[a]
PFHxS	1.2 ± 0.1[a]	1.2 ± 0.1[a]	0.8 ± 0.1[a]	0.9 ± 0.1[a]	0.7 ± 0.4[a]	ND	2.88 ± 0.36	5.51 ± 0.64
PFOA	9.63 ± 0.79	9.43 ± 0.71	3.00 ± 0.33	2.79 ± 0.17	0.8 ± 0.5[a]	1.9 ± 1.1[a]	4.03 ± 0.52	3.80 ± 0.60
PFNA	ND	ND	ND	ND	ND	0.7 ± 0.5[a]	ND	ND
PFOS	ND	ND	ND	ND	1.1 ± 0.7[a]	1.0 ± 0.6[a]	2.49 ± 0.29	9.60 ± 0.57

Abbreviations: DW, drinking water; GW, groundwater; ND, not detected; PFBA, perfluorobutanoic acid; PFBS, perfluorobutanesulfonic acid; PFHpA, perfluoroheptanoic acid; PFHpA, perfluoroheptanoic acid; PFHxA, perfluorohexanoic acid; PFHxS, perfluorohexanesulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctanesulfonic acid; PFPeA, perfluoropentanoic acid; SW, surface water; WW, wastewater.
[a]Estimated results of less than the minimum reporting level (MRL) of 2.0 ng/L.
[b]Estimated results of less than Lab D's MRL of 5.0 ng/L.

(SDs) of PFAS measured by the four participating laboratories, which were based on eight replicate samples (i.e., two replicate unfortified samples were run by each laboratory), with an exception of GW1. EPA Method 533 results for GW1 were based on six replicate unfortified samples run by three participating laboratories. The results measured between the MRLs and one-third of the MRLs were included as estimates. Table 3 does not include the analytes, which were not detected by any of the four participating laboratories. As shown in Table 3, the native concentrations resulting from both EPA Methods 537.1 and 533 were generally consistent for the potable and nonpotable water matrices. All four participating laboratories achieved relatively low SDs for all PFAS measured at or greater than the MRLs. In addition, the obtained relative percent differences (RPDs) between the two methods were less than 20% for all PFAS at or greater than the MRLs with a couple of exceptions. At the native concentrations close to the MRL of 2.0 ng/L, the RPDs were 24% for PFHxA in DW1 and 21% for PFOS in WW1, respectively.

3.2 | Accuracy and precision of PFAS analyses

Four MS replicates were extracted and analyzed by each participating laboratory to demonstrate the accuracy and precision. The obtained percent recoveries and trending from all four laboratories were very similar, which can be found in Appendix S1. The overall mean recoveries were calculated from all 16 MS replicates analyzed by the four laboratories. Figures 1 and 2 represent the overall mean MS recoveries of the analytes and SS for EPA Method 537.1, respectively. Figures 3 and 4 represent the overall mean MS recoveries of the analytes and IDAs for EPA Method 533, respectively.

As shown in Figure 1, the obtained mean recoveries of all EPA Method 537.1 analytes were within 80%–120% for the RW, DW, and GW matrices. The mean recoveries for PFAS eluting before NMeFOSAA were within 84%–116% for the SW and treated WW matrices. It is notable that the mean recoveries gradually decreased with the increase in carbon chain lengths or retention time for
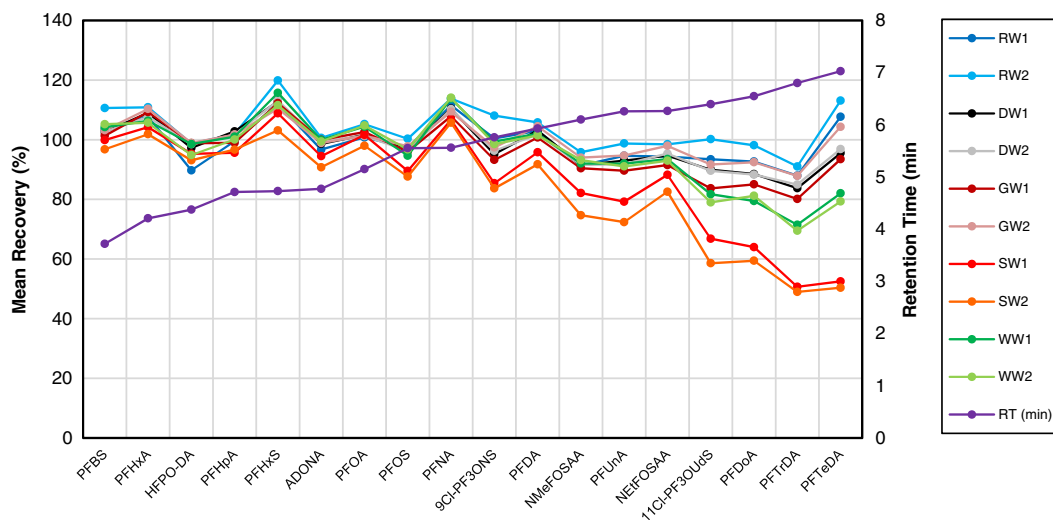
FIGURE 1 Mean recoveries of EPA Method 537.1 analytes resulting from the four participating laboratories, based on 16 matrix spike replicates. The fortification concentrations were 2 ng/L for RW1; 10 ng/L for DW1, DW2, GW1, GW2, SW1, and SW2; and 50 ng/L for RW2, WW1, and WW2, respectively. DW, drinking water; GW, groundwater; RW, reagent water; SW, surface water; WW, wastewater
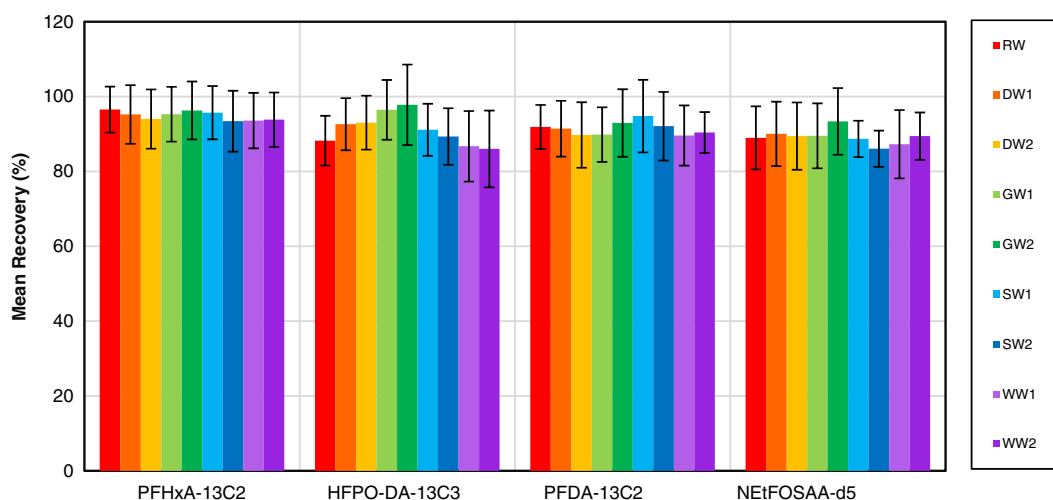

FIGURE 2 Mean recoveries with positive/negative percent relative standard deviations of EPA Method 537.1 surrogate standards resulting from the four participating laboratories, based on 40 reagent water (RW) replicates (32 fortified blanks and 8 unfortified RW samples) and 24 replicates for the other water matrices (16 fortified matrix spikes and 8 unfortified native water samples). The fortification concentrations were 40 ng/L for HFPO-DA-$^{13}C_3$, PFHxA-$^{13}C_2$, and PFDA-$^{13}C_2$; and 160 ng/L for NEtFOSAA-d$_5$, respectively. DW, drinking water; GW, groundwater; SW, surface water; WW, wastewater

long-chain PFAS. From PFDA to PFTeDA, the mean recoveries varied from 102% to 82% for WW1, from 101% to 79% for WW2, from 96% to 53% for SW1, and from 92% to 50% for SW2, respectively.

Figure 1 indicates that common inorganic water quality parameters (e.g., total hardness, total alkalinity, and common anion concentrations as shown in Table 1) might not significantly interfere with recoveries of PFAS, including late-eluting 11Cl-PF3OUdS, PFDoA, PFTrDA, and PFTeDA. The decreased recoveries of these PFAS for the SW and treated WW matrices might be due to their

surface adsorption enhanced by the presence of relatively high HPC and/or TOC concentrations. A hypothesis is that these long-chain PFAS could conjugate with large organic molecules, biological matter, and/or porous debris likely present in the SW matrices. The conjugation might create a co-adsorption effect, which could make the extraction, bottle rinsing, and elution more challenging.

Figure 2 indicates that the mean MS recoveries of all four SS were within 86%–98% with percent relative standard deviations (RSDs) of 4.8%–10.8% for all the studied matrices. However, the results also indicate that the
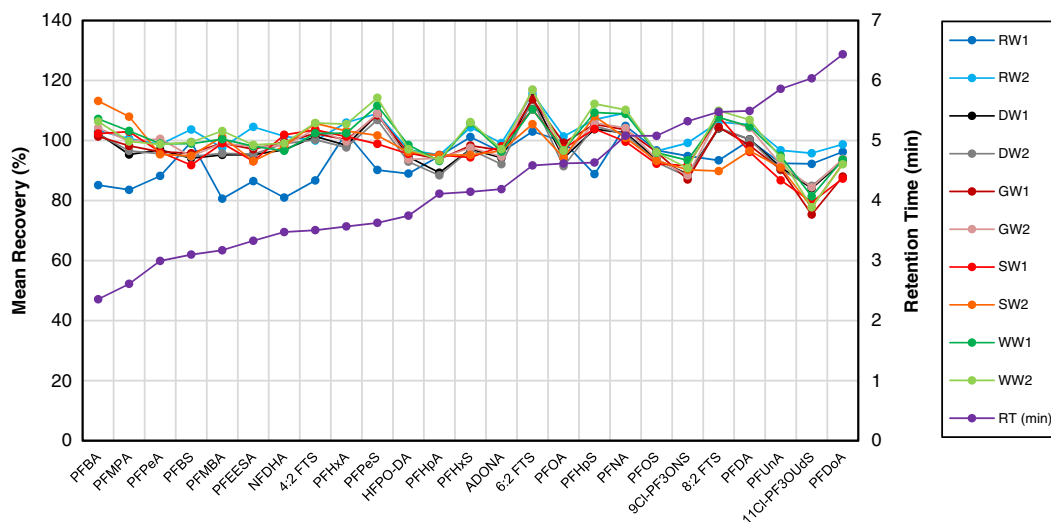
FIGURE 3 Mean recoveries of EPA Method 533 analytes resulting from the four participating laboratories, based on 16 matrix spike replicates. The fortification concentrations were 2 ng/L for RW1; 10 ng/L for DW1, DW2, GW1, GW2, SW1, and SW2; and 50 ng/L for RW2, WW1, and WW2, respectively. DW, drinking water; GW, groundwater; RW, reagent water; SW, surface water; WW, wastewater
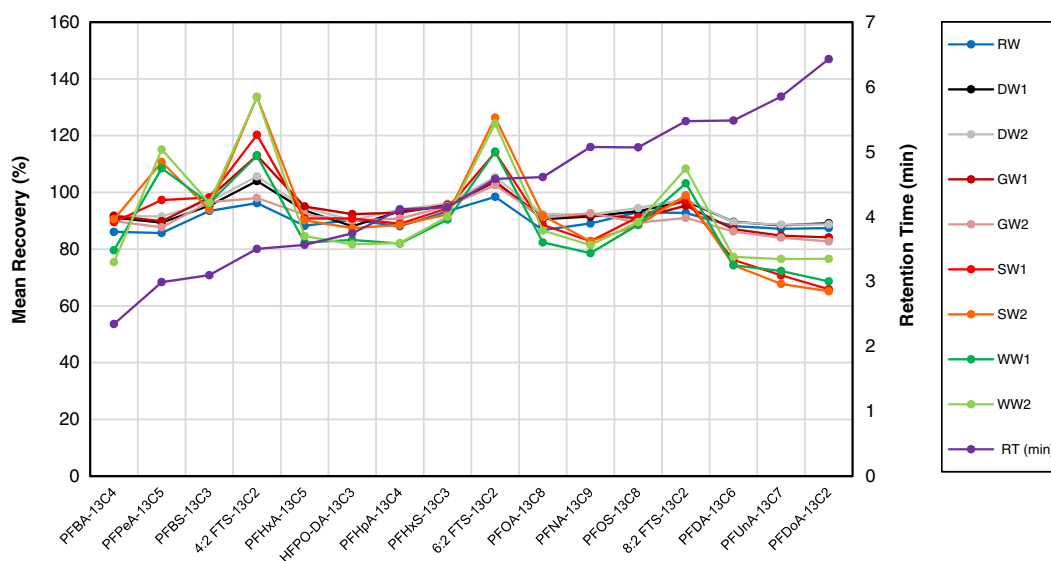

FIGURE 4 Mean recoveries of EPA Method 533 isotope dilution analogues resulting from the four participating laboratories, based on 40 reagent water (RW) replicates (32 fortified blanks and 8 unfortified RW samples) and 24 replicates for the other water matrices (16 fortified matrix spikes and 8 unfortified native water samples). The fortification concentrations were 160 ng/L for 4:2 FTS-$^{13}C_2$, 6:2 FTS-$^{13}C_2$, and 8:2 FTS-$^{13}C_2$; and 40 ng/L for the other isotope dilution analogues, respectively. DW, drinking water; GW, groundwater; SW, surface water; WW, wastewater

latest-eluting surrogate NEtFOSAA-d$_5$, passing the acceptance criteria of 70%–130% recoveries, does not necessarily predict good extraction efficiency for long-chain PFAS typically eluting after NEtFOSAA or NEtFOSAA-d$_5$ in challenging nonpotable water matrices like SW1 and SW2. On the other hand, the SS falling below the acceptance criteria will clearly indicate the likelihood of significantly low biased results for the long-chain PFAS.

Figure 3 indicates that the mean MS recoveries of EPA Method 533 analytes did not obviously decrease with the increase in carbon chain lengths for long-chain PFAS. All the analytes were measured with mean recoveries at 75%–117% for all the studied water matrices. It is worth mentioning that EPA Method 533 did not include NMeFOSAA, NEtFOSAA, PFTrDA, and PFTeDA, possibly due to the relatively low extraction recoveries, poor precision, and/or the solubility concern of PFTrDA and
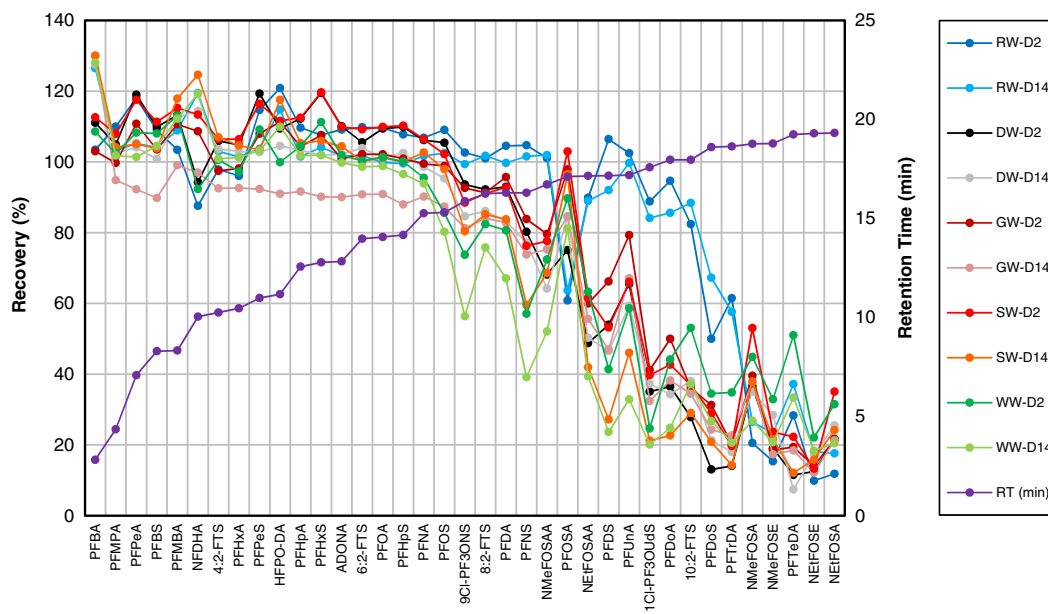
FIGURE 5 Recoveries of per- and polyfluoroalkyl substances fortified into 8-oz high-density polyethylene sample bottles at 400 ng/L and stored at refrigeration temperature (1–6 °C). DW, drinking water; GW, groundwater; RW, reagent water; SW, surface water; WW, wastewater

PFTeDA in the 80:20 volumetric ratio of methanol/water solvent. As shown in Figure 4, all IDAs were measured with the mean recoveries of 65%–134% for all studied water matrices, which were well within the acceptable limits of 50%–200% as described in the method. Similar to Figure 1, the obtained mean recoveries of all EPA Method 533 IDAs were within 83%–113% for the studied RW, DW, and GW matrices. The mean recoveries of 79%–134% were also obtained for the IDAs eluting before PFDA-$^{13}C_6$. It is also notable that the mean recoveries slightly decreased with the increase in carbon chain lengths for long-chain IDAs for the SW and treated WW matrices. From 8:2 FTS-$^{13}C_2$ to PFDoA-$^{13}C_2$, the mean recoveries varied from 103% to 69% for WW1, from 108% to 77% for WW2, from 97% to 66% for SW1, and from 99% to 65% for SW2, respectively. Similarly, the decreased IDA recoveries might be due to their surface adsorption enhanced by the presence of relatively high HPC and/or TOC concentrations in these SW and treated WW matrices. Although the recoveries of some IDAs were slightly low-biased for these challenging matrices, the isotope dilution technique effectively compensated for the matrix interferences, as shown in Figure 3.

It is worth mentioning that it is critical to choose appropriate reference IDAs for those analytes that do not have their own isotopically labeled analogues available. The mean recoveries of early-eluting PFMPA shown in Figure 3 for WW1 and WW2 were quantitated against the IDAs PFPeA-$^{13}C_5$ (Labs A and B) and PFBA-$^{13}C_4$ (Labs C and D),

respectively. For Lab A, PFMPA was initially measured with mean recoveries of 143% with RSD of 2.4% for WW1 and 188% with RSD of 1.6% for WW2, corresponding to the mean recoveries of IDA PFBA-$^{13}C_4$ at 65% with RSD of 2.6% and 68% with RSD of 1.9%, respectively. However, PFMPA was measured with mean recoveries of 89% with RSD of 1.6% for WW1 and 90% with RSD of 1.2% for WW2 when the corresponding IDA was switched to PFPeA-$^{13}C_5$. For Lab B, PFMPA was initially measured with mean recoveries of 152% with RSD of 1.6% for WW1 and 161% with RSD of 3.2% for WW2, corresponding to the mean recoveries of IDA PFBA-$^{13}C_4$ at 59% with RSD of 1.2% and 51% with RSD of 7.1%, respectively. However, PFMPA was measured with mean recoveries of 95% with RSD of 0.6% for WW1 and 85% with RSD of 2.8% for WW2 when the corresponding IDA was switched to PFPeA-$^{13}C_5$. As shown in Tables S7–S10 in Appendix S1, compared with the RW, DW, GW, and SW matrices, lower recoveries of IDA PFBA-$^{13}C_4$ were obtained from the treated WW matrices for all four laboratories. The co-extracted polar organic compounds and/or common anions from the WW matrices could co-elute with PFBA-$^{13}C_4$ and cause electrospray ionization suppression.

3.3 | Surface adsorption of PFAS

In a separate surface adsorption study, the same water matrices were collected in 8-oz HDPE bottles, fortified

with PFAS at 400 ng/L, and stored at refrigeration temperature (typically, 1–6 °C). An aliquot of the samples was then transferred into autosampler vials after storage of 2 and 14 days, mixed with an aliquot of ammonium acetate buffer, fortified with isotopically labeled PFAS analogues used as the IS, diluted with methanol in a 60:40 volumetric ratio of water to methanol, and then mixed well prior to the LC/MS/MS analysis. In order to match the adsorption losses in the autosampler vials and analytical system, the calibration standards were also prepared in 40% methanol in RW. This study also included several commonly analyzed PFAS additional to EPA Method 537.1 and EPA Method 533 analyte lists. The obtained results are similar to those resulting from the surface adsorption study on 8-oz high-density polypropylene bottles (Whitaker & Li, 2018). As shown in Figure 5, the recoveries of PFAS were measured as a reciprocal of surface adsorption losses. First of all, the trending variations of mean recoveries were not notably different from 2-day storage to 14-day storage. Secondly, in general, the recoveries of PFAS gradually decreased with the increase in carbon chain lengths or retention time for long-chain PFAS, which means that the surface adsorption of PFAS on the inner walls of the HDPE bottles generally increased with the increase in carbon chain lengths after PFNA. Thirdly, compared with the RW results, the recoveries of PFAS substantially decreased in the DW, GW, SW, and WW, particularly for PFAS between 9Cl-PF3ONS and PFTrDA except neutral PFOSA. This observation indicates that these water matrices could

substantially enhance bottle surface adsorption, particularly for those acidic PFAS. However, the RW might result in slightly more bottle surface adsorption of neutral PFOSA than the DW, GW, SW, and WW matrices. Finally, for long-chain PFAS eluting after PFUnA, approximately 50% or more PFAS could be adsorbed on the inner walls of the HDPE bottles.

The obtained relatively low recoveries shown in Figures 1 and 3 of late-eluting EPA Method 537.1 analytes and EPA Method 533 IDAs are basically consistent with the PFAS recoveries shown in Figure 5. These results indicate that hydrolytic, photolytic, and/or biological degradations of environmentally persistent PFAS may be minimal. PFAS adsorption losses are a primary challenge for LC/MS/MS analyses combined with SPE because manual or automated SPE procedures normally involve more surface contact areas in addition to sample bottles. Because adsorption losses of long-chain PFAS are likely enhanced by water matrices (particularly for nonpotable water samples containing high concentration of TOC, large organic molecules, biological matter, and porous debris), enhancing postextraction bottle rinsing and elution will become critical. In order to confirm this hypothesis, the participating Lab A repeated once the normal postextraction bottle rinsing and elution steps; i.e., doubled volumes of methanol were used in the postextraction bottle rinsing and elution steps. As shown in Figure 6, the mean recoveries of PFAS were significantly improved, increasing by 6%–30% for SW1 and by 4%–30% for SW2, depending on specific PFAS.
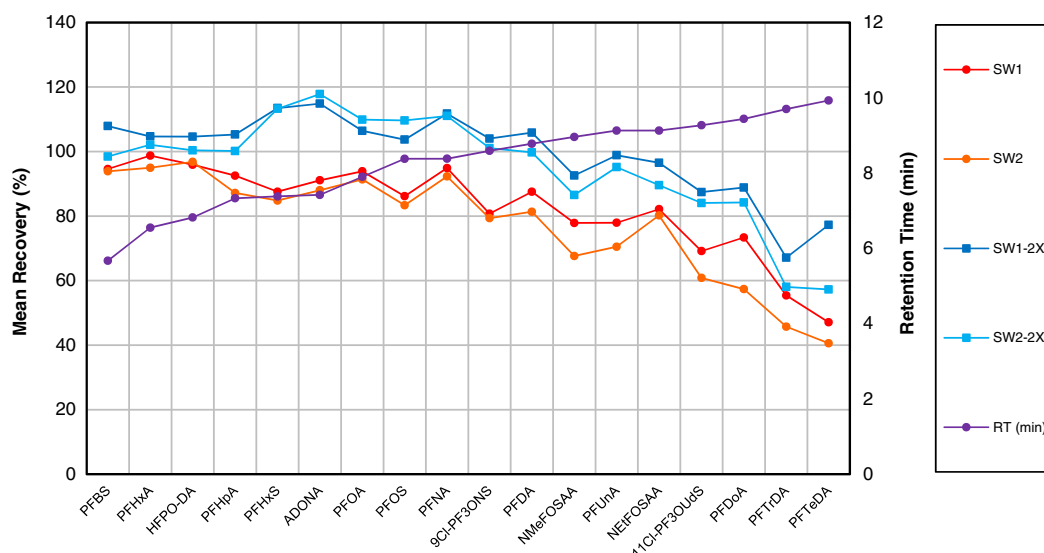

FIGURE 6 Mean recoveries of EPA Method 537.1 analytes, based on four matrix spike replicates fortified at 10 ng/L from Lab a. SW1 and SW2: Normal postextraction bottle rinsing and elution steps. SW1-2X and SW2-2X: Repeated the normal postextraction bottle rinsing and elution steps once. SW, surface water

4 | CONCLUSIONS

The experimental results have demonstrated that both EPA Methods 537.1 and 533 are accurate and precise DW methods. They may be likely applicable for pristine non-potable water analyses. Bottle surface adsorption losses were demonstrated as a primary challenge for meeting the quality control acceptance criteria as described in the reference methods and for providing accurate analytical results, particularly for nonpotable water matrices such as SW in which high concentrations of TOC, large organic molecules, biological matter, and porous debris may be present. Slightly low biased recoveries (typically, 50%–69%) of late-eluting surrogate NEtFOSAA-d$_5$ are commonly associated with EPA Method 537.1, which may occur in a small fraction of potable and nonpotable samples. The experimental results indicate that this issue can be significantly reduced by using enhanced postextraction bottle rinsing and elution procedures. Compared with EPA Method 537.1, EPA Method 533 using isotope dilution analysis can tolerate more matrix interference and subsequently provide more accurate results. In addition to bottle surface adsorption losses of late-eluting PFAS, another notable challenge for EPA Method 533 is to choose appropriate IDAs for those analytes that do not have their own isotopically labeled analogues available. Ionization suppression likely resulting from co-extracted polar organic compounds and/or common inorganic anions can significantly affect the response factors between early-eluting analytes and the reference IDAs, which may be dependent on specific SPE procedures. Similar to EPA Method 537.1, enhancing the postextraction bottle rinsing and elution steps can also be expected to a good practice for EPA Method 533, possibly reducing the extraction of common inorganic anions and surface adsorption losses of late-eluting PFAS.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

DATA AVAILABILITY STATEMENT

Data available in article supplementary material.

AUTHOR CONTRIBUTIONS

Yongtao Li: Conceptualization; resources; data curation; formal analysis; supervision; funding acquisition; validation; investigation; visualization; methodology; writing - original draft; project administration; writing-review & editing. **Joshua S. Whitaker:** Conceptualization; data curation; validation; methodology; writing-review & editing. **Robert B. Hrabak:** Data curation; supervision; investigation; methodology; writing-review & editing. **Marnellie Ramos:** Data curation; supervision; investigation; methodology; writing-review & editing. **Charles Neslund:** Conceptualization; data curation; supervision; investigation; methodology; writing-review & editing.

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