

Appendix A

EPA Memorandum – PFAS National Primary Drinking Water Regulation Monitoring and Reporting



OFFICE OF GROUND WATER AND DRINKING WATER

WASHINGTON, D.C. 20460

November 21, 2024

MEMORANDUM

SUBJECT: PFAS National Primary Drinking Water Regulation Monitoring and Reporting

FROM: Eric Burneson, Director *Eric Burneson*
Standards and Risk Management Division, Office of Ground Water and Drinking Water

Marietta Echeverria, Director
Compliance and Capacity Assistance Division, Office of Ground Water and Drinking Water

TO: EPA Regional Water Division Directors
Drinking Water Primacy Agencies

The purpose of the memorandum is to provide information on how drinking water primacy agencies can consider previously collected PFAS drinking water data (e.g., from UCMR 5 or state monitoring programs) to satisfy some or all of the initial monitoring requirements and determine the ongoing compliance monitoring frequency under the [final Per- and Polyfluoroalkyl Substance \(PFAS\) National Primary Drinking Water Regulation \(NPDWR\)](#). This memorandum also provides information and guidance on implementing other aspects of the PFAS NPDWR monitoring and reporting requirements.

The final PFAS NPDWR includes individual Maximum Contaminant Level Goals (MCLGs) and Maximum Contaminant Levels (MCLs) for five PFAS: perfluorooctanoic acid (PFOA), perfluorooctane sulfonic acid (PFOS), perfluorononanoic acid (PFNA), hexafluoropropylene oxide dimer acid (HFPO-DA, commonly known as GenX Chemicals), and perfluorohexane sulfonic acid (PFHxS). The final NPDWR also includes a Hazard Index MCLG and MCL for mixture combinations containing two or more of four PFAS (PFHxS, PFNA, HFPO-DA, and perfluorobutane sulfonic acid (PFBS)). Community and non-transient non community water systems must comply with the PFAS NPDWR by the deadlines described in 40 C.F.R. 141.900(b).

Initial Monitoring Requirements

Community water systems and non-transient non-community water systems are required to conduct both initial monitoring and ongoing compliance monitoring of regulated PFAS during normal operating conditions at all entry points to the distribution system according to the requirements in 40 C.F.R.

141.902. These systems must complete initial monitoring by April 26, 2027, and begin ongoing compliance monitoring thereafter.

To comply with initial monitoring requirements groundwater systems serving more than 10,000 customers and all surface water systems must collect four quarterly samples (2 to 4 months apart) over a 12-month period, and groundwater systems serving 10,000 or fewer customers must collect two semi-annual samples (5 to 7 months apart) over a 12-month period by April 26, 2027. Per Section 141.902(b)(1)(vi), primacy agencies can also allow water systems to use previously collected monitoring data to satisfy some or all of these initial monitoring requirements, if (1) the sampling was conducted according to specific data quality assurance criteria as part of the [UCMR 5](#), state-level, or other appropriate monitoring efforts that use EPA methods 533 or 537.1 with no modifications and (2) samples were collected starting on or after January 1, 2019. Further, under 141.902(b)(1)(viii), water systems using previously acquired data with fewer than the number of required initial monitoring samples (i.e., 2 or 4) or samples that do not meet the sample collection timing requirements (i.e., 2 to 4 or 5 to 7 months apart) may take additional samples in a different calendar year to supplement the existing data; samples must be taken such that all the required number of initial monitoring samples and sample collection timing requirements are satisfied according to the source water type and system size in the initial monitoring requirements described at the beginning of this paragraph. For example, large groundwater systems utilizing their UCMR 5 data from their two UCMR 5 sampling events would be required to collect two additional samples (according to the rule's timing requirements) to satisfy the NPDWR's initial monitoring requirements for four quarterly samples every 2 to 4 months apart. Moreover, if UCMR 5 samples were collected at a sample location in February and August, then in a different calendar year the water system would be required to collect one sample in April, May, or June and another sample in October, November, or December at that sample location.

The final rule also allows for primacy agencies to delete results of obvious sampling errors (e.g., detection of PFAS in the field reagent blank) and require a new sample to meet the rule's initial monitoring requirements. In the case of resampling due to sampling error(s), the water system should seek to resample as soon as possible during the required sample collection timeframe (i.e., 2 to 4 or 5 to 7 months apart). If the system does not conduct the required resampling in the regulatory timeframe, then the system is in violation of the rule's sampling requirements. After noncompliance has been identified, primacy agencies may consider, on a case-by-case basis, whether to exercise enforcement discretion as the system in noncompliance attempts to conduct resampling to satisfy the initial monitoring requirements. The EPA recommends primacy agencies memorialize in writing, whether informally or with formal enforcement actions, the resampling schedule to ensure the system returns to compliance in a timely manner. For more details on the NPDWR initial monitoring requirements, please refer to 40 C.F.R. 141.902(b)(vi) – (viii).

Compliance Monitoring Frequency Determination

The results of initial monitoring are required to be used by the primacy agency to determine the ongoing compliance monitoring frequency per 40 C.F.R. 141.902(b)(2)(i). To be eligible for reduced ongoing triennial compliance monitoring at an entry point, all regulated-PFAS initial monitoring sample results must be below all rule trigger levels. The trigger levels in the final PFAS NPDWR are equal to one-half of the regulated PFAS MCLs, and the Practical Quantitation Levels (PQLs) are based on the UCMR 5 minimum reporting levels (MRLs):

Contaminant	MCL (ppt or ng/L)	Trigger Level (1/2 of MCL) (ppt or ng/L)	PQL (based on UCMR 5 MRL) (ppt or ng/L)
PFOA	4.0	2.0	4.0
PFOS	4.0	2.0	4.0
HFPO-DA	10	5	5.0
PFHxS	10	5	3.0
PFNA	10	5	4.0
PFBS	N/A	N/A	3.0
Hazard Index (mixtures of PFHxS, PFNA, HFPO-DA, and PFBS)	1 (unitless)	0.5 (unitless)	N/A

Laboratory Requirements and Analytical Methods

Under the final rule, 40 C.F.R. 141.901(b)(1), all regulated PFAS analyses must be conducted using approved analytical methods (EPA Methods 533 or 537.1). Additionally, per 141.902(b)(2), for samples that are collected after June 24, 2024, all analyses must be performed by laboratories that have been “certified” by the EPA or the primacy agency. For initial monitoring purposes, the EPA has deemed its [UCMR 5 Laboratory Approval program](#), as well as state laboratory certification programs and state NELAP accreditation programs that use TNI standard, equivalent for purposes of meeting this requirement. This equivalency determination, with respect to the UCMR 5 Laboratory Approval Program, is based upon laboratories meeting strict UCMR 5 Laboratory Approval Program requirements, that are comparable to state laboratory certification and accreditation programs. Primacy agencies likewise have the discretion to accept sample results from laboratories that were part of a state-led drinking water PFAS sampling program that assessed laboratories for conformance with Method 533 or 537.1.

UCMR 5 Monitoring Data

Laboratories analyzing UCMR 5 samples only report data at or above UCMR 5 MRLs to the EPA. For small systems serving 10,000 or fewer customers, the EPA is currently working with the EPA-contracted laboratories (i.e., those analyzing UCMR 5 samples for these small systems) to reprocess the UCMR 5 small-system data and produce updated, lower-level results that could support compliance monitoring frequency determinations. The EPA will provide these results to the small water system (with a copy to the primacy agency) for the system to determine if they wish for their primacy agency to consider these results as all or part of their initial monitoring. Large systems serving more than 10,000 people arrange for their UCMR laboratory analysis and have direct relationships with the laboratories. With this in mind, the agency is strongly encouraging laboratories to accommodate large system requests for producing reprocessed (lower level) data reports (i.e., reexamine data housed in the laboratory’s information-management system) to support the large system’s submission of initial monitoring data. Such data may inform the primacy agency’s determination of NPDWR compliance monitoring frequency and eligibility for reduced triennial compliance monitoring.

PFAS Monitoring Data Reporting

Per Section 141.902(a)(7), for initial and compliance monitoring required under the rule, systems must report all results *provided by a laboratory* to the primacy agency and these results must be used specifically for the purpose of determining compliance monitoring frequency; this includes values

below the PQLs. The EPA recommends that laboratories use 1/3 of the PQL for each of the regulated PFAS as a minimum threshold when providing results to the system to reduce the possibility of background PFAS contamination impacts on measured sample results. The rule requires use of EPA Methods 533 and 537.1 both of which have QC requirements for field and laboratory reagent blank sample results to be less than 1/3 of the laboratory's MRLs (which must be at or below the regulatory PQLs) to ensure that background contamination is not mistaken for PFAS legitimately present in the field sample (see 89 FR 32606). Thus, EPA is recommending that laboratories use 1/3 of the PQL as the lower bound of its reporting to systems to ensure that the reported value accurately reflect the presence of PFAS in the water sample. The EPA is making this recommendation to allow for more consistent laboratory reporting of regulated PFAS monitoring data and assessment of the results by drinking water primacy agencies and water systems. Those laboratories that follow the agency's recommendation should treat results below 1/3 of the PQLs as "not detected" (ND) and would not report a numeric value.

When reporting values below PQLs laboratories must be able to qualitatively detect, not necessarily quantify, the regulated PFAS analytes. Furthermore, for UCMR 5 laboratories and other laboratories that cannot meet the method data quality objectives (DQOs¹) for the reprocessed UCMR 5 results or monitoring results under the rule (i.e., if laboratory quantitation that meets the DQOs is not possible for results below the PQLs), the lower-level data (i.e., data between the PQL and 1/3 of the PQL) should still be reported as a numeric values but notated (using a data "flag") as "qualitative." The EPA notes that these qualitative data are not used to demonstrate compliance with MCLs (laboratories must be able to produce quantitative data, meeting DQOs, at or above the PQLs to demonstrate compliance with the MCLs) but, as noted in the preamble to the final rule, while these lower level data may not have the same precision and accuracy as higher-level measurements (i.e., PQLs), they can be determined and used to ensure that water systems are monitoring at an appropriate frequency based on contamination risk.

Additional resources are listed below:

- Final PFAS National Primary Drinking Water Regulation Rulemaking Federal Register Notice: <https://www.federalregister.gov/documents/2024/04/26/2024-07773/pfas-national-primary-drinking-water-regulation>
- EPA Final PFAS National Primary Drinking Water Regulation Website: <https://www.epa.gov/sdwa/and-polyfluoroalkyl-substances-pfas>
- Final PFAS National Primary Drinking Water Regulation Monitoring and Reporting Factsheet: https://www.epa.gov/system/files/documents/2024-04/pfas-npdwr_fact-sheet_monitoring_4.8.24_0.pdf

¹ For data to be reliably reported below the UCMR 5 MRLs for PFAS, the laboratory should have generated initial method quality control data, in the form of calibration points at the lower levels and should have completed an Initial Demonstration of Capability (IDC) demonstrating successful performance at the lower levels, as described in Method 533 (Section 9.1) and Method 537.1 (Section 9.2). Furthermore, ongoing batch QC must be performed at the lower concentrations. Batch QC includes lower concentrations of initial continuing calibration checks (CCC) as described in Method 533 (Section 9.2.2 and 10.4) and Method 537.1 (Section 9.3.2 and 10.3), and a Laboratory Reagent Blank and Field Reagent Blank that passes the criteria of nothing observed above 1/3 the concentration of the lower reporting levels, as described in Method 533 (Sections 9.2.1 and 9.2.8) and Method 537.1 (Sections 9.3.1 and 9.3.8).

- UCMR 5 Resources:

<https://www.epa.gov/dwucmr/fifth-unregulated-contaminant-monitoring-rule>

Any questions about the information provided above may be addressed to PFASNPDWR@epa.gov.

cc: Association of State Drinking Water Administrators

Appendix D

Sample Locations

Appendix D - Sample Locations

County	PWS Type	PWS #	PWS Name	Source Type	Location Name	ID	AVAILABILITY_CODE	External Number	Water Type
HUMBOLDT	NC	NV0000915	A1 FUEL STOP	WL	WELL 1	W01	P	2311	GW
WASHOE	C	NV0000200	ALPINE MOBILE HOME PARK	WL	WELL 1	W01	P	56	GW
NYE	NC	NV0000155	AMARGOSA VALLEY RV PARK	WL	WELL	W01	P	2302	GW
CLARK	NTNC	NV0000379	APEX REGIONAL WASTE MGMT CENTER	WL	WELL 5 MW 5	W01	P	2074	GW
CLARK	C	NV0004092	BIG BEND WATER DISTRICT	IN	COLORADO RIVER INTAKE	IN01	P	1956	SW
NYE	C	NV0000369	BIG VALLEY MHP	WL	WELL 1	W01	P	509	GW
WASHOE	NC	NV0000710	BOWERS MANSION	WL	WELL	W01	P	3642	GW
WASHOE	C	NV0000277	CONESTOGA MHP	WL	WELL 1	W01	P	73	GW
WASHOE	NC	NV0000717	CROSBYS LODGE	WL	WELL 1	W01	P	95	GW
WASHOE	NC	NV0000718	CRYSTAL PEAK PARK	WL	WELL	W01	P	96	GW
WASHOE	NC	NV0000719	DAVIS CREEK PARK	WL	WELL	W01	P	97	GW
WASHOE	NC	NV0000736	DESERT INVESTMENT GROUP LTD	WL	SHOP WELL 1	W01	P	105	GW
WASHOE	NC	NV0000736	DESERT INVESTMENT GROUP LTD	WL	HOUSE WELL 2	W02	P	294	GW
CLARK	NC	NV0001008	DESERT SOUTHWEST CONFERENCE	WL	WELL 1	W01	P	1858	GW
WASHOE	C	NV0000042	EMPIRE MINING COMPANY	WL	WELL 7	W01	P	254	GW
WASHOE	C	NV0000042	EMPIRE MINING COMPANY	WL	WELL 10	W04	P	3387	GW
DOUGLAS	C	NV0002540	FAIRGROUNDS/SUNRISE ESTATES	WL	FAIRGROUNDS WELL 1	W01	P	654	GW
DOUGLAS	C	NV0002540	FAIRGROUNDS/SUNRISE ESTATES	WL	FAIRGROUNDS WELL 2	W02	P	6101	GW
DOUGLAS	C	NV0002540	FAIRGROUNDS/SUNRISE ESTATES	WL	FAIRGROUNDS WELL 3	W03	E	6102	GW
CHURCHILL	C	NV0000045	FALLON CITY OF	CC	CC TO FALLON NAVAL AIR STN NV0000350	CC01	P	5330	GW
CHURCHILL	C	NV0000045	FALLON CITY OF	WL	WELL 1	W01	P	414	GW
CHURCHILL	C	NV0000045	FALLON CITY OF	WL	WELL 2	W02	P	910	GW
CHURCHILL	C	NV0000045	FALLON CITY OF	WL	WELL 3	W03	P	1047	GW
CHURCHILL	C	NV0000045	FALLON CITY OF	WL	WELL 4	W04	P	1985	GW
CHURCHILL	NC	NV0002534	FALLON RV PARK	WL	WELL 1 EAST	W01	P	650	GW
CLARK	NTNC	NV0000372	GEORGIA PACIFIC GYPSUM LLC	WL	WELL 1	W01	P	2307	GW
WASHOE	NTNC	NV0000751	GOLD RANCH CASINO	WL	RV PARK WELL 2	W02	P	3644	GW
WASHOE	NTNC	NV0000751	GOLD RANCH CASINO	WL	WELL 3	W03	P	10261	GW
CLARK	NTNC	NV0001045	GOODSPRINGS ES CLARK COMMUNITY CENTER	WL	WELL 1	W01	P	147	GW
WASHOE	C	NV0000035	GRANDVIEW TERRACE WATER DISTRICT	WL	TANK WELL	W01	P	3	GW
WASHOE	C	NV0000035	GRANDVIEW TERRACE WATER DISTRICT	WL	GULCH WELL	W02	E	253	GW
WASHOE	C	NV0000035	GRANDVIEW TERRACE WATER DISTRICT	WL	MEDGAR WELL	W03	P	329	GW
CLARK	NC	NV0004073	IDLE SPURS LOUNGE AND RESTAURANT	WL	WELL	W01	P	2294	GW
WASHOE	C	NV0000158	INCLINE VILLAGE GID	IN	BURNT CEDAR LK TAHOE INTAKE	IN01	P	38	SW
WASHOE	NC	NV0000731	JOHNNYS RISTORANTE ITALIANO	WL	WELL 1	W01	P	103	GW
WASHOE	C	NV0000199	KE TA MOBILE HOME PARK	WL	WELL 1	W01	P	55	GW
WASHOE	C	NV0000199	KE TA MOBILE HOME PARK	WL	WELL 2	W02	E	5008	GW
ELKO	NTNC	NV0000928	LAMOILLE VALLEY PLAZA	WL	WELL 1	W01	P	2308	GW
WASHOE	NC	NV0000738	MAGIC CARPET GOLF	WL	WELL 1	W01	P	106	GW
WASHOE	NC	NV0000757	MELS DINER	WL	WELL 2	W02	P	6804	GW
CLARK	C	NV0000160	MOAPA VALLEY WATER DISTRICT	SP	BALDWIN SPRING	SP04	P	2298	GW
CLARK	C	NV0000160	MOAPA VALLEY WATER DISTRICT	SP	JONES SPRING	SP05	P	2320	GW
CLARK	C	NV0000160	MOAPA VALLEY WATER DISTRICT	WL	MX6 WELL	W01	S	333	GW
CLARK	C	NV0000160	MOAPA VALLEY WATER DISTRICT	WL	ARROW CANYON WELL 1	W02	P	350	GW
CLARK	C	NV0000160	MOAPA VALLEY WATER DISTRICT	WL	LOGANDALE WELL	W03	E	2312	GW

Appendix D - Table of Active Sources for PWS in Nevada

CLARK	C	NV0000160	MOAPA VALLEY WATER DISTRICT	WL	ARROW CANYON WELL 2	W06	P	5298	GW
WASHOE	C	NV0000732	MOUNT ROSE BOWL PROPERTY OWNERS WATER CO	SP	SPRING	SP02	P	3476	GW
WASHOE	C	NV0000732	MOUNT ROSE BOWL PROPERTY OWNERS WATER CO	WL	WELL 1	W01	E	1830	GW
ELKO	C	NV0000170	MOUNTAIN CITY WATER AND SEWER	SP	SPRING	SP01	P	3073	GW
WASHOE	NC	NV0000789	MSJK PROPERTIES LLC	WL	WELL 1	W01	P	127	GW
CLARK	NC	NV0001010	MT CHARLESTON PUB LLC	WL	MT CHARLESTON LODGE WELL	W01	P	140	GW
WASHOE	NC	NV0000743	MT ROSE CAMPGROUND USFS	WL	WELL 1	W01	P	6926	GW
WASHOE	NC	NV0000744	MT ROSE SKI TAHOE	SP	SPRING	SP01	P	3522	GW
CHURCHILL	NTNC	NV0002541	NAS CENTROID EW RANGE	WL	CENTROID WELL	W01	P	655	GW
WASHOE	NTNC	NV0000774	NATCHEZ SCHOOL	WL	WELL 1	W01	P	122	GW
WASHOE	NC	NV0004022	NDOT WADSWORTH REST STOP	WL	WELL	W01	P	193	GW
WASHOE	NC	NV0000720	NITECAP BAR	WL	WELL	W01	P	98	GW
WASHOE	NTNC	NV0000750	PLEASANT VALLEY SCHOOL	WL	WELL	W01	P	111	GW
CLARK	C	NV0000370	RAINBOW GARDENS WATER USERS	WL	WELL 1	W01	P	86	GW
WASHOE	C	NV0000701	RENO SAHARA TRAILER PARK	WL	WELL 1	W01	P	87	GW
WASHOE	C	NV0000701	RENO SAHARA TRAILER PARK	WL	WELL 2	W02	E	290	GW
WASHOE	C	NV0000191	RIO VISTA NV LLC	WL	WELL 1 OFFICE	W01	P	47	GW
WASHOE	C	NV0000191	RIO VISTA NV LLC	WL	WELL 2	W02	P	276	GW
WASHOE	NC	NV0003001	RIVERBELLE PROPERTIES LLC SASQUATCH INN	WL	WELL 2	W02	P	8103	GW
NYE	C	NV0004074	ROUND MOUNTAIN PUC	WL	WELL HW 1	W01	P	759	GW
NYE	C	NV0004074	ROUND MOUNTAIN PUC	WL	WELL HW 2	W02	P	991	GW
WASHOE	NC	NV0000746	SAND HARBOR STATE PARK	WL	WELL 1	W01	P	110	GW
WASHOE	NC	NV0000760	SKY TAVERN	SP	SPRING	SP01	P	3523	GW
WASHOE	C	NV0000211	SUN VALLEY GID	CC	CC FROM TMWA GOLDEN VALLEY RD	CC01	P	63	SW
WASHOE	C	NV0000211	SUN VALLEY GID	CC	CC FROM TMWA SUN VALLEY BLVD	CC02	P	8866	SW
WASHOE	C	NV0000765	SUTCLIFFE MOBILE PARK	WL	WELL 1	W01	P	296	GW
WASHOE	C	NV0000765	SUTCLIFFE MOBILE PARK	WL	WELL 2	W02	P	342	GW
CLARK	NC	NV0000329	T AND J INVESTMENTS DBA THE GROVE	WL	SILK PURSE RANCH WELL 1	W01	P	495	GW
CLARK	C	NV0004060	TROUT CANYON LAND AND WATER USERS ASSOC	WL	WELL 1	W01	P	7519	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	CC	CC FROM SVGID NV0211	CC01	E	9068	SW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	IN	CHALK BLUFF RAW WATER INTAKE	IN33	P	5316	SW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	IN	GLENDALE RAW WATER INTAKE	IN34	P	5317	SW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	IN	MT ROSE WATER INTAKE	IN35	P	8989	SW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	HOLCOMB WELL	W01	P	384	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	PATRIOT WELL	W02	P	4074	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	LONGLEY LN WELL	W03	P	4075	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	S VIRGINIA ST WELL	W04	P	4076	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	SIERRA PLAZA WELL	W05	P	4077	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	DELUCCHI WELL	W06	P	4078	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	LAKESIDE DR WELL	W07	P	4079	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	PEZZI WELL	W09	P	4080	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	GREG ST WELL	W10	P	4081	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	POPLAR ST WELL 2	W11	P	374	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	POPLAR ST WELL 1	W12	P	382	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	SPARKS AVE WELL	W13	P	4082	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	CORBETT WELL	W15	P	4083	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	TERMINAL WAY WELL	W16	P	371	GW

Appendix D - Table of Active Sources for PWS in Nevada

WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	MILL ST WELL	W17	P	366	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	S TWENTYFIRST ST WELL	W18	P	4084	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	GALLETTI WELL	W19	P	4085	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	HIGH STREET WELL	W20	P	376	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	MORRILL AVE WELL	W21	P	378	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	KIETZKE LN WELL	W22	P	4086	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	FOURTH ST WELL	W23	P	4087	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	VIEW ST WELL	W24	P	4088	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	EL RANCHO WELL	W25	P	4089	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	SWOPE WELL	W26	P	4090	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	HUNTER LAKE WELL	W27	P	4091	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	GLEN HARE WELL	W28	P	4092	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	RENO HIGH WELL	W29	P	4093	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	SILVER LAKE REPLACEMENT WELL	W33	P	5557	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	SILVER KNOLLS WELL	W34	P	5727	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	HAWKINGS COURT WELL	W36	P	5884	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	HUFFAKER PLACE WELL	W37	P	7888	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	INNOVATION WELL	W40	P	8092	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	ARROWCREEK WELL 1	W41	P	8144	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	ARROWCREEK WELL 2	W42	P	8145	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	ARROWCREEK WELL 3	W43	P	8146	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	TESSA WELL 1	W44	P	8147	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	TESSA WELL 2	W45	P	8148	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	DOUBLE DIAMOND WELL 1	W46	P	8152	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	LEMMON VALLEY WELL 5	W47	P	8296	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	HIDDEN VALLEY WELL 5	W49	P	8160	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	LEMMON VALLEY WELL 7	W50	P	8298	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	DESERT SPRINGS WELL 1	W51	P	7952	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	DESERT SPRINGS WELL 2	W52	P	7953	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	DESERT SPRINGS WELL 3	W53	P	8269	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	DESERT SPRINGS WELL 4	W54	P	8270	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	LEMMON VALLEY WELL 9	W56	P	8300	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	FISH SPRINGS WELL A	W57	P	8301	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	FISH SPRINGS WELL B	W58	P	8302	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	FISH SPRINGS WELL C	W59	P	8303	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	FISH SPRINGS WELL D	W60	P	8304	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	FISH SPRINGS WELL E	W61	P	8305	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	MT ROSE WELL 3	W62	P	8332	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	MT ROSE WELL 5	W63	P	8333	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	MT ROSE WELL 6	W64	P	8334	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	ST JAMES WELL 1	W65	P	8343	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	ST JAMES WELL 2	W66	P	8344	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	STMGID WELL 1	W67	P	8347	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	STMGID WELL 2	W68	P	8348	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	STMGID WELL 3	W69	P	8349	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	STMGID WELL 4	W70	P	8350	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	STMGID WELL 5	W71	P	8351	GW

Appendix D - Table of Active Sources for PWS in Nevada

WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	STMGID WELL 6	W72	P	8352	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	STMGID WELL 11	W73	P	8353	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	STMGID WELL 12	W74	P	8354	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	SPRING CREEK WELL 2	W75	P	8371	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	SPRING CREEK WELL 6	W77	P	8373	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	SPRING CREEK WELL 7	W78	P	8374	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	THOMAS CREEK WELL 1	W79	P	8391	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	STMGID WELL 7	W80	P	8393	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	DOUBLE DIAMOND WELL 3	W83	P	8837	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	WEST RENO WELL 7	W84	P	9703	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	WEST RENO WELL 8	W85	P	9704	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	WEST RENO WELL 10	W86	P	9705	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	WEST RENO WELL 12	W87	P	9706	GW
WASHOE	C	NV0000190	TRUCKEE MEADOWS WATER AUTHORITY	WL	SPRING CREEK WELL 8	W88	P	9753	GW
WASHOE	C	NV0000281	WADSWORTH MOBILE HOME PARK	WL	WELL 1	W01	P	76	GW
WASHOE	C	NV0000281	WADSWORTH MOBILE HOME PARK	WL	WELL 2	W02	E	2409	GW
WASHOE	NC	NV0000793	WASHOE LAKE STATE PARK	WL	WELL	W01	P	3651	GW
WASHOE	NC	NV0003094	WASHOE REGIONAL SHOOTING RANGE	WL	WELL	W01	P	188	GW
WASHOE	C	NV0000280	WEBB MOBILE HOME PARK	WL	WELL 1	W01	E	75	GW
WASHOE	C	NV0000280	WEBB MOBILE HOME PARK	WL	WELL 3	W03	P	5756	GW
WASHOE	NC	NV0000727	WHISPERING VINE	WL	WELL 1	W01	P	101	GW
CLARK	C	NV0000289	SOUTHERN NEVADA WATER SYSTEM	IN	SNWS INTAKE 3	IN03	P	7603	SW
WASHOE	C	NV0000282	STEAMBOAT SPRINGS WATERWORKS INC	WL	WELL 1	W01	P	1842	GW
WASHOE	C	NV0000282	STEAMBOAT SPRINGS WATERWORKS INC	WL	WELL 2	W02	P	1843	GW
WASHOE	C	NV0000196	VERDI MEADOWS UTILITY COMPANY INC	WL	WELL GW2	W02	P	2554	GW
WASHOE	C	NV0000196	VERDI MEADOWS UTILITY COMPANY INC	WL	WELL GW3	W03	P	4042	GW
WHITE PINE	NC	NV0004055	ALL IN ONE TRUCK STOP	WL	WELL 1	W01	P	755	GW
NYE	NTNC	NV0002190	AMARGOSA ELEMENTARY SCHOOL	CC	CC INTERTIE FROM AMARGOSA TWN CMLPX 5037	CC01	E	8940	GW
NYE	NTNC	NV0002190	AMARGOSA ELEMENTARY SCHOOL	WL	WELL 2	W02	P	9106	GW
NYE	NTNC	NV0003061	AMARGOSA SENIOR CENTER	WL	WELL 2	W02	P	5331	GW
NYE	C	NV0002558	AMARGOSA WATER COMPANY	WL	WELL 1	W01	P	668	GW
NYE	C	NV0002558	AMARGOSA WATER COMPANY	WL	WELL 2	W02	E	970	GW
NYE	C	NV0005033	ANCHOR IN MHP	WL	WELL	W01	P	786	GW
ELKO	NC	NV0002088	ANGEL CREEK CAMPGROUND USFS	WL	WELL	W01	P	576	GW
ELKO	NC	NV0003037	ANGEL LAKE CAMPGROUND USFS	SP	SPRING	SP01	P	3518	GW
NYE	NC	NV0003010	BAILEYS HOT SPRINGS	SP	SPRING	SP01	P	3137	GW
WHITE PINE	NC	NV0003052	BAKER CREEK CAMPGROUND NPS	SP	SPRING	SP01	P	3519	GW
NYE	NC	NV0005038	BEATTY RV PARK	WL	WELL 1	W01	P	791	GW
NYE	C	NV0000009	BEATTY WATER AND SANITATION DISTRICT	WL	WELL 2	W02	E	893	GW
NYE	C	NV0000009	BEATTY WATER AND SANITATION DISTRICT	WL	WELL 3	W03	E	1039	GW
NYE	C	NV0000009	BEATTY WATER AND SANITATION DISTRICT	WL	INDIAN SPRING WELL	W04	P	2385	GW
NYE	C	NV0000009	BEATTY WATER AND SANITATION DISTRICT	WL	SUMMIT WELL	W06	P	2386	GW
NYE	C	NV0000009	BEATTY WATER AND SANITATION DISTRICT	WL	WELL EW4	W07	P	3336	GW
LINCOLN	NC	NV0002120	BEAVER DAM STATE PARK	WL	WELL 1	W01	P	6767	GW
NYE	NC	NV0002151	BERLIN ICHTHYOSAUR STATE PARK	WL	WELL 1	W01	P	604	GW
CLARK	C	NV0000147	BERMUDA PALMS MANAGEMENT LLC	WL	WELL 2	W01	P	35	GW
NYE	C	NV0000362	BIG FIVE PARK	WL	WELL 1	W01	P	507	GW

Appendix D - Table of Active Sources for PWS in Nevada

CLARK	NTNC	NV0000323	BLUE DIAMOND RAINBOW SW PLAZA WATER	WL	WELL	W01	P	83	GW
CLARK	C	NV0000092	BLUE DIAMOND WATER CO OP INC	SP	COTTONWOOD SPRING	SP01	P	3328	GW
CLARK	NC	NV0000321	BRANDOS BLUE DIAMOND RD	WL	WELL	W01	P	82	GW
NYE	C	NV0002538	C VALLEY MHP	WL	WELL 1	W01	P	653	GW
NYE	C	NV0002538	C VALLEY MHP	WL	WELL 2	W02	P	967	GW
LINCOLN	C	NV0000013	CALIENTE PUBLIC UTILITIES	WL	WELL 1 RR WELL	W01	P	393	GW
LINCOLN	C	NV0000013	CALIENTE PUBLIC UTILITIES	WL	WELL 5	W05	P	6222	GW
LINCOLN	C	NV0000013	CALIENTE PUBLIC UTILITIES	WL	WELL 6	W06	P	6207	GW
CLARK	NC	NV0002509	CAMP LADY OF THE SNOWS	WL	WELL	W02	P	309	GW
CLARK	NC	NV0005009	CAMP LEE CANYON CCPR	WL	CAMP LEE CANYON WELL	W01	P	6387	GW
STOREY	C	NV0005056	CANYON GID	WL	WELL 1	W01	P	793	GW
STOREY	C	NV0005056	CANYON GID	WL	WELL 2	W02	P	4564	GW
ELKO	C	NV0000014	CARLIN UTILITIES	SP	SPRING	SP02	P	2540	GW
ELKO	C	NV0000014	CARLIN UTILITIES	WL	WELL 3	W01	P	394	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	CC	CC INTERTIE FROM DAYTON NV0032	CC01	P	6667	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	CC	CC INTERTIE FROM EAST VALLEY NV2216	CC02	P	6850	SW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	CC	CC INTERTIE FROM NORTH COUNTY NV0977	CC03	E	6854	SW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	IN	INTAKE FOR HOBART MARLETTE	IN28	P	3100	SW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	IN	INTAKE AT ASH CANYON	IN29	P	1212	SW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	IN	INTAKE AT KINGS CANYON	IN30	P	1209	SW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	SP	CUSTERS SPRINGS	SP37	P	5763	GU
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 33 BRUNSWICK	W01	P	1197	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 40 MORGAN MILL	W02	P	1810	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 44 EMPIRE RANCH RD	W03	P	2334	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 34 MILLS PARK	W08	P	1809	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 11B SECOND ST	W09	P	1194	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 11A SECOND ST	W10	P	1171	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 4A ELAINE ST	W11	P	896	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 45 SONOMA ST	W12	P	2348	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 8 SILVER SAGE	W13	P	1151	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 43 BENNETT AVE	W14	P	3055	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 38 S EDMONDS	W15	P	1202	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 9 SNYDER	W16	P	1161	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 6 FOOTHILL	W18	P	1104	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 5 BATH ST	W19	P	1041	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 46 SILVER OAK	W20	P	2349	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 48 THAMES LN	W25	P	2350	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 49 STEWART ST	W26	P	4361	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 51 FOOTHILL	W28	P	5462	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 53 SUNSET	W29	P	5463	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 54 FUJI	W30	P	5464	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 55 WNCC	W31	P	5465	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 10B COLLEGE PARKWAY (ASR)	W32	P	5469	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 25B RIVERVIEW PARK NORTH	W33	P	5471	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 16B LAKEVIEW	W34	P	5695	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 24B E 5TH ST	W35	P	5722	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 41B RIVERVIEW PARK SOUTH	W38	P	6280	GW

Appendix D - Table of Active Sources for PWS in Nevada

CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WELL 50	W39	P	6325	GW
CARSON CITY	C	NV0000015	CARSON CITY PUBLIC WORKS	WL	WINNIE LANE WELL 3 REPLACEMENT	W40	P	9461	GW
CHURCHILL	C	NV0003068	CARSON RIVER ESTATES	WL	WELL 1	W01	P	724	GW
CHURCHILL	C	NV0003068	CARSON RIVER ESTATES	WL	WELL 2	W02	P	981	GW
CHURCHILL	C	NV0003068	CARSON RIVER ESTATES	WL	WELL 3	W03	P	2355	GW
NYE	C	NV0000218	CARVERS SMOKEY VALLEY RV AND MHP	WL	WELL 1	W01	P	452	GW
WHITE PINE	NC	NV0004035	CAVE LAKE STATE PARK	SP	SPRING	SP01	P	3157	GW
DOUGLAS	C	NV0000259	CAVE ROCK SKYLAND	IN	LAKE TAHOE INTAKE	IN01	P	473	SW
NYE	NC	NV0004085	CHICKEN RANCH	WL	BACK WELL 1	W01	P	762	GW
NYE	NC	NV0004085	CHICKEN RANCH	WL	FRONT WELL 2	W02	P	992	GW
DOUGLAS	NTNC	NV0005078	CHINA SPRINGS YOUTH CAMP	WL	WELL 2	W02	P	5263	GW
NYE	C	NV0002554	CHIPMUNK RETREAT	WL	WELL 2	W02	P	9642	GW
LYON	NC	NV0005020	CHURCHILL BUTTE COMPLEX	WL	WELL 1	W01	P	778	GW
PERSHING	NTNC	NV0003076	COEUR ROCHESTER INC	WL	WELL PW-3A	W06	P	7183	GW
CLARK	C	NV0004000	COLD CREEK CANYON HOMEOWNERS	WL	WELL 1 UPPER WELL	W01	P	190	GW
CLARK	C	NV0004000	COLD CREEK CANYON HOMEOWNERS	WL	WELL 3 LOWER WELL	W03	P	7861	GW
WHITE PINE	C	NV0005057	COLD CREEK MHP	WL	COLD CREEK WELL 1A	W02	P	7990	GW
HUMBOLDT	NC	NV0002113	COMMUNITY HALL PARADISE VALLEY	WL	WELL 1	W01	P	587	GW
CARSON CITY	C	NV0000017	COTTONWOOD MHP	WL	NORTH WELL	W01	P	397	GW
CARSON CITY	C	NV0000017	COTTONWOOD MHP	WL	MIDDLE WELL	W02	P	898	GW
CARSON CITY	C	NV0000017	COTTONWOOD MHP	WL	SOUTH WELL	W03	P	1042	GW
NYE	C	NV0005032	COUNTRY VIEW ESTATES GBWC	WL	WELL 48 NORTH EAST	W01	P	785	GW
NYE	C	NV0005032	COUNTRY VIEW ESTATES GBWC	WL	WELL 48 A SOUTH WEST	W02	P	4096	GW
NYE	C	NV0005032	COUNTRY VIEW ESTATES GBWC	WL	CALVADA NORTH WELL	W03	P	5956	GW
CLARK	NTNC	NV0001081	CREECH AIR FORCE BASE	WL	WELL 106-4	W04	P	5677	GW
CLARK	NTNC	NV0001081	CREECH AIR FORCE BASE	WL	WELL 62-5	W05	P	5791	GW
CLARK	NTNC	NV0001081	CREECH AIR FORCE BASE	WL	WELL 3 REPLACEMENT	W06	P	10106	GW
EUREKA	C	NV0000043	CRESCENT VALLEY WATER SYSTEM	WL	WELL 4	W04	P	6150	GW
EUREKA	C	NV0000043	CRESCENT VALLEY WATER SYSTEM	WL	WELL 5	W05	P	6827	GW
LYON	C	NV0000032	DAYTON VALLEY WATER SYSTEM	CC	CC INTERTIE FROM CARSON CITY NV0015	CC01	P	5992	GW
LYON	C	NV0000032	DAYTON VALLEY WATER SYSTEM	WL	WELL 2	W02	P	903	GW
LYON	C	NV0000032	DAYTON VALLEY WATER SYSTEM	WL	WELL 3	W03	P	1043	GW
LYON	C	NV0000032	DAYTON VALLEY WATER SYSTEM	WL	WELL 4	W04	P	3281	GW
LYON	C	NV0000032	DAYTON VALLEY WATER SYSTEM	WL	WELL 5 0029-04	W05	P	5381	GW
LYON	C	NV0000032	DAYTON VALLEY WATER SYSTEM	WL	WELL 6 0029-01	W06	P	5382	GW
LYON	C	NV0000032	DAYTON VALLEY WATER SYSTEM	WL	WELL 7 0029-02	W07	P	5383	GW
LYON	C	NV0000032	DAYTON VALLEY WATER SYSTEM	WL	WELL 8 CARDELLI	W08	P	5377	GW
LYON	C	NV0000032	DAYTON VALLEY WATER SYSTEM	WL	WELL 20 ROLLING A	W20	P	5696	GW
LYON	C	NV0000032	DAYTON VALLEY WATER SYSTEM	WL	WELL 21 ALAMOSA ST	W21	P	8543	GW
NYE	C	NV0000831	DESERT MIRAGE HOMEOWNERS ASSOCIATION	WL	WELL 2	W02	P	2397	GW
NYE	C	NV0000831	DESERT MIRAGE HOMEOWNERS ASSOCIATION	WL	WELL 3	W03	P	10115	GW
NYE	C	NV0000300	DESERT UTILITIES	WL	WELL 1	W01	P	483	GW
NYE	C	NV0000300	DESERT UTILITIES	WL	WELL 2	W02	P	3356	GW
NYE	C	NV0000300	DESERT UTILITIES	WL	WELL 4	W04	P	3745	GW
EUREKA	C	NV0002573	DEVILS GATE WATER SYSTEM GID 2	CC	INTERTIE FROM DISTRICT 1 NV0002574	CC01	P	6182	GW
ELKO	NC	NV0004096	DOUBLE D BAR & GRILL	WL	WELL 1	W01	P	764	GW
DOUGLAS	C	NV0002216	DOUGLAS COUNTY CARSON VALLEY SYSTEM	CC	CC INTERTIE FROM MINDEN NV0168	CC01	P	6086	GW

Appendix D - Table of Active Sources for PWS in Nevada

DOUGLAS	C	NV0002216	DOUGLAS COUNTY CARSON VALLEY SYSTEM	CC	CC INTERTIE FROM CARSON CITY NV0015	CC02	E	6849	GW
DOUGLAS	C	NV0002216	DOUGLAS COUNTY CARSON VALLEY SYSTEM	WL	AIRPORT SOUTH WELL	W04	E	4005	GW
DOUGLAS	C	NV0002216	DOUGLAS COUNTY CARSON VALLEY SYSTEM	WL	NORTH COUNTY WELL 1 TOPSY	W05	P	10145	GW
DOUGLAS	C	NV0002216	DOUGLAS COUNTY CARSON VALLEY SYSTEM	WL	NORTH COUNTY WELL 2	W06	P	10146	GW
DOUGLAS	C	NV0002216	DOUGLAS COUNTY CARSON VALLEY SYSTEM	WL	GENOA LAKES WELL 2	W07	P	10154	GW
DOUGLAS	C	NV0002216	DOUGLAS COUNTY CARSON VALLEY SYSTEM	WL	WALLEYS WELL	W08	P	10155	GW
DOUGLAS	C	NV0002216	DOUGLAS COUNTY CARSON VALLEY SYSTEM	WL	MONTANA SMALL WELL 2	W09	P	10163	GW
DOUGLAS	C	NV0002216	DOUGLAS COUNTY CARSON VALLEY SYSTEM	WL	MONTANA WELL 3	W10	P	10164	GW
DOUGLAS	NC	NV0002043	DOUGLAS COUNTY LAMPE PARK	WL	WELL 1	W01	P	553	GW
DOUGLAS	NC	NV0002044	DOUGLAS COUNTY TOPAZ PARK	WL	WELL 1	W01	P	554	GW
LYON	NC	NV0002593	DRESSLER PARK	WL	WELL 1	W01	P	685	GW
PERSHING	C	NV0000809	DUTCHMAN ACRES	WL	OLD WELL	W01	P	515	GW
PERSHING	C	NV0000809	DUTCHMAN ACRES	WL	NEW WELL	W02	P	1974	GW
LINCOLN	NC	NV0000188	EAGLE VALLEY RESORT	WL	WELL 1	W01	P	450	GW
LINCOLN	NC	NV0000188	EAGLE VALLEY RESORT	WL	WELL 2	W02	P	3508	GW
LYON	NC	NV0003098	EAST 50 BAR	WL	WELL 1	W01	P	735	GW
DOUGLAS	NC	NV0002218	EAST PEAK LODGE	WL	WELL	W01	P	634	GW
CLARK	C	NV0000106	EASTLAND HEIGHTS WATER ASSOCIATION	WL	PLEASANT WELL	W01	P	17	GW
CLARK	C	NV0000106	EASTLAND HEIGHTS WATER ASSOCIATION	WL	VALLEY WELL	W02	P	261	GW
LINCOLN	NC	NV0002123	ECHO CANYON STATE PARK	WL	WELL 1	W01	P	590	GW
DOUGLAS	NTNC	NV0000235	EDGEWOOD WATER COMPANY	CC	CC INTERTIE FROM KGID NV0004	CC01	E	7886	SW
DOUGLAS	NTNC	NV0000235	EDGEWOOD WATER COMPANY	IN	LAKE TAHOE INTAKE	IN01	P	458	SW
DOUGLAS	C	NV0000271	ELK POINT COUNTRY CLUB	WL	WELL 1	W01	P	480	GW
DOUGLAS	C	NV0000271	ELK POINT COUNTRY CLUB	WL	WELL 2	W02	P	2078	GW
ELKO	C	NV0000272	ELKO CITY OF	WL	WELL 38	W01	P	2362	GW
ELKO	C	NV0000272	ELKO CITY OF	WL	WELL 27	W02	P	1119	GW
ELKO	C	NV0000272	ELKO CITY OF	WL	WELL 31	W03	P	1141	GW
ELKO	C	NV0000272	ELKO CITY OF	WL	WELL 36	W04	P	2363	GW
ELKO	C	NV0000272	ELKO CITY OF	WL	WELL 37	W06	P	2365	GW
ELKO	C	NV0000272	ELKO CITY OF	WL	WELL 13	W08	P	1177	GW
ELKO	C	NV0000272	ELKO CITY OF	WL	WELL 12	W09	P	481	GW
ELKO	C	NV0000272	ELKO CITY OF	WL	WELL 14	W10	P	1182	GW
ELKO	C	NV0000272	ELKO CITY OF	WL	WELL 10A	W11	P	1167	GW
ELKO	C	NV0000272	ELKO CITY OF	WL	WELL 30	W12	P	1210	GW
ELKO	C	NV0000272	ELKO CITY OF	WL	WELL 15	W13	P	939	GW
ELKO	C	NV0000272	ELKO CITY OF	WL	WELL 20	W14	P	1198	GW
ELKO	C	NV0000272	ELKO CITY OF	WL	WELL 18	W15	P	1192	GW
ELKO	C	NV0000272	ELKO CITY OF	WL	WELL 24	W17	P	1200	GW
ELKO	C	NV0000272	ELKO CITY OF	WL	WELL I 96	W18	P	3337	GW
ELKO	C	NV0000272	ELKO CITY OF	WL	WELL 42	W19	P	5790	GW
ELKO	C	NV0000272	ELKO CITY OF	WL	WELL 43	W20	P	6041	GW
WHITE PINE	NTNC	NV0004083	ELY CONSERVATION CAMP NDOC	WL	WELL 1	W01	P	760	GW
WHITE PINE	C	NV0005071	ELY MAXIMUM SECURITY PRISON NDOC	WL	WELL 1	W01	P	803	GW
WHITE PINE	C	NV0005071	ELY MAXIMUM SECURITY PRISON NDOC	WL	WELL 2	W02	P	1004	GW
WHITE PINE	C	NV0000038	ELY MUNICIPAL WATER DEPARTMENT	WL	NORTH ST WELL	W02	P	1045	GW
WHITE PINE	C	NV0000038	ELY MUNICIPAL WATER DEPARTMENT	WL	10TH AND M ST WELL	W03	P	907	GW
WHITE PINE	C	NV0000038	ELY MUNICIPAL WATER DEPARTMENT	WL	17TH AND M ST WELL	W04	P	2383	GW

Appendix D - Table of Active Sources for PWS in Nevada

WHITE PINE	C	NV0000038	ELY MUNICIPAL WATER DEPARTMENT	WL	TERRACE WELL PWCOE 09-02	W05	P	6228	GW
WHITE PINE	C	NV0000038	ELY MUNICIPAL WATER DEPARTMENT	WL	GOLF COURSE WELL PWCOE 09-05	W06	P	6235	GW
WHITE PINE	C	NV0000038	ELY MUNICIPAL WATER DEPARTMENT	WL	WELL RW 7P	W08	P	6429	GW
STOREY	NTNC	NV0003075	EP MINERALS LLC CLARK	WL	WELL 1	W01	P	729	GW
CLARK	C	NV0000109	EQUESTRIAN ESTATES CO OP WATER ASSOC	WL	WELL 1 EAST	W01	P	19	GW
CLARK	C	NV0000109	EQUESTRIAN ESTATES CO OP WATER ASSOC	WL	WELL 2 WEST	W02	P	263	GW
NYE	C	NV0002552	ESCAPEE CO OP OF NEVADA	WL	WELL 3	W03	P	5766	GW
NYE	C	NV0002552	ESCAPEE CO OP OF NEVADA	WL	WELL 4	W04	P	7620	GW
EUREKA	C	NV0000044	EUREKA WATER ASSOCIATION	SP	FRED SPRING	SP01	P	6908	GW
EUREKA	C	NV0000044	EUREKA WATER ASSOCIATION	SP	LUCKY SPRING	SP02	P	6909	GW
EUREKA	C	NV0000044	EUREKA WATER ASSOCIATION	SP	MIDDLE SPRING	SP03	P	2316	GW
EUREKA	C	NV0000044	EUREKA WATER ASSOCIATION	SP	LANIE SPRING	SP05	P	2321	GW
EUREKA	C	NV0000044	EUREKA WATER ASSOCIATION	SP	GORMAN SPRING	SP06	P	2322	GW
EUREKA	C	NV0000044	EUREKA WATER ASSOCIATION	SP	FLORIO SPRING	SP07	P	2323	GW
EUREKA	C	NV0000044	EUREKA WATER ASSOCIATION	SP	RICHMOND SPRING	SP08	P	6518	GW
EUREKA	C	NV0000044	EUREKA WATER ASSOCIATION	SP	SUMMIT SPRING	SP09	P	6519	GW
EUREKA	C	NV0000044	EUREKA WATER ASSOCIATION	SP	PROSPECT SPRING	SP10	P	6910	GW
EUREKA	C	NV0000044	EUREKA WATER ASSOCIATION	SP	BULLWACKER SPRING	SP11	P	6911	GW
EUREKA	C	NV0000044	EUREKA WATER ASSOCIATION	WL	WELL 1	W01	P	909	GW
EUREKA	C	NV0000044	EUREKA WATER ASSOCIATION	WL	WELL 2	W02	P	1046	GW
CHURCHILL	NC	NV0002587	FALLON FOOD MART	WL	WELL 1	W01	P	681	GW
CHURCHILL	C	NV0000350	FALLON NAVAL AIR STATION	CC	CC FROM CITY OF FALLON NV0000045	CC01	P	5328	GW
CHURCHILL	C	NV0000350	FALLON NAVAL AIR STATION	WL	WELL 1	W01	P	498	GW
CHURCHILL	C	NV0000350	FALLON NAVAL AIR STATION	WL	WELL 2	W02	P	950	GW
CHURCHILL	C	NV0000350	FALLON NAVAL AIR STATION	WL	WELL 3	W03	P	1070	GW
LYON	C	NV0000062	FERNLEY PUBLIC WORKS	WL	WELL 4	W01	P	915	GW
LYON	C	NV0000062	FERNLEY PUBLIC WORKS	WL	WELL 9A	W02	P	2221	GW
LYON	C	NV0000062	FERNLEY PUBLIC WORKS	WL	WELL 9	W03	P	1135	GW
LYON	C	NV0000062	FERNLEY PUBLIC WORKS	WL	WELL 11 SHOP WELL	W04	P	3283	GW
LYON	C	NV0000062	FERNLEY PUBLIC WORKS	WL	WELL 13	W05	P	5509	GW
LYON	C	NV0000062	FERNLEY PUBLIC WORKS	WL	WELL 14	W06	P	5697	GW
WHITE PINE	NC	NV0002170	FIRESIDE INN	WL	WELL	W01	P	612	GW
CLARK	NC	NV0001053	FLOYD LAMB PARK	WL	WELL 171A	W03	P	9838	GW
LYON	NC	NV0002132	FORT CHURCHILL HISTORIC STATE PARK	WL	WELL 1	W01	P	594	GW
DOUGLAS	C	NV0000066	GARDNERVILLE RANCHOS GID	WL	WELL 1	W01	P	428	GW
DOUGLAS	C	NV0000066	GARDNERVILLE RANCHOS GID	WL	WELL 5	W05	P	1136	GW
DOUGLAS	C	NV0000066	GARDNERVILLE RANCHOS GID	WL	WELL 6	W06	P	1152	GW
DOUGLAS	C	NV0000066	GARDNERVILLE RANCHOS GID	WL	WELL 8	W08	P	3063	GW
DOUGLAS	C	NV0000066	GARDNERVILLE RANCHOS GID	WL	WELL 9 ROCKY TERRACES ESTATES	W09	P	5483	GW
DOUGLAS	C	NV0000066	GARDNERVILLE RANCHOS GID	WL	WELL 2A	W10	P	7764	GW
DOUGLAS	C	NV0000066	GARDNERVILLE RANCHOS GID	WL	WELL 4A	W11	P	8897	GW
DOUGLAS	C	NV0000065	GARDNERVILLE WATER COMPANY	CC	CC INTERTIE FROM TOWN OF MINDEN NV0168	CC01	E	7197	GW
DOUGLAS	C	NV0000065	GARDNERVILLE WATER COMPANY	WL	WELL 1	W01	P	427	GW
DOUGLAS	C	NV0000065	GARDNERVILLE WATER COMPANY	WL	WELL 2	W02	P	916	GW
DOUGLAS	C	NV0000065	GARDNERVILLE WATER COMPANY	WL	WELL 3	W03	P	1049	GW
DOUGLAS	C	NV0000065	GARDNERVILLE WATER COMPANY	WL	WELL 4	W04	P	1107	GW
DOUGLAS	C	NV0000065	GARDNERVILLE WATER COMPANY	WL	WELL 6	W06	P	2080	GW

Appendix D- Table of Active Sources for PWS in Nevada

DOUGLAS	C	NV0000065	GARDNERVILLE WATER COMPANY	WL	WELL 7	W07	P	2081	GW
DOUGLAS	C	NV0000065	GARDNERVILLE WATER COMPANY	WL	WELL 9	W09	P	2083	GW
CLARK	NC	NV0003029	GIRL SCOUTS NV CAMP FOXTAIL	WL	WELL 1	W01	P	2542	GW
DOUGLAS	C	NV0000356	GLENBROOK WATER COOPERATIVE INC	IN	LAKE TAHOE INTAKE	IN01	P	502	SW
HUMBOLDT	C	NV0005029	GOLCONDA GID	SP	POLE SPRINGS	SP02	P	2333	GW
HUMBOLDT	C	NV0005029	GOLCONDA GID	WL	POLE CREEK WELL	W01	P	784	GW
HUMBOLDT	C	NV0005029	GOLCONDA GID	WL	I-80 WELL	W03	E	4070	GW
HUMBOLDT	C	NV0003079	GOLD COUNTRY ESTATES	CC	CC INTERTIE FROM STAR CITY NV0252	CC02	E	7815	GW
HUMBOLDT	C	NV0003079	GOLD COUNTRY ESTATES	WL	WELL 1	W01	P	731	GW
HUMBOLDT	C	NV0003079	GOLD COUNTRY ESTATES	WL	WELL 2	W02	P	984	GW
HUMBOLDT	C	NV0003079	GOLD COUNTRY ESTATES	WL	DOMESTIC WELL 3	W03	P	7066	GW
HUMBOLDT	C	NV0003079	GOLD COUNTRY ESTATES	WL	FIRE WELL 4	W04	P	7067	GW
ESMERALDA	C	NV0000072	GOLDFIELD TOWN WATER	WL	WELL 2 K2	W02	P	918	GW
ESMERALDA	C	NV0000072	GOLDFIELD TOWN WATER	WL	WELL 3 K3	W03	P	9760	GW
ESMERALDA	C	NV0000072	GOLDFIELD TOWN WATER	WL	WELL 4 K4	W04	P	9761	GW
ESMERALDA	C	NV0000072	GOLDFIELD TOWN WATER	WL	WELL 1 REPLACEMENT K1R	W05	P	9762	GW
WASHOE	C	NV0000207	GREAT BASIN WATER CO COLD SPRINGS	WL	WELL 6	W03	P	338	GW
WASHOE	C	NV0000207	GREAT BASIN WATER CO COLD SPRINGS	WL	WELL 7	W04	P	2401	GW
WASHOE	C	NV0000207	GREAT BASIN WATER CO COLD SPRINGS	WL	SWEGER WELL (WELL 8)	W05	P	3094	GW
WASHOE	C	NV0000207	GREAT BASIN WATER CO COLD SPRINGS	WL	WELL 1A	W06	P	4552	GW
WASHOE	C	NV0000207	GREAT BASIN WATER CO COLD SPRINGS	WL	VANDYKE WELL	W07	P	8502	GW
WASHOE	C	NV0001086	GREAT BASIN WATER CO SPANISH SPRINGS	CC	CC FROM TWMA NV0190	CC01	E	9420	SW
WASHOE	C	NV0001086	GREAT BASIN WATER CO SPANISH SPRINGS	WL	WELL SS6	W01	P	2059	GW
WASHOE	C	NV0001086	GREAT BASIN WATER CO SPANISH SPRINGS	WL	WELL SS1A	W02	P	2061	GW
ELKO	C	NV0000036	GREAT BASIN WATER CO SPRING CREEK	WL	WELL 8	W01	P	408	GW
ELKO	C	NV0000036	GREAT BASIN WATER CO SPRING CREEK	WL	WELL 10	W02	P	905	GW
ELKO	C	NV0000036	GREAT BASIN WATER CO SPRING CREEK	WL	WELL 14	W03	P	1044	GW
ELKO	C	NV0000036	GREAT BASIN WATER CO SPRING CREEK	WL	WELL 4	W04	P	1105	GW
ELKO	C	NV0000036	GREAT BASIN WATER CO SPRING CREEK	WL	WELL 5	W05	P	2120	GW
ELKO	C	NV0000036	GREAT BASIN WATER CO SPRING CREEK	WL	WELL 12	W06	P	2121	GW
ELKO	C	NV0000036	GREAT BASIN WATER CO SPRING CREEK	WL	WHEAR WELL 7	W07	P	2994	GW
ELKO	C	NV0000036	GREAT BASIN WATER CO SPRING CREEK	WL	CHARLWOOD WELL	W08	P	5690	GW
ELKO	C	NV0000036	GREAT BASIN WATER CO SPRING CREEK	WL	WILLINGTON WELL 9	W09	P	5913	GW
ELKO	C	NV0005027	GREAT BASIN WATER CO SPRING CREEK MHP	WL	WELL SC 1	W01	P	782	GW
ELKO	C	NV0005027	GREAT BASIN WATER CO SPRING CREEK MHP	WL	WELL 11	W02	P	1000	GW
ELKO	C	NV0005027	GREAT BASIN WATER CO SPRING CREEK MHP	WL	WELL SC 3	W03	S	2117	GW
NYE	C	NV0000270	GREAT BASIN WATER COMPANY	WL	WELL 1	W01	P	479	GW
NYE	C	NV0000270	GREAT BASIN WATER COMPANY	WL	WELL 2	W02	P	938	GW
NYE	C	NV0000270	GREAT BASIN WATER COMPANY	WL	WELL 11	W04	P	1118	GW
NYE	C	NV0000270	GREAT BASIN WATER COMPANY	WL	WELL 9	W05	P	1140	GW
NYE	C	NV0000270	GREAT BASIN WATER COMPANY	WL	WELL 12	W06	P	8573	GW
LINCOLN	NC	NV0002119	GREEN VALLEY GROCERY	SP	SPRING	SP01	P	3152	GW
CLARK	C	NV0001080	HARRIS SPRINGS RANCH	WL	WELL 2	W02	P	7843	GW
MINERAL	C	NV0000357	HAWTHORNE ARMY DEPOT	CC	EMERGENCY INTERTIE TO HAWTHORNE 0073	CC01	I	6257	GW
MINERAL	C	NV0000357	HAWTHORNE ARMY DEPOT	WL	WELL 4 W02	W02	P	1188	GW
MINERAL	C	NV0000357	HAWTHORNE ARMY DEPOT	WL	WELL 11 W03	W03	P	4359	GW
MINERAL	C	NV0000073	HAWTHORNE UTILITIES	WL	WHISKEY FLAT WELL 2	W01	P	1816	GW

Appendix D - Table of Active Sources for PWS in Nevada

MINERAL	C	NV0000073	HAWTHORNE UTILITIES	WL	I ST WELL	W02	P	1817	GW
MINERAL	C	NV0000073	HAWTHORNE UTILITIES	WL	WHISKEY FLAT WELL 1	W03	P	1818	GW
MINERAL	C	NV0000073	HAWTHORNE UTILITIES	WL	WHISKEY FLAT WELL 3	W05	P	3052	GW
MINERAL	C	NV0000073	HAWTHORNE UTILITIES	WL	BABBITT WELL 7	W07	P	6246	GW
CLARK	C	NV0000076	HENDERSON CITY OF	CC	CC FROM AMS TP AT SNWS NV0000289	CC01	P	255	SW
CLARK	C	NV0000076	HENDERSON CITY OF	CC	CC FROM RM TP AT SNWS NV0000289	CC02	P	5550	SW
LYON	NC	NV0004037	HEYDAY INN	WL	WELL	W01	P	743	GW
CLARK	C	NV0000145	HILLCREST MANOR WATER USERS ASSOCIATION	WL	WELL 1B MADRE MESA	W01	P	33	GW
CLARK	C	NV0000145	HILLCREST MANOR WATER USERS ASSOCIATION	WL	WELL 2B SHEILA	W02	P	268	GW
LYON	NTNC	NV0004053	HODGES TRANSPORTATION CO	WL	WELL 1 NORTH	W01	P	754	GW
LYON	NTNC	NV0004053	HODGES TRANSPORTATION CO	WL	WELL 2 SOUTH	W02	P	988	GW
LYON	NTNC	NV0004053	HODGES TRANSPORTATION CO	WL	WELL 3 HOUSE	W03	P	1080	GW
DOUGLAS	C	NV0002046	HOLBROOK STATION RV AND MHP	WL	OSBORN WELL	W01	P	556	GW
DOUGLAS	C	NV0002046	HOLBROOK STATION RV AND MHP	WL	MOOSBERG WELL	W03	P	7056	GW
CLARK	NTNC	NV0003006	HOOVER DAM	IN	HOOVER DAM INTAKE ARIZONA TOWER 6	IN01	E	179	SW
CLARK	NTNC	NV0003006	HOOVER DAM	IN	HOOVER DAM INTAKE ARIZONA TOWER 3	IN02	P	4929	SW
NYE	NC	NV0000833	HORIZON C STORES 1 PAHRUMP VALLEY BLVD	WL	WELL	W01	P	528	GW
NYE	NC	NV0000827	HORIZON C STORES 2 TUMBLEWEED TAVERN	WL	WELL	W01	P	138	GW
PERSHING	C	NV0000226	IMLAY WATER SYSTEM	WL	WELL 1	W01	P	2558	GW
PERSHING	C	NV0000226	IMLAY WATER SYSTEM	WL	WELL 2 BACKUP WELL	W02	P	4003	GW
DOUGLAS	C	NV0000355	INDIAN HILLS GID	CC	CC INTERTIE FROM NORTH COUNTY NV0977	CC01	E	6396	GW
DOUGLAS	C	NV0000355	INDIAN HILLS GID	CC	CC INTERTIE FROM EAST VALLEY NV2216	CC02	E	6717	GW
DOUGLAS	C	NV0000355	INDIAN HILLS GID	WL	JVES NORTH WELL	W09	P	3066	GW
DOUGLAS	C	NV0000355	INDIAN HILLS GID	WL	JVES SOUTH WELL	W10	P	3067	GW
DOUGLAS	C	NV0000355	INDIAN HILLS GID	WL	RIDGEVIEW WELL 5	W11	P	3748	GW
CLARK	C	NV0000082	INDIAN SPRINGS WATER CO INC	WL	WELL 1	W01	P	6	GW
CLARK	C	NV0000082	INDIAN SPRINGS WATER CO INC	WL	WELL 2	W02	P	2356	GW
ELKO	C	NV0000088	JACKPOT WATER SYSTEM	WL	FIRE HOUSE WELL	W01	E	436	GW
ELKO	C	NV0000088	JACKPOT WATER SYSTEM	WL	CACTUS PETE WELL	W03	E	1052	GW
ELKO	C	NV0000088	JACKPOT WATER SYSTEM	WL	GOLF COURSE WELL	W04	E	1110	GW
ELKO	C	NV0000088	JACKPOT WATER SYSTEM	WL	PARK WELL	W05	P	2368	GW
ELKO	C	NV0000088	JACKPOT WATER SYSTEM	WL	WELL 6	W06	P	6323	GW
ELKO	C	NV0002070	JARBIDGE WATER SYSTEM	IN	BEAR CREEK INTAKE	IN01	P	566	SW
ELKO	C	NV0002070	JARBIDGE WATER SYSTEM	IN	JARBIDGE RIVER EMERGENCY INTAKE	IN02	E	6428	SW
CLARK	C	NV0000089	JEAN UTILITY SERVICES INC	WL	MIDWAY WELL J4	W01	P	9	GW
CLARK	C	NV0000089	JEAN UTILITY SERVICES INC	WL	STATE WELL J2	W03	P	330	GW
CLARK	C	NV0000089	JEAN UTILITY SERVICES INC	WL	GOLD STRIKE WELL	W04	P	349	GW
ELKO	NC	NV0002071	JIGGS BAR	WL	WELL 1	W01	P	567	GW
CHURCHILL	NTNC	NV0002024	KENNAMETAL INC	WL	WELL 1	W01	P	545	GW
CLARK	C	NV0000287	KINGS ROW TP	CC	CONSECUTIVE CONNECTION LVVWD	CC01	E	7734	SW
CLARK	C	NV0000287	KINGS ROW TP	WL	WELL	W01	P	78	GW
DOUGLAS	C	NV0000004	KINGSBURY GID	IN	LAKE TAHOE INTAKE STATION 1 RAW	IN02	P	889	SW
LANDER	C	NV0000265	KINGSTON TOWN WATER UTILITIES	WL	BACKUP WELL 1	W01	P	476	GW
LANDER	C	NV0000265	KINGSTON TOWN WATER UTILITIES	WL	MAIN WELL 2	W02	P	936	GW
WHITE PINE	C	NV0002174	KOA OF ELY CAMPGROUND	WL	WELL 1	W01	P	614	GW
WHITE PINE	C	NV0002174	KOA OF ELY CAMPGROUND	WL	WELL 2	W02	P	3068	GW
CLARK	C	NV0000142	KYLE CANYON WATER DISTRICT	WL	ECHO WELL 3	W01	P	32	GW

Appendix D - Table of Active Sources for PWS in Nevada

CLARK	C	NV0000142	KYLE CANYON WATER DISTRICT	WL	RAINBOW WELL	W02	P	267	GW
CLARK	C	NV0000142	KYLE CANYON WATER DISTRICT	WL	ECHO WELL 4	W03	P	3931	GW
CLARK	C	NV0000142	KYLE CANYON WATER DISTRICT	WL	ECHO WELL 5	W04	P	4848	GW
CHURCHILL	NC	NV0002028	LAHONTAN DAM STATE PARK	SP	SOUTH SPRING	SP01	P	4004	GU
CHURCHILL	NC	NV0002028	LAHONTAN DAM STATE PARK	SP	NORTH SPRING	SP02	P	2447	GU
LYON	NC	NV0002133	LAHONTAN DAM STATE PARK BEACH 3	WL	WELL 1	W01	P	595	GW
LYON	NC	NV0005021	LAHONTAN DAM STATE PARK BEACH 7	WL	WELL 1	W01	P	779	GW
ELKO	C	NV0000273	LAMOILLE WATER USERS ASSOC	WL	WELL 1	W01	P	482	GW
ELKO	C	NV0000273	LAMOILLE WATER USERS ASSOC	WL	WELL 2	W02	P	7086	GW
LANDER	C	NV0000008	LANDER CO SEWER AND WATER DIST 1 BM	WL	OLD WELL 6	W01	S	391	GW
LANDER	C	NV0000008	LANDER CO SEWER AND WATER DIST 1 BM	WL	WELL 8	W04	P	6220	GW
LANDER	C	NV0000008	LANDER CO SEWER AND WATER DIST 1 BM	WL	WELL 7	W05	P	6221	GW
LANDER	C	NV0000008	LANDER CO SEWER AND WATER DIST 1 BM	WL	WELL 9	W06	P	7318	GW
LANDER	C	NV0000006	LANDER CO SEWER AND WATER DIST 2 AUSTIN	SP	PONY CANYON SPRINGS	SP05	P	2431	GW
LANDER	C	NV0000006	LANDER CO SEWER AND WATER DIST 2 AUSTIN	WL	VALLEY WELL 1	W02	E	2165	GW
LANDER	C	NV0000006	LANDER CO SEWER AND WATER DIST 2 AUSTIN	WL	REESE RIVER VALLEY WELL	W05	P	6934	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	CC	CC FROM AMS TP AT SNWS NV0000289	CC01	P	10	SW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	CC	CC FROM RM TP AT SNWS NV0000289	CC02	P	5542	SW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 7A CHARLESTON HTS 2168 ZONE	W01	P	3246	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 68 CHARLESTON HTS 2168 ZONE	W02	P	3373	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 81 CHARLESTON HTS 2168 ZONE	W03	P	3254	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 82 CHARLESTON HTS 2168 ZONE	W04	P	3255	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 83 CHARLESTON HTS 2168 ZONE	W05	P	3855	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 84 CHARLESTON HTS 2168 ZONE	W06	P	3856	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 13 CHARLESTON HTS 2168 ZONE	W07	P	3401	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 15A CHARLESTON HTS 2168 ZONE	W08	P	3247	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 34 CHARLESTON HTS 2168 ZONE	W09	P	3406	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 80 CHARLESTON HTS 2168 ZONE	W11	P	3408	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 14 CHARLESTON HTS 2168 ZONE	W12	P	3384	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 16 CHARLESTON HTS 2168 ZONE	W13	P	3402	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 17 CHARLESTON HTS 2168 ZONE	W14	P	3248	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 1A 2538 ZONE	W17	P	3398	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 24 2538 ZONE	W18	P	3251	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 89 2538 ZONE	W20	P	3370	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 86 RICE 2420 ZONE	W23	P	3846	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 76 2635 ZONE	W25	P	259	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 87 2745 ZONE	W26	E	3374	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 93 2745 ZONE	W27	P	3375	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 97 2635 ZONE	W28	P	3847	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 105 2538 ZONE	W29	P	3412	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 2A 2538 ZONE	W30	P	3399	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 5A 2538 ZONE	W31	P	3400	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 18A 2538 ZONE	W32	P	3403	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 77 RONZONE NORTH 2300 ZONE	W33	P	3364	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 85 RONZONE SOUTH 2300 ZONE	W34	P	3377	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 45 2300 ZONE	W35	P	3378	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 22A LUCE 2420 ZONE	W38	P	3376	GW

Appendix D - Table of Active Sources for PWS in Nevada

CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 23A LUCE 2420 ZONE	W39	P	3250	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 73 2420 ZONE	W40	P	3366	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 88 2538 ZONE	W41	P	3409	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 102 2635 ZONE	W42	P	3363	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 116 2420 ZONE	W43	E	3372	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 28 GOWAN 2420 ZONE	W44	P	3252	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 33 GOWAN 2420 ZONE	W46	P	3381	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 69 2538 ZONE	W47	P	3382	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 72 2538 ZONE	W48	P	3383	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 92 2538 ZONE	W49	P	3369	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 94 2538 ZONE	W50	P	3368	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 101 2538 ZONE	W51	P	3360	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 103 2538 ZONE	W52	P	3411	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 117 2538 ZONE	W53	P	3417	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 90 2635 ZONE	W54	P	3711	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 98 2635 ZONE	W59	P	3410	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 110 2635 ZONE.	W60	P	3413	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 112 2635 ZONE	W61	P	3414	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 114 2635 ZONE	W62	P	3416	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 115 2635 ZONE	W63	P	3371	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 78 2635 ZONE	W64	P	3407	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 91 2745 ZONE	W65	P	3361	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 95 FT APACHE 2538 ZONE	W66	P	3362	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 96 GRAND CANYON 2635 ZONE	W67	P	3848	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 113 2635 ZONE	W68	P	3415	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 74 2538 ZONE	W69	P	3859	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 75 2538 ZONE	W70	P	4043	GW
CLARK	C	NV0000090	LAS VEGAS VALLEY WATER DISTRICT	WL	WELL 29A GOWAN 2420 ZONE	W73	P	9108	GW
NYE	NC	NV0005068	LDS CHURCH PAHRUMP WARD	WL	WELL	W01	P	801	GW
CLARK	NC	NV0002508	LEE CANYON CAMPGROUNDS USFS	CC	CC FROM CAMP LEE CANYON NV0005009	CC02	P	6391	GW
CLARK	NC	NV0002508	LEE CANYON CAMPGROUNDS USFS	WL	US FOREST SERVICE WELL	W01	P	7915	GW
CLARK	NTNC	NV0000307	LEE CANYON SKI AREA	WL	WELL 1	W01	P	487	GW
CLARK	NTNC	NV0000307	LEE CANYON SKI AREA	WL	WELL 2	W02	P	940	GW
WHITE PINE	NC	NV0003051	LEHMAN CREEK CAMPGROUND NPS	SP	SPRING	SP01	P	3191	GW
LINCOLN	NC	NV0002192	LITTLE A LE INN	WL	WELL 1	W01	P	623	GW
DOUGLAS	C	NV0002050	LOGAN CREEK ESTATES GID	WL	NEW WELL 3	W03	P	3834	GW
NYE	NTNC	NV0000871	LONGSTREET INN AND CASINO	WL	WELL 2	W02	P	3070	GW
PERSHING	C	NV0000861	LOVELOCK CORRECTIONAL CENTER NDOC	CC	CC FROM LOVELOCK MEADOWS WATER NV0000161	CC01	P	2276	GW
PERSHING	C	NV0000161	LOVELOCK MEADOWS WATER DISTRICT	WL	WELL 7	W01	P	438	GW
PERSHING	C	NV0000161	LOVELOCK MEADOWS WATER DISTRICT	WL	WELL 5	W02	P	1053	GW
PERSHING	C	NV0000161	LOVELOCK MEADOWS WATER DISTRICT	WL	WELL 8 OREANA WELL	W03	P	5779	GW
CLARK	C	NV0000140	LUNA VISTA	WL	LAKEWAY RNT WELL	W01	P	30	GW
HUMBOLDT	NC	NV0002220	LYE CREEK CAMPGROUND USFS	SP	SPRING	SP01	P	3480	GW
NYE	C	NV0000165	MANHATTAN TOWN WATER	WL	PIPE SPRINGS WELL	W03	P	6248	GW
HUMBOLDT	C	NV0000162	MCDERMITT WATER SYSTEM	WL	WELL 1	W01	E	439	GW
HUMBOLDT	C	NV0000162	MCDERMITT WATER SYSTEM	WL	WELL 3	W03	P	7706	GW

Appendix D - Table of Active Sources for PWS in Nevada

WHITE PINE	C	NV0000163	MCGILL WATER AND SEWER DIST	WL	WELL 1	W01	P	923	GW
WHITE PINE	C	NV0000163	MCGILL WATER AND SEWER DIST	WL	WELL 2	W02	P	5596	GW
ELKO	NC	NV0003071	MIDAS WATER COOPERATIVE	WL	WELL 1 MIDAS TOWN WELL	W01	P	982	GW
ELKO	NC	NV0003071	MIDAS WATER COOPERATIVE	WL	WELL 2	W02	P	5356	GW
CHURCHILL	NC	NV0002027	MIDDLEGATE STATION	WL	WELL 1	W01	P	546	GW
CARSON CITY	NC	NV0002006	MILL HOUSE INN MOTEL	WL	WELL 1	W01	P	537	GW
MINERAL	C	NV0000074	MINA LUNING WATER SYSTEM	WL	RASSLER UPPER WELL	W01	P	1137	GW
MINERAL	C	NV0000074	MINA LUNING WATER SYSTEM	WL	RASSLER LOWER WELL	W02	P	1109	GW
MINERAL	C	NV0000074	MINA LUNING WATER SYSTEM	WL	MINA NEW WELL 4	W04	P	3306	GW
ELKO	C	NV0000169	MONTELLO WATER SYSTEM	SP	MONTELLO NORTH SPRINGS	SP01	P	2545	GW
ELKO	C	NV0000169	MONTELLO WATER SYSTEM	SP	LOWER SPRINGS	SP02	P	4767	GW
ELKO	C	NV0000169	MONTELLO WATER SYSTEM	SP	MONTELLO WEST SPRINGS	SP03	E	8904	GW
NYE	NC	NV0002142	MOOSE LODGE 808	WL	WELL	W01	P	600	GW
CLARK	NC	NV0001082	MOUNTAIN SPRINGS BAR INC	WL	WELL 1	W01	P	162	GW
DOUGLAS	C	NV0003081	MOUNTAIN VIEW MHP	WL	WELL	W01	P	732	GW
CLARK	C	NV0001015	MT CHARLESTON WATER COMPANY	WL	WELL 1	W01	P	142	GW
EUREKA	NC	NV0002214	NDOT BEOWAWE ROADSIDE PARK RP802 EU	WL	WELL 1	W01	P	632	GW
NYE	NC	NV0002147	NDOT BIG SMOKEY ROADSIDE PARK RP807NY	WL	WELL	W01	P	164	GW
PERSHING	NC	NV0002160	NDOT COSGRAVE ROADSIDE PARK RP804PE	WL	WELL 2 REPLACEMENT WELL	W02	P	7320	GW
NYE	NC	NV0002146	NDOT LATHROP WELLS ROADSIDE PARK RP801NY	WL	WELL	W01	P	603	GW
ESMERALDA	NC	NV0002089	NDOT MILLERS NV ROADSIDE PARK RP801ES	WL	EAST WELL 1	W01	P	577	GW
CHURCHILL	NC	NV0002034	NDOT TRINITY ROADSIDE PARK RP806CH	CC	NP TRUCK FROM LOVELOCK MEADOWS	CC01	P	2602	GW
HUMBOLDT	NC	NV0002102	NDOT VALMY ROADSIDE PARK RP809HU	WL	WELL	W01	P	583	GW
CLARK	C	NV0003028	NELLIS AIR FORCE BASE	CC	CC FROM AMS TP AT SNWS NV0000289	CC01	P	2539	SW
CLARK	C	NV0003028	NELLIS AIR FORCE BASE	CC	CC FROM RM TP AT SNWS NV0000289	CC02	P	5547	SW
CLARK	C	NV0003028	NELLIS AIR FORCE BASE	WL	AREA 1 WELL 1 NON-POTABLE EMERGENCY	W01	E	3455	GW
CLARK	C	NV0003028	NELLIS AIR FORCE BASE	WL	AREA 1 WELL 7 489 NON-POTABLE EMERGENCY	W02	E	3312	GW
CLARK	C	NV0003028	NELLIS AIR FORCE BASE	WL	AREA 1 WELL 14 NON-POTABLE EMERGENCY	W03	E	3456	GW
CLARK	C	NV0003028	NELLIS AIR FORCE BASE	WL	AREA 1 WELL 11 NON-POTABLE EMERGENCY	W05	E	3459	GW
CLARK	C	NV0003028	NELLIS AIR FORCE BASE	WL	AREA 1 WELL 12 NON-POTABLE EMERGENCY	W06	E	3460	GW
CLARK	C	NV0003028	NELLIS AIR FORCE BASE	WL	CRAIG RD WELL 2	W09	P	3462	GW
CLARK	C	NV0003028	NELLIS AIR FORCE BASE	WL	CRAIG RD WELL 8	W10	P	3314	GW
EUREKA	NTNC	NV0005077	NEVADA GOLD MINES CARLIN GOLDSTRIKE FCTY	WL	EW-14	W07	P	8560	GW
EUREKA	NTNC	NV0002603	NEVADA GOLD MINES CARLIN NORTH AREA LCH	WL	GEN 3	W02	P	8877	GW
EUREKA	NTNC	NV0002600	NEVADA GOLD MINES GOLD QUARRY	WL	WELL PW4	W01	P	3223	GW
EUREKA	NTNC	NV0002600	NEVADA GOLD MINES GOLD QUARRY	WL	WELL PW2	W02	P	3224	GW
EUREKA	NTNC	NV0002600	NEVADA GOLD MINES GOLD QUARRY	WL	WELL PW12	W03	P	6685	GW
HUMBOLDT	NTNC	NV0002528	NEVADA GOLD MINES TURQUOISE RIDGE	WL	WELL 4	W03	P	6497	GW
HUMBOLDT	NTNC	NV0005070	NEVADA GOLD MINES TWIN CREEKS	WL	WELL 1	W01	P	1874	GW
CLARK	NTNC	NV0005006	NEVADA TEST AND TRAINING RANGE PT BRAVO	WL	POINT BRAVO WELL	W01	P	771	GW
ELKO	C	NV0000037	NEVADA YOUTH TRAINING CENTER	WL	WELL 1	W01	P	409	GW
NYE	NC	NV0004099	NNSS AREA 12	WL	WELL 8	W01	P	766	GW
NYE	NC	NV0004098	NNSS AREA 25	WL	WELL J12	W01	P	993	GW
NYE	NC	NV0004098	NNSS AREA 25	WL	WELL J14	W03	P	6463	GW
NYE	NTNC	NV0000360	NNSS SITE AREA A23 AND 6	WL	WELL 5 B	W02	P	2392	GW
NYE	NTNC	NV0000360	NNSS SITE AREA A23 AND 6	WL	WELL 4	W04	P	1123	GW
NYE	NTNC	NV0000360	NNSS SITE AREA A23 AND 6	WL	WELL 4 A	W05	P	2394	GW

Appendix D - Table of Active Sources for PWS in Nevada

CLARK	C	NV0000175	NORTH LAS VEGAS UTILITIES	CC	CC FROM AMS TP AT SNWS NV0000289	CC01	P	41	SW
CLARK	C	NV0000175	NORTH LAS VEGAS UTILITIES	CC	CC FROM RM TP AT SNWS NV0000289	CC02	E	5545	SW
CLARK	C	NV0000175	NORTH LAS VEGAS UTILITIES	WL	WELL ROBINSON	W01	P	273	GW
CLARK	C	NV0000175	NORTH LAS VEGAS UTILITIES	WL	ELSTNER ESTATES REPLACEMENT WELL W03	W08	P	9576	GW
CLARK	C	NV0000175	NORTH LAS VEGAS UTILITIES	WL	SILVER MESA REPLACEMENT WELL W05	W10	P	9578	GW
HUMBOLDT	NTNC	NV0002513	NORTH VALMY POWER PLANT	WL	WELL GP 3B	W03	P	2367	GW
HUMBOLDT	NTNC	NV0002513	NORTH VALMY POWER PLANT	WL	WELL 4 GP 2R	W04	P	8978	GW
CLARK	C	NV0002506	NPS CALLVILLE BAY	IN	CALLVILLE BAY LK MEAD INTAKE	IN01	P	2296	SW
CLARK	NTNC	NV0002501	NPS COTTONWOOD COVE	WL	WELL 1	W01	P	166	GW
CLARK	NTNC	NV0002501	NPS COTTONWOOD COVE	WL	WELL 3	W02	P	2359	GW
CLARK	C	NV0002503	NPS ECHO BAY	IN	ECHO BAY LAKE MEAD INTAKE	IN01	P	168	SW
NYE	NTNC	NV0002152	NYE COUNTY COMPLEX	WL	WELL 1	W01	P	605	GW
NYE	NTNC	NV0002152	NYE COUNTY COMPLEX	WL	WELL 2	W02	P	5208	GW
ELKO	C	NV0004017	OASIS MHP	WL	WELL	W01	P	738	GW
CHURCHILL	C	NV0000052	OK MOBILE HOME PARK	WL	WELL 1	W01	P	419	GW
CHURCHILL	C	NV0000303	OLD RIVER WATER COMPANY	WL	WELL 2 SECONDARY	W02	P	1813	GW
CHURCHILL	C	NV0000303	OLD RIVER WATER COMPANY	WL	WELL 3 PRIMARY	W03	P	4714	GW
WASHOE	C	NV0002526	OLD WASHOE ESTATES	WL	OLD WASHOE WELL 3	W03	P	2416	GW
WASHOE	C	NV0002526	OLD WASHOE ESTATES	WL	OLD WASHOE WELL 4	W04	P	7194	GW
HUMBOLDT	C	NV0003032	OROVADA WATER DISTRICT	WL	WELL 1	W01	P	701	GW
HUMBOLDT	C	NV0003032	OROVADA WATER DISTRICT	WL	WELL 2	W02	P	979	GW
NYE	NC	NV0005034	PAHRUMP RV PARK	WL	WELL	W01	P	787	GW
NYE	NTNC	NV0003036	PAHRUMP SENIOR CENTER INC	WL	GEEZER WELL	W01	P	702	GW
CLARK	C	NV0000819	PALM GARDENS WATER CO OP	WL	WELL 1	W01	P	135	GW
CLARK	C	NV0000819	PALM GARDENS WATER CO OP	WL	WELL 2	W02	P	303	GW
LINCOLN	C	NV0000185	PANACA FARMSTEAD ASSOCIATION	WL	WELL 3 - EMERGENCY ONLY	W03	E	447	GW
LINCOLN	C	NV0000185	PANACA FARMSTEAD ASSOCIATION	WL	WELL 5 NEW WELL	W05	P	6659	GW
HUMBOLDT	NTNC	NV0002186	PARADISE VALLEY ELEMENTARY	WL	WELL 1	W01	P	620	GW
HUMBOLDT	NC	NV0002106	PARADISE VALLEY PARK	WL	WELL 1	W01	P	585	GW
DOUGLAS	C	NV0000067	PINION PINES MHP	WL	WELL 1	W01	P	429	GW
LINCOLN	C	NV0000186	PIOCHE PUBLIC UTILITIES	WL	BLANCHARD WELL 3	W01	P	448	GW
LINCOLN	C	NV0000186	PIOCHE PUBLIC UTILITIES	WL	MURRAY WELL 1	W03	P	1057	GW
LINCOLN	C	NV0000186	PIOCHE PUBLIC UTILITIES	WL	CLAY WELL 2	W04	E	3286	GW
LINCOLN	C	NV0000187	PIOCHE PUBLIC UTILITIES CASTLETON	SP	FLORAL SPRING	SP01	P	2339	GW
CLARK	NC	NV0001012	PIONEER SALOON	WL	PIONEER SALOON WELL	W01	P	141	GW
CLARK	C	NV0001073	PRIMM VALLEY CASINO RESORTS	WL	WELL 6	W02	P	343	GW
CLARK	C	NV0001073	PRIMM VALLEY CASINO RESORTS	WL	WELL 5A	W05	P	7184	GW
ELKO	NC	NV0002064	R&R RV PARK	WL	WELL 2	W02	P	7143	GW
CLARK	NC	NV0001069	RANCH HOUSE	WL	SP CL WELL	W01	P	153	GW
CLARK	C	NV0000129	RANCHO COOPERATIVE WATER COMPANY	WL	SOUTH WELL	W01	P	24	GW
CLARK	C	NV0000129	RANCHO COOPERATIVE WATER COMPANY	WL	NORTH WELL	W02	P	265	GW
CLARK	NTNC	NV0005011	RED ROCK CANYON VISITOR CENTER BLM	WL	WATER WELL 2	W02	P	8046	GW
CLARK	NTNC	NV0001042	RIVERSIDE RESORT	WL	WELL 4 EAST	W04	P	4178	GW
CLARK	NTNC	NV0001042	RIVERSIDE RESORT	WL	WELL 5 WEST OLD WELL 1	W05	P	4505	GW
CLARK	NTNC	NV0001042	RIVERSIDE RESORT	WL	WELL 6 SOUTH	W06	P	5495	GW
DOUGLAS	C	NV0000068	RIVERVIEW MOBILE HOME PARK	WL	UPPER WELL 2	W02	P	430	GW
CLARK	C	NV0000319	ROARK ESTATES WATER ASSOC	WL	WELL 2	W02	P	6658	GW

Appendix D - Table of Active Sources for PWS in Nevada

DOUGLAS	C	NV0000260	ROUND HILL GID	IN	NEW INTAKE	IN02	P	5827	SW
NYE	NTNC	NV0002535	ROUND MOUNTAIN SMOKY VALLEY MINE	WL	WEST WELL	W01	P	651	GW
NYE	NTNC	NV0002535	ROUND MOUNTAIN SMOKY VALLEY MINE	WL	EAST WELL 1 (MIDDLE WELL)	W02	P	2398	GW
NYE	NTNC	NV0002535	ROUND MOUNTAIN SMOKY VALLEY MINE	WL	EAST WELL 2	W03	P	2399	GW
HUMBOLDT	NC	NV0002542	ROYAL PEACOCK RV PARK	WL	WELL 1	W01	P	656	GW
ELKO	C	NV0002075	RUBY DOME HOLDINGS DBA ELKO RV PARK	WL	WELL 1	W01	P	569	GW
ELKO	C	NV0002075	RUBY DOME HOLDINGS DBA ELKO RV PARK	WL	WELL 2	W02	P	2366	GW
ELKO	NC	NV0003070	RUBY VALLEY LDS CHAPEL	WL	WELL	W01	P	725	GW
WHITE PINE	C	NV0000164	RUTH WATER DISTRICT	CC	CC INTERTIE FROM CITY OF ELY	CC01	P	6022	GW
PERSHING	NC	NV0002223	RYE PATCH STATE PARK	WL	WELL 1	W01	P	638	GW
PERSHING	NC	NV0005023	RYE PATCH STATE PARK PICNIC AREA	WL	WELL 1	W01	P	780	GW
DOUGLAS	NC	NV0002047	S AND J VENTURES DBA JUNCTION BAR	WL	WELL 1	W01	P	557	GW
ELKO	NTNC	NV0002566	SAGE SCHOOLS	WL	WELL 1 SAGE SCHOOL	W01	P	673	GW
ELKO	NTNC	NV0002566	SAGE SCHOOLS	WL	WELL 2	W02	P	971	GW
CHURCHILL	C	NV0002023	SAGE VALLEY MHP LLC	WL	WELL 1	W01	P	544	GW
CHURCHILL	C	NV0000054	SAGE VALLEY PARK ANNEX	WL	SOUTH WELL OLD WELL 2	W02	P	913	GW
CHURCHILL	C	NV0000054	SAGE VALLEY PARK ANNEX	WL	NORTH WELL OLD WELL 1 BACKUP	W03	P	3741	GW
CHURCHILL	NC	NV0000872	SAGE VALLEY RV PARK LLC	WL	WELL 3	W03	P	8861	GW
CLARK	NC	NV0000317	SANDY VALLEY LIBRARY COMM CTR	WL	WELL 04	W03	P	9473	GW
CLARK	NTNC	NV0004001	SANDY VALLEY SCHOOLS ES MS CCSD	WL	WELL 1	W01	P	191	GW
CLARK	C	NV0000219	SEARCHLIGHT WATER COMPANY	WL	WELL S2	W01	P	68	GW
CLARK	C	NV0000219	SEARCHLIGHT WATER COMPANY	WL	WELL S1	W02	P	2357	GW
CLARK	C	NV0000219	SEARCHLIGHT WATER COMPANY	WL	WELL S3	W03	P	2358	GW
DOUGLAS	NC	NV0000844	SEVEN ELEVEN NO 23074	WL	WELL 1	W01	P	1881	GW
CLARK	C	NV0000126	SHETLAND WATER DISTRICT	WL	WELL 1B	W02	P	9501	GW
NYE	NC	NV0003074	SHORT BRANCH	WL	WELL 1	W01	P	728	GW
NYE	C	NV0005028	SHOSHONE ESTATES WATER CO INC	WL	WELL 1	W01	P	783	GW
NYE	C	NV0005028	SHOSHONE ESTATES WATER CO INC	WL	WELL 2	W02	P	3304	GW
DOUGLAS	C	NV0000030	SIERRA ESTATES GID	CC	NORTH COUNTY INTERTIE	CC01	E	6149	GW
DOUGLAS	C	NV0000030	SIERRA ESTATES GID	WL	WELL 1 BUCKS WAY	W01	P	404	GW
WASHOE	C	NV0004021	SILVER KNOLLS MUTUAL WATER COMPANY	WL	WELL 2 LOWER WELL	W01	P	192	GW
ESMERALDA	C	NV0000363	SILVER PEAK WATER SYSTEM	WL	WELL 2	W01	P	508	GW
ESMERALDA	C	NV0000363	SILVER PEAK WATER SYSTEM	WL	WELL 4	W03	P	6043	GW
LYON	C	NV0000223	SILVER SPRINGS MUTUAL WATER COMPANY	WL	WELL 5 LAKE STREET	W02	P	928	GW
LYON	C	NV0000223	SILVER SPRINGS MUTUAL WATER COMPANY	WL	WELL 4 IDAHO ST	W03	P	1058	GW
LYON	C	NV0000223	SILVER SPRINGS MUTUAL WATER COMPANY	WL	DEODAR REPLACEMENT WELL	W05	P	8862	GW
WASHOE	NC	NV0000761	SLIDE MOUNTAIN SKI AREA	SP	BRUCES SPRING	SP01	P	3524	GW
WASHOE	NC	NV0000761	SLIDE MOUNTAIN SKI AREA	SP	WINTERS SPRING	SP02	P	3750	GW
WASHOE	NC	NV0000761	SLIDE MOUNTAIN SKI AREA	SP	ZEPHER SPRING	SP03	P	3751	GW
LYON	NTNC	NV0002189	SMITH VALLEY SCHOOLS	WL	WELL	W01	P	621	GW
ELKO	C	NV0000351	SOUTH CRESTVIEW HOMEOWNERS	WL	WELL 1	W01	P	499	GW
ELKO	C	NV0000351	SOUTH CRESTVIEW HOMEOWNERS	WL	WELL 2	W02	P	2114	GW
ELKO	C	NV0000351	SOUTH CRESTVIEW HOMEOWNERS	WL	WELL 3	W03	P	5349	GW
ELKO	NC	NV0002209	SOUTH RUBY CAMPGROUND USFS	WL	WELL	W01	P	630	GW
CLARK	C	NV0005062	SOUTHERN DESERT CORRECTIONAL CTR NDOC	WL	WELL 2	W02	P	1002	GW
CLARK	C	NV0005062	SOUTHERN DESERT CORRECTIONAL CTR NDOC	WL	WELL 3	W03	P	3082	GW
CLARK	C	NV0005062	SOUTHERN DESERT CORRECTIONAL CTR NDOC	WL	WELL 4	W04	P	3083	GW

Appendix D - Table of Active Sources for PWS in Nevada

CLARK	C	NV0005062	SOUTHERN DESERT CORRECTIONAL CTR NDOC	WL	WELL 5	W05	P	6230	GW
CLARK	C	NV0005062	SOUTHERN DESERT CORRECTIONAL CTR NDOC	WL	WELL 6	W06	P	8624	GW
CLARK	C	NV0000221	SPIRIT MOUNTAIN UTILITY COMPANY	WL	WELL 1 COMMANCHE WAY	W01	P	69	GW
CLARK	C	NV0000221	SPIRIT MOUNTAIN UTILITY COMPANY	WL	WELL 2 EAST	W02	E	286	GW
DOUGLAS	NC	NV0004041	SPOONER LAKE STATE PARK	WL	WELL 1	W01	P	746	GW
CLARK	NC	NV0001071	SPRING MOUNTAIN RANCH STATE PARK	WL	WELL SPRING MTN RANCH CY1988	W01	P	155	GW
CLARK	C	NV0000292	SPRING MOUNTAIN YOUTH CAMP	SP	DEER CREEK SPRING	SP01	P	3517	GW
LINCOLN	NC	NV0002124	SPRING VALLEY STATE PARK	WL	WELL 3	W01	P	591	GW
LYON	C	NV0000224	STAGECOACH GID	WL	CENTRAL WELL CW	W01	P	455	GW
LYON	C	NV0000224	STAGECOACH GID	WL	CHURCHILL DOWNS WELL 5	W05	P	1138	GW
LYON	C	NV0000224	STAGECOACH GID	WL	SOUTH WELL	W06	E	8558	GW
LYON	C	NV0000224	STAGECOACH GID	WL	NORTH WELL	W07	E	8559	GW
NYE	NC	NV0002598	STAGESTOP RESTAURANT	WL	WELL 1	W01	P	688	GW
HUMBOLDT	C	NV0000252	STAR CITY PROPERTIES	WL	WELL 1	W01	P	468	GW
HUMBOLDT	C	NV0000252	STAR CITY PROPERTIES	WL	WELL 2	W02	P	2148	GW
PERSHING	NC	NV0002517	STAR POINT MHP RV PARK	WL	WELL 1	W01	P	644	GW
STOREY	C	NV0000240	STOREY COUNTY WATER DISTRICT	IN	VCWTP RAW WATER LINE	IN01	P	461	SW
NYE	C	NV0005066	SUNSET MHP	WL	WELL 1	W01	P	799	GW
NYE	C	NV0005066	SUNSET MHP	WL	WELL 2	W02	P	5441	GW
CARSON CITY	C	NV0000028	TERRACE GARDEN APARTMENTS	WL	WELL 1	W01	P	402	GW
ELKO	NC	NV0003044	TERRACES PICNIC GROUNDS USFS	SP	TERRACE SPRING	SP01	P	3196	GW
ELKO	NC	NV0003043	THOMAS CANYON CAMPGROUND USFS	SP	THOMAS CREEK SPRING	SP01	P	3527	GW
NYE	NTNC	NV0000804	TOLICHA PEAK ELECTRONIC COMBAT RANGE	WL	WELL 1	W01	P	511	GW
NYE	NTNC	NV0000823	TONOPAH CONSERVATION CAMP NDOC	CC	CC FROM TONOPAH PUBLIC UTIL NV0000237	CC01	P	136	GW
NYE	NTNC	NV0005002	TONOPAH ELECTRONIC COMBAT RANGE O AND M	WL	O AND M WELL	W01	P	769	GW
NYE	C	NV0000237	TONOPAH PUBLIC UTILITIES	WL	WELL 5	W05	P	459	GW
NYE	C	NV0000237	TONOPAH PUBLIC UTILITIES	WL	WELL 6	W06	P	930	GW
NYE	C	NV0000237	TONOPAH PUBLIC UTILITIES	WL	WELL 7	W07	P	1060	GW
NYE	C	NV0000237	TONOPAH PUBLIC UTILITIES	WL	WELL 8	W08	P	2391	GW
NYE	C	NV0000237	TONOPAH PUBLIC UTILITIES	WL	WELL 9	W09	P	6930	GW
NYE	C	NV0000237	TONOPAH PUBLIC UTILITIES	WL	WELL 10	W10	P	6931	GW
NYE	NTNC	NV0005001	TONOPAH TEST RANGE AREA 10 INDUSTRIAL	WL	WELL EH 2	W01	P	768	GW
NYE	NTNC	NV0004068	TONOPAH TEST RANGE MANCAMP	WL	NORTH CACTUS FLATS WELL	W01	P	1081	GW
NYE	NTNC	NV0004068	TONOPAH TEST RANGE MANCAMP	WL	WELL 3 REPLACEMENT FOR WELL 2	W03	P	8091	GW
NYE	NTNC	NV0003014	TONOPAH TEST RANGE SITE 6	WL	WELL 6	W01	P	1077	GW
DOUGLAS	C	NV0000070	TOPAZ LAKE WATER CO INC	WL	WELL 1	W01	P	432	GW
DOUGLAS	C	NV0000070	TOPAZ LAKE WATER CO INC	WL	WELL 2 - EMERGENCY USE ONLY	W02	E	2103	GW
DOUGLAS	C	NV0000070	TOPAZ LAKE WATER CO INC	WL	WELL 3	W03	P	2104	GW
DOUGLAS	C	NV0000070	TOPAZ LAKE WATER CO INC	WL	WELL 4 PINE NUT WELL	W04	P	6262	GW
DOUGLAS	C	NV0000239	TOPAZ RANCH ESTATES GID AND WATER CO	WL	WELL 1	W01	P	460	GW
DOUGLAS	C	NV0000239	TOPAZ RANCH ESTATES GID AND WATER CO	WL	W07 WELL 6 REPLACEMENT	W07	P	10222	GW
CLARK	C	NV0000314	TORREY PINES TINA LN WTR ASSOC	WL	WATER WELL 1996 REPLACEMENT	W01	P	489	GW
DOUGLAS	C	NV0000168	TOWN OF MINDEN	CC	CC INTERTIE FROM GARDERVILLE TOWN NV0065	CC01	E	6671	GW
DOUGLAS	C	NV0000168	TOWN OF MINDEN	CC	CC INTERTIE FROM EAST VALLEY NV2216	CC02	E	6903	GW
DOUGLAS	C	NV0000168	TOWN OF MINDEN	WL	WELL 5 1715 BOUGAINVILLEA	W02	P	925	GW
DOUGLAS	C	NV0000168	TOWN OF MINDEN	WL	WELL 3 1648 COUNTY RD	W03	P	1055	GW
DOUGLAS	C	NV0000168	TOWN OF MINDEN	WL	WELL 4 1769 IRONWOOD DR	W04	P	1111	GW

Appendix D - Table of Active Sources for PWS in Nevada

DOUGLAS	C	NV0000168	TOWN OF MINDEN	WL	WELL 8 1745 BUCKEYE ROAD	W05	P	4583	GW
DOUGLAS	C	NV0000168	TOWN OF MINDEN	WL	WELL 2 1573 COUNTY RD	W06	P	3824	GW
DOUGLAS	C	NV0000168	TOWN OF MINDEN	WL	WELL 7 1730 U ORBIT WAY	W07	P	7319	GW
DOUGLAS	C	NV0000168	TOWN OF MINDEN	WL	WELL 9 1330 BUCKEYE RD	W09	P	7558	GW
DOUGLAS	C	NV0000168	TOWN OF MINDEN	WL	WELL 11 1800 HEYBOURNE RD	W10	P	8030	GW
DOUGLAS	C	NV0000168	TOWN OF MINDEN	WL	1613 WATER ST WELL 1 REPLACEMENT	W11	P	8504	GW
PERSHING	NTNC	NV0002159	TRAVEL CENTERS OF AMERICA NO 181	WL	WELL 1	W01	E	608	GW
PERSHING	NTNC	NV0002159	TRAVEL CENTERS OF AMERICA NO 181	WL	WELL 3	W03	P	8802	GW
CLARK	NTNC	NV0001059	TROPICANA RESORT AND CASINO	CC	LVVWD INTERTIE N IS 2	CC01	P	6100	SW
CLARK	NTNC	NV0001059	TROPICANA RESORT AND CASINO	WL	TROP WELL 3 EAST	W01	P	151	GW
CLARK	NTNC	NV0001059	TROPICANA RESORT AND CASINO	WL	TROP WELL 4 SOUTH	W02	P	306	GW
PERSHING	NC	NV0003072	TURN 3 RV PARK AND STORE	WL	WELL 1	W01	P	727	GW
ELKO	C	NV0000189	TUSCARORA WATER SYSTEM	WL	WELL	W01	P	451	GW
DOUGLAS	C	NV0000817	UPPAWAY WATER SYSTEM	WL	WELL 1 WEST	W01	P	2596	GW
DOUGLAS	C	NV0000817	UPPAWAY WATER SYSTEM	WL	WELL 2 EAST BACK UP	W02	P	3351	GW
CLARK	NC	NV0001075	VALLEY OF FIRE STATE PARK	WL	VALLEY OF FIRE WELL	W01	P	158	GW
ELKO	C	NV0000805	VALLEY VIEW RV PARK	WL	WELL 1 WEST FIRE	W01	P	512	GW
ELKO	C	NV0000805	VALLEY VIEW RV PARK	WL	WELL 2 EAST	W02	E	3581	GW
HUMBOLDT	NC	NV0002112	VALMY CHEVRON	WL	WELL 2	W02	P	3936	GW
HUMBOLDT	C	NV0003016	VALMY STATION MOBILE HOME PARK	WL	WELL 1	W01	P	699	GW
NYE	NC	NV0002556	VFW PAHRUMP POST 10054	WL	WELL 2	W02	P	8980	GW
HUMBOLDT	NC	NV0002543	VIRGIN VALLEY CAMPGROUND FWS	WL	WELL	W01	P	657	GW
CLARK	C	NV0000167	VIRGIN VALLEY WATER DISTRICT	WL	WELL 02 BUNKERVILLE	W08	P	373	GW
CLARK	C	NV0000167	VIRGIN VALLEY WATER DISTRICT	WL	WELL 28	W09	P	3333	GW
CLARK	C	NV0000167	VIRGIN VALLEY WATER DISTRICT	WL	WELL 29	W10	E	3332	GW
CLARK	C	NV0000167	VIRGIN VALLEY WATER DISTRICT	WL	WELL 30	W11	E	3266	GW
CLARK	C	NV0000167	VIRGIN VALLEY WATER DISTRICT	WL	WELL 32	W12	P	4572	GW
CLARK	C	NV0000167	VIRGIN VALLEY WATER DISTRICT	WL	WELL 31	W13	P	4570	GW
CLARK	C	NV0000167	VIRGIN VALLEY WATER DISTRICT	WL	WELL 33	W14	P	4571	GW
CLARK	C	NV0000167	VIRGIN VALLEY WATER DISTRICT	WL	WELL 27A	W15	P	8804	GW
CLARK	C	NV0000167	VIRGIN VALLEY WATER DISTRICT	WL	WELL 01A BUNKERVILLE	W16	P	8854	GW
CLARK	C	NV0000167	VIRGIN VALLEY WATER DISTRICT	WL	WELL 26A	W17	P	9393	GW
MINERAL	C	NV0000268	WALKER LAKE GID	WL	MALLET WELL	W01	E	478	GW
MINERAL	C	NV0000268	WALKER LAKE GID	WL	BIGHORN WELL	W03	P	6919	GW
LYON	NC	NV0004048	WALKER RIVER RESORT	WL	WELL 1	W01	P	751	GW
LYON	NC	NV0004048	WALKER RIVER RESORT	WL	WELL 2	W02	P	8893	GW
LYON	C	NV0000242	WEED HEIGHTS DEVELOPMENT	WL	WELL 1 TAC WELL	W01	P	462	GW
LYON	C	NV0000242	WEED HEIGHTS DEVELOPMENT	WL	WELL 2 BLM WELL	W02	P	5502	GW
ELKO	NC	NV0002078	WELCOME STATION RV PARK SONO LLC	WL	WELL	W01	P	571	GW
LYON	NC	NV0000243	WELLINGTON STATION RESORT	WL	WELL 1	W01	P	463	GW
ELKO	NTNC	NV0005005	WELLS HONOR CAMP NDOC	WL	WELL 1	W01	P	770	GW
ELKO	C	NV0000245	WELLS MUNICIPAL WATER DEPARTMENT	WL	WELL 2 PRIMARY	W02	P	932	GW
ELKO	C	NV0000245	WELLS MUNICIPAL WATER DEPARTMENT	WL	HEAVY INDUSTRIAL PARK WELL	W05	P	4615	GW
ELKO	C	NV0000245	WELLS MUNICIPAL WATER DEPARTMENT	WL	WELL 6 RODEO	W06	P	5594	GW
ELKO	C	NV0000245	WELLS MUNICIPAL WATER DEPARTMENT	WL	CHARIOT WELL NEW WELL	W07	P	6175	GW
ELKO	C	NV0000246	WEST WENDOVER WATER SYSTEM	WL	SHAFTER 1 WELL	W01	P	465	GW
ELKO	C	NV0000246	WEST WENDOVER WATER SYSTEM	WL	SHAFTER 2 WELL	W02	P	933	GW

Appendix D - Table of Active Sources for PWS in Nevada

ELKO	C	NV0000246	WEST WENDOVER WATER SYSTEM	WL	SHAFTER 3 WELL	W03	P	1116	GW
ELKO	C	NV0000246	WEST WENDOVER WATER SYSTEM	WL	SHAFTER 4 WELL	W04	P	2130	GW
ELKO	C	NV0000246	WEST WENDOVER WATER SYSTEM	WL	SHAFTER 5 WELL	W05	P	2133	GW
ELKO	C	NV0000246	WEST WENDOVER WATER SYSTEM	WL	WELL PEQUOP 1	W07	P	7707	GW
ELKO	C	NV0000246	WEST WENDOVER WATER SYSTEM	WL	WELL PEQUOP 2	W08	P	7708	GW
ELKO	C	NV0000246	WEST WENDOVER WATER SYSTEM	WL	SHAFTER 6 WELL REPLACEMENT	W09	P	8064	GW
WHITE PINE	NC	NV0003054	WHEELER PEAK CAMPGROUND NPS	SP	SPRING	SP01	P	3167	GW
ELKO	NC	NV0003059	WILDHORSE STATE PARK AND RECREATION AREA	WL	WELL 1	W01	P	719	GW
DOUGLAS	NTNC	NV0000816	WILLIAMS RIDGE TECH PARK	CC	CC INTERTIE FROM FAIRGROUNDS NV2540	CC01	P	9491	GW
DOUGLAS	NTNC	NV0000816	WILLIAMS RIDGE TECH PARK	WL	WELL 1	W01	P	520	GW
HUMBOLDT	C	NV0000248	WINNEMUCCA CITY OF	SP	RAILROAD SPRING	SP05	P	2326	GW
HUMBOLDT	C	NV0000248	WINNEMUCCA CITY OF	WL	WELL 5	W03	P	1847	GW
HUMBOLDT	C	NV0000248	WINNEMUCCA CITY OF	WL	WELL 6	W04	P	1117	GW
HUMBOLDT	C	NV0000248	WINNEMUCCA CITY OF	WL	WELL 7	W06	P	2965	GW
HUMBOLDT	C	NV0000248	WINNEMUCCA CITY OF	WL	WELL 4	W07	P	4018	GW
HUMBOLDT	C	NV0000248	WINNEMUCCA CITY OF	WL	WELL 2A	W08	P	5846	GW
LYON	C	NV0000255	YERINGTON CITY OF	WL	WELL 3 BROADWAY	W02	P	935	GW
LYON	C	NV0000255	YERINGTON CITY OF	WL	MASON ROAD WELL	W05	P	4008	GW
LYON	C	NV0000255	YERINGTON CITY OF	WL	WELL 6 MOUNTAIN VIEW (REPLACEMENT)	W06	P	5707	GW
DOUGLAS	NTNC	NV0002059	ZEPHYR COVE LODGE AND RESORT	WL	WELL 1	W01	P	2380	GW
DOUGLAS	NTNC	NV0002059	ZEPHYR COVE LODGE AND RESORT	WL	WELL 2	W02	P	2381	GW
DOUGLAS	NTNC	NV0002059	ZEPHYR COVE LODGE AND RESORT	WL	WELL 3	W03	P	2382	GW
DOUGLAS	C	NV0000258	ZEPHYR COVE WATER UTILITY DISTR	IN	LAKE TAHOE INTAKE	IN01	P	2597	SW
NYE	C	NV0000063	GABBS WATER SYSTEM	WL	WELL 1	W01	P	426	GW
NYE	C	NV0000063	GABBS WATER SYSTEM	WL	WELL 3	W03	P	6787	GW
CLARK	C	NV0000011	BOULDER CITY	CC	CC FROM SNWA NV0000289	CC01	P	2	SW
CLARK	NC	NV0000942	LAKER PLAZA INC	WL	WELL 1	W01	P	3690	GW
ELKO	NTNC	NV0002524	CARLIN HONOR CAMP NDOC	WL	WELL 1	W01	P	647	GW
HUMBOLDT	NTNC	NV0005069	HUMBOLDT CONSERVATION CAMP NDOC	WL	WELL	W01	P	802	GW
LINCOLN	C	NV0000005	ALAMO SEWER AND WATER GID	WL	SANDHILL WELL	W07	P	6937	GW
LINCOLN	C	NV0000005	ALAMO SEWER AND WATER GID	WL	INDUSTRIAL PARK WELL AKA AIP	W08	P	6938	GW
NYE	NC	NV0000828	AMARGOSA PARK	WL	WELL	W01	P	525	GW
CHURCHILL	NC	NV0002581	SKIPS MINI MARKET	WL	SKIPS WELL	W01	P	679	GW
CLARK	C	NV0000010	BLUE DIAMOND WATER COMPANY	CC	CC FROM CERTAIN TEED GYPSUM NV0364	CC01	P	7759	GW
ESMERALDA	NTNC	NV0002184	DYER ELEMENTARY SCHOOL	WL	WELL 3	W03	P	6716	GW
WASHOE	C	NV0000071	GERLACH GID	SP	RAILROAD SPRING	SP01	P	3430	GW
WASHOE	C	NV0000071	GERLACH GID	SP	GARDEN SPRING	SP02	P	3431	GW
ELKO	NTNC	NV0003013	JERRITT CANYON GOLD JERRITT CANYON MINE	WL	WELL 5	W02	P	976	GW
ELKO	NTNC	NV0003013	JERRITT CANYON GOLD JERRITT CANYON MINE	WL	WELL WW7	W03	P	6470	GW
NYE	NTNC	NV0005067	MOUNTAIN VIEW MHP GBWC	WL	WELL 1	W01	P	800	GW
CLARK	NC	NV0004071	SPENCER W KIMBALL SCOUT RESERVATION	WL	SOUTH WELL UP MTN	W01	P	196	GW
CLARK	NC	NV0004071	SPENCER W KIMBALL SCOUT RESERVATION	WL	NORTH WELL BY POOL	W02	P	3441	GW
WASHOE	NC	NV0004064	GOLDEN VALLEY PARK	WL	WELL	W01	P	194	GW
ELKO	NC	NV0002069	JACK CREEK LODGE	WL	WELL 1	W01	P	2305	GW
CLARK	C	NV0004031	SKY RANCH ESTATES	WL	MAIN WELL	W01	P	1992	GW
CLARK	C	NV0004031	SKY RANCH ESTATES	WL	BACKUP WELL	W02	E	2361	GW
WASHOE	C	NV0000801	STAMPMILL	WL	EAST WELL	W01	P	1946	GW

Appendix D - Table of Active Sources for PWS in Nevada

WASHOE	C	NV0000801	STAMPMILL	WL	WEST WELL	W02	P	2411	GW
WASHOE	C	NV0002525	SUNRISE ESTATES	WL	SUNRISE ESTATES WELL 1	W01	P	1947	GW
WASHOE	C	NV0002525	SUNRISE ESTATES	WL	SUNRISE ESTATES WELL 3	W03	E	3396	GW
CHURCHILL	C	NV0000055	TOLAS WATERWORKS CO OP	WL	WELL 1A	W03	P	6520	GW
WASHOE	NTNC	NV0000773	VERDI MUTUAL WATER COMPANY	SP	SPRING	SP01	P	2342	GW
LYON	NC	NV0002128	CG BAR	WL	WELL 1	W01	P	592	GW
WHITE PINE	C	NV0000863	BAKER WATER AND SEWER GID	WL	WELL 1	W01	P	1905	GW
NYE	NTNC	NV0005037	AMARGOSA TOWN COMPLEX	WL	WELL	W01	P	790	GW
HUMBOLDT	NTNC	NV0000864	HYCROFT RESOURCES & DEVELOPMENT LLC	WL	WELL 1	W01	P	1922	GW
WASHOE	C	NV0000865	LIGHTNING W	WL	LIGHTNING W WELL 1	W01	P	1937	GW
WASHOE	C	NV0000865	LIGHTNING W	WL	LIGHTNING W WELL 2	W02	P	3388	GW
WASHOE	C	NV0000865	LIGHTNING W	WL	LIGHTNING W WELL 3	W03	P	5894	GW
ELKO	NC	NV0000873	SOUTHFORK STATE PARK AND RECREATION AREA	WL	WELL 1	W01	P	1915	GW
NYE	NC	NV0002141	HIGHWAY 95 FUEL & FOOD	WL	WELL	W01	P	599	GW
STOREY	NTNC	NV0000879	ASIA UNION ELECTRONIC CHEMICALS RENO	WL	WELL 1	W01	P	1926	GW
PERSHING	NTNC	NV0000884	FLORIDA CANYON MINING INC	WL	WELL 1 PW5	W01	P	1924	GW
STOREY	NTNC	NV0000878	MARS PETCARE US INC	WL	WELL 1	W01	P	1927	GW
WHITE PINE	NC	NV0000301	CAVE SPRINGS WATER SYSTEM NPS	SP	SPRING 1	SP01	P	484	GW
WHITE PINE	NC	NV0000301	CAVE SPRINGS WATER SYSTEM NPS	SP	SPRING 2	SP02	P	5537	GW
WHITE PINE	NC	NV0000301	CAVE SPRINGS WATER SYSTEM NPS	SP	SPRING 3	SP03	P	5538	GW
CHURCHILL	NC	NV0002586	TOP GUN SKATE	WL	WELL 1	W01	P	1964	GW
WHITE PINE	NC	NV0005059	BORDER INN	WL	WELL	W01	P	796	GW
WHITE PINE	NC	NV0002165	CHERRY CREEK WATER DISTRICT	WL	WELL 2	W02	P	5802	GW
CHURCHILL	NC	NV0002594	FALLON LIVESTOCK LLC	WL	WELL 2	W02	P	7629	GW
LINCOLN	NC	NV0002121	KERSHAW RYAN STATE PARK	WL	WELL 2	W02	P	6031	GW
WHITE PINE	NC	NV0002175	MAJORS STATION	WL	WELL	W01	P	1966	GW
EUREKA	C	NV0002574	DEVILS GATE GID DISTRICT 1	WL	DG1 FRONTIER STREET WELL	W01	P	3298	GW
NYE	NC	NV0002575	TATUADO WILD SIDE TAVERN	WL	WELL 1	W01	P	1967	GW
CLARK	NC	NV0000378	NEVADA TEST AND TRAINING RANGE SILVER FL	WL	SILVER FLAG WELL 2372	W01	P	1989	GW
NYE	NTNC	NV0000834	LAKESIDE CASINO AND RV PARK	WL	WELL 1	W01	P	529	GW
LANDER	NTNC	NV0000892	NEVADA GOLD MINES CORTEZ PIPELINE	WL	WELL 2	W02	P	6506	GW
ESMERALDA	NC	NV0000890	ESMERALDA MARKET AND GAS LLC	WL	WELL 1 STEEL WELL	W01	P	2289	GW
ESMERALDA	NC	NV0000890	ESMERALDA MARKET AND GAS LLC	WL	WELL 2 PVC	W02	P	3756	GW
WASHOE	NC	NV0000893	FRANKTOWN MEADOWS	WL	WELL 1	W01	P	2286	GW
WASHOE	NC	NV0000893	FRANKTOWN MEADOWS	WL	WELL 2	W02	P	3643	GW
NYE	NC	NV0000918	HORIZON C STORES 3 LINDA STREET	WL	WELL	W01	P	2283	GW
NYE	C	NV0000926	PAHRUMP UTILITY COMPANY INC	WL	WELL 2 BRIDGER ST	W02	S	3101	GW
NYE	C	NV0000926	PAHRUMP UTILITY COMPANY INC	WL	WELL 3 MANSE RD WELL	W03	S	5692	GW
NYE	C	NV0000926	PAHRUMP UTILITY COMPANY INC	WL	WELL 4 FOX AVE	W04	S	5784	GW
NYE	C	NV0000926	PAHRUMP UTILITY COMPANY INC	WL	HAFEN WELL 5 HERITAGE DR	W05	E	5785	GW
NYE	C	NV0000926	PAHRUMP UTILITY COMPANY INC	WL	WELL 1 REPLACEMENT	W06	P	6638	GW
CLARK	NC	NV0000380	SANDY VALLEY CENTER	WL	WELL	W01	P	2280	GW
STOREY	NTNC	NV0000913	TRI GENERAL IMPROVEMENT DISTRICT	WL	WELL 1	W01	P	2279	GW
STOREY	NTNC	NV0000913	TRI GENERAL IMPROVEMENT DISTRICT	WL	WELL 2	W02	P	3355	GW
STOREY	NTNC	NV0000913	TRI GENERAL IMPROVEMENT DISTRICT	WL	WELL 3	W03	P	3826	GW
STOREY	NTNC	NV0000913	TRI GENERAL IMPROVEMENT DISTRICT	WL	WELL 4 PORTOFINO	W04	P	5860	GW
STOREY	NTNC	NV0000913	TRI GENERAL IMPROVEMENT DISTRICT	WL	WELL 5	W05	P	6630	GW

Appendix D - Table of Active Sources for PWS in Nevada

STOREY	NTNC	NV0000913	TRI GENERAL IMPROVEMENT DISTRICT	WL	WELL 6	W06	P	6631	GW
STOREY	NTNC	NV0000913	TRI GENERAL IMPROVEMENT DISTRICT	WL	WELL 7	W07	P	8977	GW
STOREY	NTNC	NV0000913	TRI GENERAL IMPROVEMENT DISTRICT	WL	WELL 8	W08	P	8976	GW
MINERAL	NTNC	NV0000897	SCHURZ ELEMENTARY SCHOOL	WL	WELL 1	W01	P	2232	GW
LINCOLN	NC	NV0000898	UNR 4H CAMP AND LEARNING CENTER	WL	WELL 1	W01	P	2300	GW
LINCOLN	NC	NV0000898	UNR 4H CAMP AND LEARNING CENTER	WL	WELL 2	W02	P	4874	GW
CHURCHILL	NTNC	NV0000903	NEW MILLENNIUM BUILDING SYSTEMS, LLC	WL	WELL 1	W01	P	2234	GW
NYE	NTNC	NV0005036	SHERIS RANCH	WL	WELL 1	W01	P	789	GW
NYE	NTNC	NV0005036	SHERIS RANCH	WL	NEW WELL 2	W03	P	7072	GW
CLARK	NTNC	NV0001000	LHOIST NORTH AMERICA	WL	WELL 1 NORTH OLD WELL	W01	P	2292	GW
CLARK	NTNC	NV0001000	LHOIST NORTH AMERICA	WL	WELL 2 SOUTH NEW WELL	W02	P	3679	GW
CLARK	C	NV0003005	NPS BOULDER BEACH	CC	CC FROM BOULDER CITY	CC01	P	2553	SW
HUMBOLDT	NTNC	NV0000907	LONE TREE MINE	WL	WELL SS 13	W03	P	3215	GW
PERSHING	NC	NV0002222	RYE PATCH PETROLEUM	WL	WELL 1	W01	P	637	GW
WASHOE	C	NV0000767	ROSEMOUNT WATER CO	SP	SPRING 1 MAIN	SP01	P	2547	GW
DOUGLAS	NC	NV0004033	CAMP GALILEE	WL	WELL 1	W01	P	741	GW
ELKO	NC	NV0004050	CAMP LAMOILLE LIONS CLUB	WL	WELL	W01	P	3450	GW
CLARK	NTNC	NV0001083	HOOVER DAM LODGE DBA HACIENDA	CC	CC FROM BOULDER CITY WATER NV0000011	CC01	P	2566	SW
WASHOE	NC	NV0004077	LEMMON VALLEY HORSEMANS PARK	WL	WELL	W01	P	3285	GW
WASHOE	NTNC	NV0000870	NORTH VALLEY BUS FACILITY	WL	WELL	W01	P	1942	GW
WASHOE	NC	NV0004079	ROYAL MARKET 2	WL	WELL	W01	P	2306	GW
CHURCHILL	NC	NV0000938	LATTIN FARMS	WL	KITCHEN WELL	W01	P	3475	GW
CHURCHILL	NC	NV0000929	SONIC BURGER	WL	WELL 1	W01	P	2304	GW
NYE	NC	NV0000946	WHOS DUNES	WL	WELL 1	W01	P	3706	GW
NYE	NC	NV0000943	NDOT SUNNYSIDE ROADSIDE PARK RP810NY	WL	SUNNYSIDE WELL	W01	P	3723	GW
WASHOE	NC	NV0000950	MEMORIAL POINT STATE PARK	WL	WELL	W01	P	3828	GW
LYON	NC	NV0000951	ROSIES PLACE	WL	WELL 1	W01	P	3906	GW
CLARK	NC	NV0003004	NPS LAS VEGAS BAY	CC	CC SW FROM CITY OF HENDERSON NV0000076	CC01	P	4010	SW
CLARK	NC	NV0000932	KEYSTONE CENTER	WL	SOUTH WELL	W01	P	3910	GW
WASHOE	NTNC	NV0000978	TRUCKEE CANYON	WL	TRUCKEE CANYON WELL 3	W03	P	8429	GW
WASHOE	NTNC	NV0000978	TRUCKEE CANYON	WL	TRUCKEE CANYON WELL 4	W04	P	9337	GW
WHITE PINE	NTNC	NV0000979	LUND SCHOOL	WL	WELL 1	W01	P	4144	GW
WHITE PINE	NC	NV0000961	HIDDEN CANYON RANCH	WL	WELL 1	W01	P	3990	GW
CLARK	NTNC	NV0000364	CERTAIN TEED GYPSUM	WL	SOUTH WELL	W01	P	7880	GW
CLARK	NTNC	NV0000364	CERTAIN TEED GYPSUM	WL	NORTH WELL	W02	P	7881	GW
CHURCHILL	NC	NV0000306	THE GRID	WL	WELL	W01	P	4204	GW
CLARK	NC	NV0000955	BLUE DIAMOND TRAVEL CENTER H325	WL	TRAVEL CENTER WELL 1 400FT BGS	W01	P	4201	GW
NYE	NC	NV0000917	GAMEBIRD PLAZA LLC	WL	WELL	W01	P	2284	GW
DOUGLAS	C	NV0000959	FOOTHILL WATER SYSTEM	WL	UPPER WELL/ JOBS PEAK WELL 1	W01	P	4019	GW
DOUGLAS	C	NV0000959	FOOTHILL WATER SYSTEM	WL	WELL 4 SHERIDAN	W04	P	5699	GW
DOUGLAS	C	NV0000959	FOOTHILL WATER SYSTEM	WL	WELL 2R	W05	P	6188	GW
EUREKA	NTNC	NV0000338	NEVADA GOLD MINES CARLIN LEEVILLE	WL	HDDW-06	W03	P	7396	GW
EUREKA	NTNC	NV0000338	NEVADA GOLD MINES CARLIN LEEVILLE	WL	HDDW-08	W04	P	7397	GW
NYE	NC	NV0000923	ELKS LODGE PAHRUMP	WL	WELL	W01	P	3919	GW
CLARK	NTNC	NV0000342	SILVERHAWK POWER PLANT	WL	WELL 1	W01	P	4587	GW
NYE	C	NV0000920	MOUNTAIN FALLS WATER SYSTEM GBWC	WL	WELL 1	W01	P	2282	GW
NYE	C	NV0000920	MOUNTAIN FALLS WATER SYSTEM GBWC	WL	WELL 2	W02	P	3300	GW

Appendix D - Table of Active Sources for PWS in Nevada

DOUGLAS	C	NV0000931	SIERRA COUNTRY ESTATES	WL	LOWER EAST WELL	W01	P	3915	GW
DOUGLAS	C	NV0000931	SIERRA COUNTRY ESTATES	WL	UPPER WEST WELL	W02	P	4371	GW
CLARK	NC	NV0000331	J AND J GENERAL STORE	WL	WELL	W01	P	496	GW
WHITE PINE	NTNC	NV0000855	ROBINSON NEVADA MINING COMPANY	WL	WELL NRC-1P	W01	P	1892	GW
NYE	NC	NV0002565	COYOTE CORNER MARKET	WL	WELL 2	W02	P	5775	GW
WASHOE	NC	NV0000381	BLACK ROCK STATION	WL	WELL	W01	P	5322	GW
LYON	NC	NV0000389	BUCKBOARD STATION	WL	WELL	W01	P	5364	GW
HUMBOLDT	NC	NV0000001	DENIO JUNCTION	WL	WELL 1	W01	P	388	GW
WASHOE	NC	NV0000866	AIR SAILING INC GLIDERPORT	WL	WELL	W01	P	5429	GW
LANDER	NTNC	NV0000902	NEVADA GOLD MINES PHOENIX	WL	WELL 2	W02	P	6608	GW
CHURCHILL	NC	NV0000921	OASIS SPRINGS	WL	WELL 1	W01	P	2281	GW
CLARK	NC	NV0000392	LOVELL CANYON RESIDENTIAL LLC	WL	WELL	W01	P	5515	GW
EUREKA	NTNC	NV0000885	RUBY HILL MINE	WL	WELL 3	W03	P	5519	GW
LINCOLN	NC	NV0000399	LCWS RANCH	WL	WELL	W01	P	5521	GW
EUREKA	NTNC	NV0000403	NEVADA GOLD ENERGY TS POWER PLANT	WL	WELL	W01	P	5645	GW
NYE	C	NV0000402	PLEASANT VALLEY	WL	CONCORDIA EAST WELL 1	W01	P	5610	GW
NYE	C	NV0000402	PLEASANT VALLEY	WL	WELL 2 REPLACEMENT	W03	P	9374	GW
CHURCHILL	C	NV0000406	SAND CREEK	WL	WELL 1	W01	P	5760	GW
CHURCHILL	C	NV0000406	SAND CREEK	WL	WELL 2	W02	P	9684	GW
NYE	NC	NV0000405	CARBERRY SQUARE	WL	WELL	W01	P	5701	GW
LANDER	NC	NV0003066	BOB SCOTT CAMPGROUND USFS	WL	WELL	W01	P	5725	GW
CHURCHILL	NC	NV0002014	NEVADA LIVESTOCK MARKETING	WL	WELL 1	W01	P	540	GW
ELKO	NC	NV0000949	DOVETAIL RANCH	WL	WELL 1	W01	P	3714	GW
WASHOE	NTNC	NV0002202	GRANITE CONSTRUCTION	WL	WELL	W01	P	6024	GW
CARSON CITY	NP	NV0000031	STATE WATER SHED	IN	MARLETTE INTAKE	IN01	P	405	SW
CARSON CITY	NP	NV0000031	STATE WATER SHED	SP	TUNNEL SPRINGS DIVERSION	SP06	P	5922	GU
HUMBOLDT	NTNC	NV0002095	PILOT TRAVEL CENTER 485	WL	WELL	W01	P	5911	GW
NYE	NC	NV0002196	DESERT CENTER PLAZA	WL	WELL	W01	P	5986	GW
WHITE PINE	NC	NV0003049	TIMBER CREEK CAMPGROUND USFS	SP	SPRING	SP01	P	3198	GW
LYON	NTNC	NV0002515	SNYDER LIVESTOCK COMPANY INC	WL	WELL	W01	P	6061	GW
CLARK	NTNC	NV0002533	CHUCK LENZIE GENERATING STATION	WL	CHUCK WELL WS-1	W01	P	6092	GW
CLARK	NTNC	NV0002533	CHUCK LENZIE GENERATING STATION	WL	HARRY WELL HARVEY	W03	P	6232	GW
CLARK	NTNC	NV0002533	CHUCK LENZIE GENERATING STATION	WL	CHUCK WELL WS-3	W04	P	8067	GW
LANDER	NTNC	NV0002570	NEVADA GOLD MINES CORTEZ MILL 1	WL	WELL 1	W01	P	6117	GW
CLARK	NTNC	NV0002577	SCPPA APEX GENERATING STATION	WL	WELL	W01	P	6206	GW
CLARK	NTNC	NV0002602	LOVES TRAVEL STOP AND COUNTRY STORE	WL	WELL	W01	P	6211	GW
CLARK	C	NV0001102	SNWA COYOTE SPRING VALLEY-MOAPA PWS	WL	MOAPA MX-5 WELL	W71	P	6618	GW
ELKO	NC	NV0001091	CALIFORNIA NATIONAL HISTORIC TRAILS CTR	WL	WELL	W01	P	6292	GW
CHURCHILL	NC	NV0002017	COLD SPRINGS STATION RESORT	WL	WELL 2	W02	P	6281	GW
LINCOLN	NC	NV0002563	CAMP FRIAS FRONTIER	WL	WELL 2	W02	P	7557	GW
NYE	C	NV0001093	SPRING MOUNTAIN MOTOR SPORTS RANCH	WL	SMMR WELL 1	W02	P	7837	GW
NYE	C	NV0001093	SPRING MOUNTAIN MOTOR SPORTS RANCH	WL	SMMR WELL 2	W03	P	7838	GW
NYE	NC	NV0001094	PATCH OF HEAVEN	WL	WELL	W01	P	6376	GW
NYE	C	NV0000408	CALVADA MEADOWS GBWC	WL	WELL	W01	P	5854	GW
WHITE PINE	NC	NV0000922	WARD CHARCOAL OVENS STATE PARK	WL	CG WELL	W01	P	3702	GW
NYE	NTNC	NV0001095	CEDAR PASS WATER SYSTEM	WL	WELL	W01	P	6438	GW
NYE	NC	NV0001096	SANDERS WINERY	WL	WELL	W01	P	6444	GW

Appendix D - Table of Active Sources for PWS in Nevada

LYON	NC	NV0002608	STAGECOACH COUNTRY MARKET	WL	WELL 1	W01	P	695	GW
LANDER	NTNC	NV0001097	NEVADA GOLD MINES CORTEZ HILLS	WL	WELL 1	W01	P	6492	GW
NYE	NC	NV0000385	COYOTE CORNER III	WL	WELL 2	W02	P	9089	GW
NYE	NC	NV0001105	EL VALLE RESTAURANT	WL	WELL	W01	P	6690	GW
CLARK	NTNC	NV0001106	CITY CENTER HOTELS	CC	LVVWD INTERTIE N2 ARIA N1 VDARA	CC01	P	6715	SW
WHITE PINE	NC	NV0003053	BIRD CREEK CAMPGROUND USFS	WL	WELL	W01	P	716	GW
CLARK	NC	NV0001024	CORN CREEK FIELD STATION FWS	WL	WELL 1	W01	P	6730	GW
HUMBOLDT	NTNC	NV0001103	MARIGOLD MINE POTABLE WATER SYSTEM	WL	WW-1	W01	P	6653	GW
WHITE PINE	NTNC	NV0000982	BALD MOUNTAIN MINE	WL	WELL	W01	P	6253	GW
CLARK	NTNC	NV0001111	LUXOR RESORT AND CASINO	CC	LVVWD INTERTIE N IS 2	CC01	P	6811	SW
CLARK	NTNC	NV0001112	NEW YORK NEW YORK HOTEL AND CASINO	CC	LVVWD INTERTIE QTY 2	CC01	P	6817	SW
CLARK	NTNC	NV0001113	CIRCUS CIRCUS CASINO	CC	LVVWD INTERTIE N IS 5	CC01	P	6829	SW
CLARK	NTNC	NV0001114	EXCALIBUR RESORT AND CASINO	CC	LVVWD INTERTIE	CC01	P	6836	SW
CLARK	C	NV0001116	SIGNATURE TOWERS	CC	LVVWD INTERTIE N IS 2	CC01	P	6844	SW
CLARK	NTNC	NV0001107	KAPEX WATER SYSTEM CITY OF NLV	WL	WELL 1	W01	P	6719	GW
CLARK	NTNC	NV0001117	BELLAGIO RESORT AND CASINO	CC	LVVWD INTERTIE	CC01	P	6869	SW
CLARK	NTNC	NV0001118	PARK MGM	CC	LVVWD INTERTIE N IS 2	CC01	P	6876	SW
CLARK	NTNC	NV0001119	MANDALAY BAY RESORT AND CASINO	CC	LVVWD INTERTIE N IS 4	CC01	P	6882	SW
CLARK	NTNC	NV0001120	MIRAGE RESORT AND CASINO	CC	LVVWD INTERTIE	CC01	P	6891	SW
NYE	NC	NV0000829	MEXICAN GRUB 5280	WL	WELL 1	W01	P	526	GW
CLARK	NTNC	NV0001121	MGM GRAND HOTEL AND CASINO	CC	LVVWD INTERTIE N IS 2	CC01	P	7037	SW
NYE	C	NV0002571	RANCHO VISTA 4	WL	WELL 1	W01	P	5855	GW
NYE	C	NV0002571	RANCHO VISTA 4	WL	WELL 2	W02	E	5856	GW
LYON	NC	NV0000341	CARMENS MEXICAN RESTAURANT	WL	WELL	W01	P	4581	GW
LYON	NC	NV0004040	STAGECOACH MARKET	WL	WELL 1	W01	P	745	GW
PERSHING	NC	NV0001125	HUMBOLDT RIVER RANCH ASSOCIATION	WL	WELL 2	W02	P	8194	GW
NYE	NTNC	NV0001122	ROUND MOUNTAIN GOLD HILL WATER SYSTEM	WL	SINTER 2 WELL	W02	P	7372	GW
ELKO	NC	NV0001092	RYNDON GAS & FOOD LLC	WL	WELL 1	W01	P	7095	GW
MINERAL	NC	NV0001128	WILDKAT RANCH	WL	WELL 1	W01	P	7134	GW
DOUGLAS	NC	NV0002041	BEST WESTERN TOPAZ LAKE INN	WL	WELL 1	W01	P	1993	GW
CLARK	NTNC	NV0001127	POLO TOWERS	CC	LVVWD INTERTIE	CC01	P	7057	SW
CLARK	NC	NV0001101	NDOT SEARCHLIGHT WELCOME CENTER	WL	WELL 1	W01	P	6514	GW
WHITE PINE	NC	NV0003046	WARD MOUNTAIN CAMPGROUND USFS	WL	WELL 2	W02	P	9034	GW
WASHOE	NTNC	NV0001132	RENO TECHNOLOGY PARK WATER COMPANY	WL	RTP 2 WELL	W01	P	7214	GW
WASHOE	NTNC	NV0001132	RENO TECHNOLOGY PARK WATER COMPANY	WL	RTP 3 WELL	W02	P	7215	GW
WASHOE	NTNC	NV0001132	RENO TECHNOLOGY PARK WATER COMPANY	WL	RTP APPLE WELL 4	W03	P	9643	GW
WASHOE	NC	NV0004065	WASHOE VALLEY MEETINGHOUSE FACILITY LDS	WL	WELL 1	W01	P	7337	GW
NYE	NC	NV0000386	TOWER PIZZA	WL	WELL	W01	P	5343	GW
NYE	NC	NV0003035	THE HUBB	WL	WELL 1	W01	P	1968	GW
CLARK	NC	NV0004024	CAMP STIMSON LDS	SP	STIMPSON SPRING	SP01	P	7334	GW
CLARK	NTNC	NV0001160	HILTON GRAND VACATIONS CONVENTION	CC	LVVWD INTERTIE	CC01	P	7604	SW
LYON	NC	NV0000881	DAYTON TAP HOUSE LLC	WL	WELL	W01	P	1919	GW
CLARK	NTNC	NV0001141	WYNN RESORTS	CC	LVVWD INTERTIES N IS 3	CC01	P	7554	SW
CLARK	NTNC	NV0001137	COSMOPOLITAN HOTEL	CC	LVVWD INTERTIE N IS 2	CC01	P	7347	SW
ELKO	NC	NV0004005	LDS LEE RECREATION CAMP	WL	WELL 2	W02	P	8597	GW
HUMBOLDT	NTNC	NV0002198	KINGS RIVER ELEMENTARY SCHOOL	WL	WELL 2	W02	P	7648	GW
CLARK	NTNC	NV0001140	SLOAN ARMY RESERVE CENTER	WL	WELL 1	W01	P	7353	GW

Appendix D - Table of Active Sources for PWS in Nevada

ELKO	NC	NV0002080	WILDHORSE RESORT	WL	WELL 1	W01	P	572	GW
PERSHING	NC	NV0001124	GOLD DIGGERS SALOON & GRUBB HOUSE	WL	WELL 1	W01	P	7045	GW
CLARK	NC	NV0003103	SPRING MOUNTAINS VISITOR GATEWAY	WL	USFS WELL	W01	P	7198	GW
CLARK	NTNC	NV0001144	GRANDVIEW AT LAS VEGAS	CC	LVVWD INTERTIE N IS 2	CC01	P	7926	SW
LANDER	NC	NV0004105	ANTELOPE VALLEY MEETINGHOUSE LDS	WL	WELL 1	W01	P	7998	GW
EUREKA	NC	NV0004104	EUREKA MEETINGHOUSE LDS	WL	WELL 1	W01	P	8000	GW
CLARK	NC	NV0004102	SANDY VALLEY MEETINGHOUSE LDS	WL	WELL	W01	P	8005	GW
LYON	NC	NV0004100	SILVER SPRINGS MEETINGHOUSE LDS	WL	WELL 1	W01	P	8004	GW
WASHOE	NC	NV0004103	LEMMON VALLEY MEETINGHOUSE LDS	WL	WELL	W01	P	8009	GW
WHITE PINE	NC	NV0004101	LUND MEETINGHOUSE LDS	WL	WELL 1	W01	P	8002	GW
CHURCHILL	NC	NV0002580	HARMON JUNCTION	WL	WELL 1	W01	P	678	GW
CHURCHILL	NTNC	NV0004028	OLAM SVI	WL	WELL	W01	P	8085	GW
LYON	NC	NV0004107	SMITH VALLEY BAPTIST CHURCH	WL	WELL	W01	P	8094	GW
WASHOE	NC	NV0004088	ANIMAL ARK	WL	WELL 1	W01	P	7339	GW
NYE	NTNC	NV0001145	US ECOLOGY NEVADA	WL	WELL 1	W01	P	7965	GW
WHITE PINE	NTNC	NV0001142	IORE GOLD PAN MINE	WL	WELL 1	W01	P	7792	GW
ELKO	NTNC	NV0001138	NEVADA GOLD MINES LONG CANYON	WL	WELL 1	W01	P	7614	GW
HUMBOLDT	NTNC	NV0001148	DIAMOND PLASTICS	WL	WELL 2	W02	P	8472	GW
WASHOE	NC	NV0004109	GRANITE HILLS BAPTIST CHURCH	WL	WELL 1	W01	P	8431	GW
NYE	NC	NV0004063	PICK AND SHOVEL INVESTMENTS	WL	WELL	W01	P	7776	GW
WASHOE	NTNC	NV0004061	SPRINGWOOD EQUESTRIAN CENTER	WL	WELL	W01	P	8484	GW
CLARK	NTNC	NV0001130	M RESORT SPA AND CASINO	CC	CC FROM HENDERSON NV0076	CC01	P	7204	SW
CHURCHILL	NTNC	NV0004120	EP MINERALS LLC FERNLEY	WL	WELL	W01	P	8794	GW
CLARK	NTNC	NV0004108	SPEED VEGAS	WL	WELL 1	W01	P	8397	GW
ELKO	NC	NV0004112	RUBY 360 LODGE	WL	WELL	W01	P	8566	GW
CLARK	NTNC	NV0001150	HILTON GRAND VACATIONS LAS VEGAS BLVD	CC	CC INTERTIE FROM LVVWD NV0090	CC01	P	7597	SW
CLARK	NC	NV0004029	RITCHIE BROTHERS	WL	WELL	W01	P	8307	GW
CLARK	NTNC	NV0001143	HILTON GRAND VACATIONS ELARA	CC	LVVWD INTERTIE	CC01	P	7850	SW
CLARK	NTNC	NV0001136	HILTON GRAND VACATIONS FLAMINGO	CC	CC INTERTIE FROM LVVWD NV0099	CC01	P	7328	SW
CLARK	NC	NV0003100	RED ROCK CAMPGROUND BLM	WL	WELL 1 LISTED ACTIVE WITH OUTS 2015 TBA	W01	P	7188	GW
CLARK	NC	NV0003101	RED ROCK FIRE STN AND LAW ENF BLM	WL	WELL	W01	P	7190	GW
NYE	NTNC	NV0000418	CRESCENT DUNES SOLAR ENERGY	WL	WELL 2	W02	P	7904	GW
NYE	NTNC	NV0000418	CRESCENT DUNES SOLAR ENERGY	WL	WELL 3	W03	P	7950	GW
NYE	NC	NV0004026	BASE CAMP US AIR FORCE	WL	WELL 1	W01	P	7609	GW
NYE	NC	NV0004026	BASE CAMP US AIR FORCE	WL	WELL 2	W02	P	8478	GW
NYE	NC	NV0004027	ASH MEADOWS NATIONAL WILDLIFE REFUGE FWS	WL	WELL	W01	P	7781	GW
CLARK	NTNC	NV0001165	VENETIAN PALAZZO	CC	LVVWD INTERTIE QTY 3	CC01	P	8792	SW
CLARK	NTNC	NV0001167	RIO RESORT AND CASINO	CC	LVVWD INTERTIE N IS 2	CC01	P	8900	SW
CLARK	C	NV0000699	NEVADA STATE VETERANS HOME BOULDER CITY	CC	CC01 BOULDER CITY N IS 2	CC01	P	8919	SW
HUMBOLDT	NC	NV0002094	DIAMOND INN BAR	WL	WELL	W01	P	581	GW
CLARK	NTNC	NV0001155	BERKLEY	CC	LVVWD INTERTIE N IS 2	CC01	P	8790	SW
LYON	NC	NV0004118	WALKER RIVER STATE REC AREA PITCHFORK	WL	WELL 1	W01	P	8998	GW
CLARK	C	NV0000124	SUNRISE ACRES WATER ASSOCIATION	WL	WELL 1	W01	P	21	GW
CLARK	C	NV0000124	SUNRISE ACRES WATER ASSOCIATION	WL	WELL 2	W02	P	264	GW
LINCOLN	NC	NV0004106	PAHRANAGAT NATIONAL WILDLIFE REFUGE FWS	WL	WELL 1	W01	P	8039	GW
WASHOE	NC	NV0000318	WASHOE LAKE STATE PARK NORTH BOAT RAMP	WL	NORTH RAMP WELL	W01	P	4540	GW
WASHOE	NC	NV0004076	THE LODGE AT GALENA	WL	WELL	W01	P	198	GW

Appendix D - Table of Active Sources for PWS in Nevada

CLARK	NTNC	NV0001162	HORSESHOE LAS VEGAS HOTEL & CASINO	CC	LVVWD INTERTIE QTY 2	CC01	P	9148	SW
WASHOE	NC	NV0004135	ECOLOGICA CORPORATION	WL	WELL 1	W01	P	9138	GW
WASHOE	NC	NV0002609	DOLLAR GENERAL	WL	WELL 1	W01	P	9122	GW
EUREKA	NTNC	NV0001159	GOLD BAR MINE	WL	WELL 210	W01	P	8944	GW
EUREKA	NTNC	NV0001159	GOLD BAR MINE	WL	WELL 213	W02	P	8945	GW
CLARK	NTNC	NV0001175	FLAMINGO LAS VEGAS HOTEL AND CASINO	CC	LVVWD INTERTIE N IS 2	CC01	P	9192	SW
CLARK	NTNC	NV0001171	HARRAHS LAS VEGAS	CC	LVVWD INTERTIE N IS 2	CC01	P	9153	SW
CLARK	NTNC	NV0001166	PARIS LAS VEGAS HOTEL AND CASINO	CC	LVVWD INTERTIE N IS 2	CC01	P	9219	SW
ELKO	NC	NV0001088	MUSTANG MONUMENT	WL	WELL 1	W01	P	7259	GW
CLARK	NTNC	NV0001163	LINQ HOTEL AND CASINO	CC	INTERTIE LVVWD N IS 2	CC01	P	9244	SW
CLARK	NTNC	NV0001172	WALDORF ASTORIA HOTEL LAS VEGAS	CC	CC LVVWD VIA CITY CENTER	CC01	P	9216	SW
NYE	NTNC	NV0001174	FRONT SIGHT FIREARMS TRAINING INSTITUTE	WL	WELL 1	W01	P	9416	GW
CLARK	NTNC	NV0001180	PLANET HOLLYWOOD HOTEL AND CASINO	CC	CC INTERTIE FROM LVVWD NV0090	CC01	P	9205	SW
MINERAL	NTNC	NV0001178	ISABELLA PEARL MINE	WL	WELL 1	W01	P	9503	GW
MINERAL	NTNC	NV0001178	ISABELLA PEARL MINE	WL	WELL 3	W03	P	9504	GW
HUMBOLDT	NTNC	NV0001191	CSS FARMS LLC	WL	WELL 1	W01	P	9533	GW
NYE	NTNC	NV0001181	ACRES CULTIVATION	WL	WELL 1	W01	P	9548	GW
CLARK	NTNC	NV0001164	CROMWELL HOTEL AND CASINO	CC	INTERTIE LVVWD N IS 2	CC01	P	9246	SW
CLARK	NTNC	NV0001190	T MOBILE ARENA	CC	CC INTERTIE FROM LVVWD NV0090	CC01	P	9488	SW
DOUGLAS	C	NV0000172	PINEVIEW ESTATES	WL	WELL 1	W01	P	9529	GW
CLARK	NC	NV0004131	BLUE DIAMOND RAINBOW NW PLAZA WATER	WL	WELL 1	W01	P	9591	GW
WASHOE	NC	NV0004145	HOLY SPIRIT CATHOLIC CHURCH	WL	WELL 1	W01	P	9619	GW
NYE	NC	NV0004132	FORELAND REFINERY	WL	WELL 1	W01	P	9616	GW
CLARK	C	NV0000430	FORT APACHE ANN NE WATER ASSOC	WL	WELL 01	W01	P	9626	GW
CLARK	NC	NV0004119	COYOTE SPRINGS GOLF COURSE	WL	WELL 1	W01	P	9638	GW
CLARK	NC	NV0004119	COYOTE SPRINGS GOLF COURSE	WL	WELL 4	W02	P	10292	GW
CLARK	NTNC	NV0001177	MARRIOTT GRAND CHATEAU	CC	INTERTIE LVVWD N IS 1	CC01	P	9260	SW
CLARK	NTNC	NV0001210	RESORTS WORLD LAS VEGAS	CC	CC INTERTIE FROM LVVWD NV0090	CC01	P	9725	SW
DOUGLAS	NTNC	NV0001201	HARRAHS LAKE TAHOE	CC	CC FROM EDGEWOOD NV0000235	CC01	P	9768	SW
DOUGLAS	NTNC	NV0001182	HARVEYS LAKE TAHOE	CC	CC01 FROM EDGEWOOD NV0000235	CC01	P	9765	SW
CLARK	NTNC	NV0001169	CAESARS PALACE	CC	INTERTIE LVVWD N IS 3	CC01	P	9239	SW
CLARK	NTNC	NV0004137	WALLFLOWER	WL	WELL 1	W01	P	9824	GW
CLARK	NTNC	NV0001168	HARRAHS LAUGHLIN	CC	CC INTERTIE WITH BBWD N IS 2	CC01	P	9227	SW
CLARK	NTNC	NV0001206	WYNDHAM GRAND DESERT	CC	CC INTERTIE FROM LVVWD NV0090	CC01	P	9941	SW
NYE	NC	NV0004139	PAHRUMP MUSEUM	WL	WELL 1	W01	P	10109	GW
CLARK	C	NV0000431	ELKHORN WATER ASSOCIATION LLC	WL	WELL 1	W01	P	10134	GW
NYE	NC	NV0004138	BLOSSER PARK	WL	WELL 1	W01	P	10183	GW
NYE	NC	NV0004115	KELLOGG PARK	WL	WELL 1	W01	P	9917	GW
WASHOE	NTNC	NV0001196	PARR RENO WATER COMPANY	CC	CC FROM TMWA	CC01	P	9683	SW
NYE	NC	NV0004142	GAMEBIRD CIRCLE K	WL	WELL 1	W01	P	10241	GW
CLARK	C	NV0000426	DESERT SUNRISE WATER USERS ASSOC	WL	WELL 1 W FORD AVE	W01	P	9717	GW
NYE	NC	NV0004147	DOLLAR GENERAL 15044	WL	WELL 1	W01	P	10279	GW
CLARK	NTNC	NV0001188	VALLEY HEALTH SPECIALTY HOSPITAL	CC	CC INTERTIE FROM LVVWD NV0090	CC01	P	10286	SW
NYE	NC	NV0004127	THE STAGE STOP	WL	WELL 1	W01	P	9342	GW
CLARK	NC	NV0004146	HURRY IT UP WE ARE BURNING DAYLIGHT	WL	WELL 01	W01	P	9839	GW
CLARK	NTNC	NV0001186	FIRST SLOAN INDUSTRIAL WATER SYSTEM	WL	WELL 1	W01	P	10029	GW

Appendix D - Table of Active Sources for PWS in Nevada

Notes:		
PWS	Public Water System	
C	Community	
NC	Non-Community Transient	
NP	Non-Public	
NTNC	Non-Transient, Non-Community	
Type Source		
CC	Consecutive Connection	
IN	Intake	
NP	Non-Potable (Truck)	
SP	Spring	
WL	Well	
Availability		
E	Emergency	
I	Interim	
P	Permanent	
S	Seasonal	
Water Type		
GU	Groundwater UDI Surface	
GW	Groundwater	
SW	Surface water	

Appendix E

Sample Collection Form

Sample Collection Form

Project: NDEP PFAS Sampling
Client: NDEP
Project #: 24-02-172

Sample Type: Drinking Water / Surface Water (Circle one)
Date: _____



PWS #	State ID	Sample ID	Sample Time	# of sample containers		Sample Location (Decimal degrees NAD 83)	
	(W-##)						
				Method 533		Latitude	
				Method 537.1		Longitude	
Sample Collected: YES / NO (circle one)				Method 1633		GPS accuracy (ft)	
Comments (weather conditions, field parameters if surface water sampled):				Quality Control Samples (List Duplicate or MS/MSD)			
Sampler:							

PWS #	State ID	Sample ID	Sample Time	# of sample containers		Sample Location (Decimal degrees NAD 83)	
	(W-##)						
				Method 533		Latitude	
				Method 537.1		Longitude	
Sample Collected: YES / NO (circle one)				Method 1633		GPS accuracy (ft)	
Comments (weather conditions, field parameters if surface water sampled):				Quality Control Samples (List Duplicate or MS/MSD)			
Sampler:							

PWS #	State ID	Sample ID	Sample Time	# of sample containers		Sample Location (Decimal degrees NAD 83)	
	(W-##)						
				Method 533		Latitude	
				Method 537.1		Longitude	
Sample Collected: YES / NO (circle one)				Method 1633		GPS accuracy (ft)	
Comments (weather conditions, field parameters if surface water sampled):				Quality Control Samples (List Duplicate or MS/MSD)			
Sampler:							

PWS #	State ID	Sample ID	Sample Time	# of sample containers		Sample Location (Decimal degrees NAD 83)	
	(W-##)						
				Method 533		Latitude	
				Method 537.1		Longitude	
Sample Collected: YES / NO (circle one)				Method 1633		GPS accuracy (ft)	
Comments (weather conditions, field parameters if surface water sampled):				Quality Control Samples (List Duplicate or MS/MSD)			
Sampler:							

Appendix F

Chain of Custody Record

Ver: 02/24/23

880 Riverside Parkway

West Sacramento, CA 95605-1500
phone 916.373.5600 fax 303.467.7248

Chain of Custody Record

Sample Origin: State of Nevada

Regulatory Program: ☐ DW ☐ NPDES ☐ RCRA ☐ Other:

TestAmerica Laboratories, Inc. d/b/a Eurofins TestAmerica

[illegible]

INSTRUCTIONS

- 1) Choose the correct Eurofins TestAmerica Facility from the pull down list by clicking on cell A1
- 2) Fill in the appropriate information for your location and phone number
- 3) Sampler - Fill in name.
- 4) Provide information on the Regulatory Program to differentiate between Drinking Water & Compliance samples.
- 5) Choose a default TAT or enter a different one if appropriate
- 6) Please indicate whether the TAT is Working or Calendar Days
- 7) In the vertical columns enter the Method/Analysis being requested
- 8) Fill out the Sample Information -- each line represents one sample
- 9) Sample Date/Time is required on all samples
- 10) In the "# of Containers" field enter the total number of bottles for each sample
- 11) Check Y or N if the sample was filtered in the field (Filtered Sample).
- 12) Note 'C' for a Composite sample; or 'G' for a Grab Sample.
- 13) The Sample name should be the one you wish to see in the final report
- 14) In the cell where the Sample Information intersects the method information please enter the number of containers submitted for the method. Alternatively simply "x" this field
- 15) In the last row of the eCOC please choose the code for the right preservative used
- 16) Note any Possible Hazards.
- 17) Use the Special Instructions field to add any special instructions to the lab
- 18) If samples are sent across the country, consider indicating the Time Zone where samples were collected
- 19) Eurofins TestAmerica Terms and Conditions apply for the analysis performed on the submitted samples unless otherwise agreed upon between Eurofins TestAmerica and Company



CHAIN-OF-CUSTODY Analytical Request Document

Chain-of-Custody is a LEGAL DOCUMENT - Complete all relevant fields

LAB USE ONLY- Affix Workorder/Login Label Here



Scan QR Code for instructions

Company Name:	Contact/Report To:
Street Address:	Phone #:
	E-Mail:
	Cc E-Mail:
Customer Project #:	Invoice to:
Project Name:	Invoice E-mail:
Site Collection Info/Facility ID (as applicable):	Purchase Order # (if applicable):
	Quote #:
Time Zone Collected: ☐ AK ☐ PT ☐ MT ☐ CT ☐ ET	County / State origin of sample(s):

Data Deliverables:			Regulatory Program (DW, RCRA, etc.) as applicable:		Reportable ☐ Yes ☐ No	
☐ Level II	☐ Level III	☐ Level IV	Rush (Pre-approval required): ☐ Same Day ☐ 1 Day ☐ 2 Day ☐ 3 Day Other _____		DW PWSID # or WW Permit # as applicable:	
☐ EQUIS			Date Results Requested:		Field Filtered (if applicable): ☐ Yes ☐ No	
☐ Other _____					Analysis:	

* Matrix Codes (Insert in Matrix box below): Drinking Water (DW), Ground Water (GW), Wastewater (WW), Product (P), Soil/Solid (SS), Oil (OL), Wipe (WP), Tissue (TS), Bioassay (B), Vapor (V), Surface Water (SW), Sediment (SED), Sludge (SL), Caulk (CK), Leachate (LL), Biosolid (BS), Other (OT)

[illegible]

Additional Instructions from Pace® :	Collected By: Printed Name Signature

Customer Remarks / Special Conditions / Possible Hazards:									

# Coolers:	Thermometer ID:	Correction Factor (°C):	Obs. Temp. (°C):	Corrected Temp. (°C):	[] On Ice

Relinquished by/Company: (Signature)	Date/Time:	Received by/Company: (Signature)	Date/Time:	Tracking Number: Delivered by: ☐ In- Person ☐ Courier ☐ FedEX ☐ UPS ☐ Other
Relinquished by/Company: (Signature)	Date/Time:	Received by/Company: (Signature)	Date/Time:	
Relinquished by/Company: (Signature)	Date/Time:	Received by/Company: (Signature)	Date/Time:	
Relinquished by/Company: (Signature)	Date/Time:	Received by/Company: (Signature)	Date/Time:	

reservation non-conformance identified

Chain of Custody Instructions

***Pace Location Requested:** City and State of Pace Laboratory testing is to be performed at.

***Company Name:** Client's company name

***Street Address:** Client's mailing address

***City, State, Zip:** Client's city, state and zip code for mailing

***Contact/ Report to:** Person to receive results

Customer Project # and Project Name: Client's reference to the project or work involved with these samples.

Site Collection Info/ Facility ID: Client's location of project

Time Zone: Check time zone of sample to ensure proper hold times are met.

Purchase Order #: Client specific number to be listed on project invoice for client billing purposes.

Invoice To: Client contact the project invoice needs to be emailed to.

Invoice Email: Email address that project invoice will need to be emailed to

***Phone #:** Client's contact phone number

E-mail: Client's e-mail for correspondence and final report

Regulatory Program: List the program that is guiding the work to ensure proper regulations are followed: DW, RCRA, etc.

Data Deliverable: Please select or enter required deliverables.

***County/State Origin of Samples:** Enter the county to ensure proper handling of regulated soils. State required to ensure proper reporting.

Field Filtered: Indicate if samples have been filtered in the field. If samples are required to be field filtered and filtering is not indicated, a qualifier will be added to all associated data.

***Customer Sample ID:** The unique sample ID you want to appear on the analytical report

***Collected Date:** Date sample was collected. For composite samples, please fill in both beginning and end date.

***Collected Time:** Time sample was collected. For composite samples, please fill in both beginning and end time.

***Comp/Grab:** Please denote "GRAB" if the sample was collected at one time from one specific location. Please denote "COMP" if the sample is a composite of samples collected at one or more times or locations and combined to make one sample.

***Matrix:** Select from list provided list. If prepopulated chain is provided for you matrix codes may vary.

***Number and Type of Containers:** Total number of containers per container type submitted for the samples

***Container Size:** Specify container size from list.

***Container Preservation Type:** Specify sample preservation from provided list.

***Analysis Requested:** Write the analysis name (or an abbreviation), the name of a group of tests, or the method number you would like us to perform. Examples are BOD, TCLP Metals, PCBs, Method 624, etc. Place a check mark in the small boxes that correspond to the sample(s) on which you want these tests performed.

Sample Comment: List any notes or important information about the individual sample here. Please identify in the sample comment if a sample should be used for MS/MSD.

Customer Remarks/Special Conditions/Possible Hazards: List special instructions about the sample here. If the sample is known or suspected to be hazardous indicate that here and attach SDS if possible. This space can also be used for listing additional analyses, or to request an extra copy of the report to be sent to an alternate person/address, etc.

Rush request: If faster than standard turnaround time results are needed. Circle one of the rush options and note the day the results are requested by. All rush requests require preapproval by the laboratory. Surcharges will apply for non- standard turnaround times. Results will be due by the end of business on the date due based on standard turnaround time unless other arrangements have been made with your Project Manager.

Summarized Sample Acceptance Policy Requirements:

- Proper, full and completed chain-of-custody documentation
- Readable unique sample container identification written in indelible ink
- Appropriate sample container
- Sufficient sample volume to perform requested tests
- Received within required holding time
- Received within temperature preservation requirements
- Sample containers received in good condition (not leaking or broken)
- Any custody seal intact
- Properly preserved
- No headspace in volatile water samples
- **Note:** When sample specific Quality Control is required (e.g. MS/MSD) please ensure necessary sample containers and sample volume is provided.

A data qualifier and/or case narrative will be added to the final test report when the above sample acceptance requirements are not met.

Location Specific Sample Acceptance Policy available from your Project Manager

***Collected By:** Printed name of sample collector

***Collected By Signature:** Signature of sample collector

***Relinquished By/Received By:** This form must be signed each time the sample(s) changes hands. Custody seals are available upon request if needed.

***Required field:** Failure to fill in a required field may result in a sample(s) being put on hold until information can be obtained. This may result in a delay in receiving results.

Appendix G

Laboratory SOPs

	Always check on-line for validity.	Level:  Standard Operating Procedure
	533 Determination of Selected PFAS in Drinking Water by Anion Exchange SPE and LC/MS/MS Using Isotope Dilution	
	Document number: LCMS-SOP30814	
Old Reference:		
Version: 4		Organisation level: 4-Business Unit
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS/6_Extract	Responsible: 6_QAD4

EUROFINS EATON ANALYTICAL, LLC
Standard Operating Procedure

EPA Method 533

Confidential

01) Title

01) Title

Determination of Selected PFAS in Drinking Water by Anion Exchange SPE and LC/MS/MS Using Isotope Dilution

Attachment:

533-30814-Monrovia r4..11.7.22.pdf (.pdf)

End of document

Version history

Version	Approval	Revision information	
2	24.FEB.2021		
3	21.JUN.2021		
4	18.JAN.2023		

LCMS-EPA 533 Determination of Selected PFAS in Drinking Water by Anion Exchange SPE and LC/MS/MS Using Isotope Dilution

- 1) SCOPE & APPLICATION
- 2) METHOD SUMMARY
- 3) DEFINITIONS
- 4) INTERFERENCES
- 5) PERSONNEL HEALTH & SAFETY
- 6) EQUIPMENT & SUPPLIES
- 7) REAGENTS & STANDARDS
- 8) SAMPLE COLLECTION, PRESERVATION & STORAGE
- 9) QUALITY CONTROL
- 10) PREVENTIVE MAINTENANCE & TROUBLESHOOTING
- 11) CALIBRATION & STANDARDIZATION
- 12) PROCEDURE
- 13) DATA PROCESSING, DATA EVALUATION, & CALCULATIONS
- 14) METHOD PERFORMANCE
- 15) SUMMARY OF METHOD
- 16) WASTE MANAGEMENT
- 17) REFERENCES
- 18) QC TABLE
- 19) REVISIONS

1) SCOPE & APPLICATION

1.1 This is a Solid Phase Extraction (SPE) liquid chromatography/electrospray ionization/tandem mass spectrometry (LC/ESI/MS/MS) method applicable to the determination of selected per- and polyfluorinated alkyl substances (PFAS) in drinking water, groundwater, surface water, bottled water, and treated drinking water sources.

Table 1. EPA Method 533 List of Analytes

Analyte	Acronym	CAS #	MRL, ng/L
9 Perfluoroalkylcarboxylic Acids			
Perfluorobutanoic acid ^a	PFBA	375-22-4	2
Perfluoropentanoic acid ^a	PFPeA	2706-90-3	2
Perfluorohexanoic acid ^a	PFHxA	307-24-4	2
Perfluoroheptanoic acid ^a	PFHpA	375-85-9	2
Perfluorooctanoic acid ^a	PFOA	335-67-1	2
Perfluorononanoic acid ^a	PFNA	375-95-1	2
Perfluorodecanoic acid ^a	PFDA	335-76-2	2
Perfluoroundecanoic acid ^a	PFUnA	2058-94-8	2
Perfluorododecanoic acid ^a	PFDaA	307-55-1	2
6 Perfluoroalkylsulfonic Acids			
Perfluorobutanesulfonic acid ^a	PFBS	375-73-5	2
Perfluoropentanesulfonic acid ^b	PFPeS	2706-91-4	2
Perfluorohexanesulfonic acid ^a	PFHxS	355-46-4	2
Perfluoroheptanesulfonic acid ^b	PFHpS	375-92-8	2

Perfluorooctanesulfonic acid ^a	PFOS	1763-23-1	2
Perfluoro(2-ethoxyethane) sulfonic acid	PFEESA	113507-82-7	2
3 Fluorotelomer Sulfonic Acids			
1H,1H,2H,2H-Perfluorohexane sulfonic acid/4:2 Fluorotelomer sulfonic acid ^a	4:2 FTS	757124-72-4	2
1H,1H,2H,2H-Perfluorooctane sulfonic acid/6:2 Fluorotelomer sulfonic acid ^a	6:2 FTS	27619-97-2	2
1H,1H,2H,2H-Perfluorodecane sulfonic acid/8:2 Fluorotelomer sulfonic acid ^a	8:2 FTS	39108-34-4	2
7 Perfluoroalkyl Ether Carboxylic Acids and Others			
Hexafluoropropylene oxide dimer acid ^a	HFPO-DA/GenX	13252-13-6	2
4,8-Dioxa-3H-perfluorononanoic acid ^b	ADONA	919005-14-4	2
9-Chlorohexadecafluoro-3-oxanone-1-sulfonic acid ^b	9Cl-PF3ONS/ F-53B major	756426-58-1	2
11-Chloroeicosafluoro-3-oxaundecane-1-sulfonic acid ^b	11Cl-PF3OUdS/ F-53B minor	763051-92-9	2
Perfluoro-4-methoxybutanoic acid ^b	PFMBA/PFMOBA	863090-89-5	2
Perfluoro-3-methoxypropanoic acid ^b	PFMPA/PFMOPrA	377-73-1	2
Nonafluoro-3,6-dioxaheptanoic acid ^b	NFDHA/PFMOEOAA	151772-58-6	2

Notes: ^a: 16 PFAS analyzed by using an isotope dilution method. ^b: 9 PFAS analyzed by using an internal standard method.

4:2 FTS also called 4:2 Fluorotelomer sulfonic acid, 6:2 FTS also called 6:2 Fluorotelomer sulfonic acid, 8:2 FTS also called 8:2 Fluorotelomer sulfonic acid, NFDHA also called Perfluoro-2-methoxyethoxyacetic acid (PFMOEOAA), HFPO-DA/GenX (acid form) also called perfluoro-2-propoxypropanoic acid (PFPrOPrA), and ADONA (acid form) also called dodecafluoro-3H-4,8-dioxanonanoic acid.

9Cl-PF3ONS/F-53B major is also available as K salt form (CAS No. 73606-19-6). 11Cl-PF3OUdS/F-53B minor is also available as K salt (CAS No. 83329-89-9). ADONA is available as ammonium salt (CAS No. 958445-44-8) and Na salt (no CAS No.).

1.2 Applicable concentration ranges are 2-80 ng/L.

1.3 Analysts performing this method must have a clear understanding of solid phase extraction (SPE) and LC/ESI/MS/MS principles and also have demonstrated experience in the use of LC/ESI/MS/MS.

2) SUMMARY OF METHOD

2.1 A 250 mL water sample fortified with isotopically labeled analogues of the method analytes is extracted through a weak anion/mixed-mode polymeric solid phase extraction (SPE) cartridge containing weak anion, mixed-mode polymeric sorbent with a styrenedivinylbenzene (SDVB) backbone and a di-amino ligand. The cartridge is rinsed with sequential washes of aqueous ammonium acetate and methanol. Then the analytes and their isotope dilution analogues are eluted from the solid phase sorbent with a small quantity of methanol containing ammonium hydroxide. The extract is then evaporated to dryness with nitrogen and heated water bath at less than 60°C, reconstituted with 20% water in methanol (v/v), spiked with the Isotope Performance Standards (IPS), and then adjusted to 1.0 mL with 20% water in methanol.

2.2 An aliquot of the sample extract is injected onto a C18 column. The analytes, isotope dilution analogues, and internal standards are separated by using a gradient composition of methanol and 20 mM ammonium acetate in reagent water as the mobile phase. After elution from the column, the analytes are detected by ESI/MS/MS in a multiple reaction monitoring (MRM) mode. In the negative ESI process, a deprotonated molecular ion (M-H) is typically formed. In the MS/MS process, these precursor ions then form product ions which are used for quantitation.

2.3 A minimum of five points are used to generate a linear or 6 points for quadratic calibration curves for the analytes using peak area and an isotope dilution technique. The calibration function is forced thru the origin. IPS and IDA/SS are calibrated using Average Response factor as these are added at a single concentration level to the calibration standards. The IDA/SS uses an internal standard technique. The IPS/IS uses an external calibration technique.

2.4 Routine quality assurance is performed by analyzing IPS/IS, IDA/SS, Continuing Calibration Checks (CCC), Laboratory Reagent Blanks (LRB/MBLK), Laboratory Fortified Blanks (LFB/LCS), Laboratory Fortified Sample Matrix (LFSM/MS), Laboratory Fortified Sample Matrix Duplicate (LFSMD/MSD), Field Duplicates (FD/DUP), Field Reagent Blank (FRB)), and external Quality Control Standard samples (QCS).

3) DEFINITIONS

3.1 Refer to the W126472, Glossary for a complete list of terms and definitions.

3.2 Isotope Dilution Analogues (IDA)/Surrogate Standards (SS) – Isotopically labeled analogues of the method analytes that are added to the sample prior to extraction in a known amount. Note: Not all target PFAS currently have an isotopically labelled analogue. In these cases, an alternate isotopically labelled analogue is used as recommended in the Table 5 of the method.

3.3 Isotope Performance Standards (IPS)/Internal Standards (IS) – Quality control compounds that are added to all standard solutions and extracts in a known amount and used to measure the relative response of the isotopically labelled analogues that are components of the same solution. For this method, the isotope performance standards are three isotopically labeled analogues of the method analytes. The isotope performance standards are indicators of instrument performance and are used to calculate the recovery of the isotope dilution analogues through the extraction procedure. In this method, the isotope performance standards are not used in the calculation of the recovery of the native analytes.

4) INTERFERENCES

4.1 Method interferences may be caused by multiple sources. All sources or items must be routinely demonstrated to be free from the interferences. An LRB can provide information regarding the presence or absence of such interferences and needs to be less than 1/3 the MRL for the method analytes. **Note:** Subtracting blank values from sample results is not permitted.

4.2 Method interferences may be caused by contaminants in solvents, reagents (including reagent water), sample bottles, autosampler vials, and other sample processing hardware.

4.2.1 LCMS grade methanol are proven to be satisfactory for this method.

4.2.2 Ammonium acetate used as a preservative potentially contains trace-level organic contaminants including PFAS.

4.2.3 Care should be taken when using gastight syringes for this method because Teflon may cause leaching PFAS contamination to samples and standards.

4.3 The SPE cartridges may be a source of interferences. Variations from lot to lot and from brand to brand may be significant to preclude analyte extraction, identification, and quantitation. Therefore, brands and lots of SPE cartridges must be recorded tested prior to use on samples to ensure that contamination does not preclude analyte identification and quantitation. A method blank (MBLK) and MRL check must be extracted and analyzed to demonstrate the SPE cartridge is free of contamination

4.4 The SPE system such as a vacuum manifold or an AutoTrace unit may be a source of interferences. All items such as these must routinely be demonstrated to be free from interferences. In order to minimize PFAS carryover on the system during extraction, the system is fully flushed with at least 50 mL of methanol before initiating a new extraction batch.

4.5 Method interferences may also be caused by contaminants: 1) in the liquid delivery system (including the solvent and in-line filters, the needle and seal pack, the plunger and seal wash, and the sample loop) and 2) in the ion generation and sampling system (including the ESI probe capillary, the sample cone, the extraction cone, and the ion block). All items such as these must routinely be demonstrated to be free from interferences. In order to minimize PFAS buildup on the system during mobile phase equilibration, the system may be flushed with 100% methanol for at least 10 minutes before initiating a sequence.

4.5.1 Carryover peaks from one analysis will affect the proper detection of analytes in a subsequent analysis. Carryover peaks may result from a sample containing high concentrations of analytes. An adequate amount of rinsing solvents for needle wash and seal pack wash are pumped through the system to clean the seal pack, needle, and sample loop. However, the analyst must carefully review data from samples analyzed immediately after high concentration samples. Also, after analysis of a sample containing high concentrations of analytes, an instrument blank can be run to ensure that accurate values are obtained for next sample.

4.6 Method interferences may also be caused by contaminants that are co-extracted from the sample. The extent of matrix interferences will vary considerably from source to source, depending upon the nature of the water. High levels of humic and/or fulvic material that is co-extracted by this method can cause enhancement and/or suppression in the ESI source. For this method, total organic carbon (TOC) is a good indicator of humic content of the sample.

4.7 The method involves the extraction and concentration of trace levels of PFAS. As such, PFAS standards, extracts and samples must not come in contact with any glass containers or pipettes as these analytes can potentially adsorb to glass surfaces. PFAS analytes, IPS

and IDA standards commercially purchased in glass ampoules are acceptable; however, all subsequent transfers or dilutions performed by the analyst must be prepared and stored in polypropylene containers.

4.8 The mass-labeled standards used as IPS and IDA may contain a small percentage of the corresponding native analyte or different mass-labeled analogs. The labeled standards must meet the purity requirements stated in the low system background requirement in Section 14.

4.8.1 A trace amount of $^{13}\text{C}_3$ -PFBA may be present in $^{13}\text{C}_4$ -PFBA. Natural abundance (1.1%) of ^{13}C present in $^{13}\text{C}_3$ -PFBA may empirically contribute to the peak areas of $^{13}\text{C}_4$ -PFBA.

4.8.2 Natural abundance (4.25%) of ^{34}S present in the corresponding native analyte may empirically contribute to the peak areas of M+2 ions from $^{13}\text{C}_2$ -labeled telomere sulfonates (4:2 FTS, 6:2 FTS, and 8:2 FTS).

4.9 Inorganic Salts – Acceptable performance defined as recovery of the isotope dilution analogues between 50–200% was confirmed for matrix ion concentrations up to 250 mg/L chloride, 250 mg/L sulfate, and 340 mg/L hardness measured as CaCO_3 . Higher matrix ion concentrations may result in unacceptable performance.

5) PERSONNEL HEALTH & SAFETY

5.1 Toxicity or carcinogenicity of each reagent used in this method has not been precisely defined. Each chemical should be treated as a potential health hazard, and exposure to these chemicals should be minimized. Consult the OSHA regulations and the SDS for all analytes, IDA, and IS for proper handling procedures. SDS are available to laboratory personnel at <https://msdsmanagement.msdsolnline.com/5c1df5b3-747d-4789-8104-42457dc3a3e5/ebinder/?nas=True>.

5.2 PFOA has been described as “likely to be carcinogenic to humans.” Pure standard materials and stock standard solutions of these method analytes should be handled with suitable protection to skin.

5.3 Wear eye protection and gloves when working with these chemicals.

5.4 Refer to the SOP “Hazardous Waste Management and Sample Disposal Procedures,” Chemical Hygiene Plan and OSHA Standard 29 CFR 1910.1450 Occupational Exposure to Hazardous Chemicals in Laboratories; Final Rule for additional safety information.

6) EQUIPMENT & SUPPLIES

UNCONTROLLED

7) REAGENTS & STANDARDS

UNCONTROLLED

UNCONTROLLED

UNCONTROLLED

8) SAMPLE COLLECTION, PRESERVATION & STORAGE

8.1 Sample Collection and Preservation – Nitrile gloves must be used when preserving bottles at the laboratory and also for collecting samples. Samples are collected in three 250 mL pre-preserved with ammonium acetate polypropylene bottles (actual volume = 280 mL to the neck) fitted with polypropylene screw caps. Alternatively, prior to shipping, 0.28 g (0.25-0.3 g) solid ammonium acetate (1 g/L) must be added to each sample container

8.1.1 The sample handler must wash their hands before sampling and wear nitrile gloves while filling and sealing the sample bottles.

8.1.2 Subsampling from a single bottle is not permitted except exceeding the calibration range as described in Section 12.5.

8.1.3 A Field Reagent Blank (FRB) sample must be handled along with each sample set. The sample set is composed of samples collected from the same sample site and at the same time. A FRB containing the reagent water must be shipped to the sampling site along with the sample bottles. For each FRB shipped, an empty sample bottle (with preservatives) must also be shipped. At the sampling site, the sampler must open the shipped FRB and pour the pre-analyzed reagent water into the empty shipped preserved sample bottle, seal and label this bottle as the FRB. The FRB is shipped back to the laboratory along with the samples and analyzed to ensure that PFAS are not introduced into the sample during sample collection/handling.

8.1.3.1 The same lots of the preservative and sample bottles must be used for the FRBs as for the field samples.

8.1.3.2 Reagent water used for the FRBs must be analyzed prior to shipment to ensure the water has minimal residual PFAS. Extract and analyze an LRB prepared with reagent water

using the same lots of the preservative and sample bottles destined for shipment to the sampling site. Ensure that analyte concentrations are $< 1/3$ the MRL.

8.2 Sample Shipment and Storage – Valid samples must be chilled on ice in a cooler during shipment, received within 2 days of collection, and below 10 °C at the laboratory. Samples must be stored at or below 6 °C and protected from light until extraction. If samples are received more than 2 days after collection, samples must not exceed 6 °C. Verify sample temperature upon receipt. Check residual chlorine within 3 days upon receipt.

8.3 Sample Holding Time – The maximum holding time for samples to be extracted is 28 days from collection.

8.4 Extract Holding Time – The maximum holding time for sample extracts is 28 days after sample extraction. Store refrigerated at $4 \pm 2^\circ\text{C}$. Bring samples to room temperature prior to analysis.

9) QUALITY CONTROL

9.1 The Quality Control Requirements of this method are presented in table form in Attachment V. Both Extraction Batch and Analysis Batch size are limited to 20 field samples.

9.2 Before analyzing any samples, the laboratory must meet the requirements of the Initial Demonstration of Capability (IDC) found in Section 14.

9.3 Isotope Performance Standards (IPS) – Add the IPS to all the calibration, QC, and field samples prior to analysis. For calibration standards, continuing calibration check, QC and field samples, the IPS response (as indicated by peak area) must be within 50-150% of the average IS area measured during the initial calibration.

9.3.1 If the criteria are not met, all data for the analytes must be considered invalid for all associated samples in the analysis batch. Corrective actions must be taken as described in Section 10.6. The analyst must reanalyze the sample as part of the same or a new analysis batch after the problems have been resolved or report the data with appropriate qualifications.

9.3.2 If the reanalysis meets the IS recovery criterion, report only the reanalysis data.

9.3.3 If the reanalysis does not meet the IPS recovery criteria, reextraction must be performed if the samples are still within the holding time or the data must be qualified. If reextraction is not possible, the ASM must be contacted in order to resolve the issue with the client.

9.4 Isotope Dilution Analogues (IDA) – Add the IDA to all the calibration, QC, and field samples prior to extraction or analysis. Calculate the percent recovery using the equation in Section 13.1. The IDA recovery in QC and field samples must be within 50-200% of the true value. IDA recovery in the calibration standards, CCCs, and QCS must be within 70-130% of the true value

9.4.1 If the IDA criteria are not met, all data for the analytes must be considered invalid for all associated samples in the analysis batch. Corrective actions must be taken as described

in Section 10.6. The analyst must reanalyze the sample as part of the same or a new analysis batch after the problems have been resolved.

9.4.2 If the reanalysis meets the IDA criteria, report only the reanalysis data.

9.4.3 If the reanalysis does not meet the IDA criteria, reextraction must be performed if the samples are still within the holding time. If reextraction is not possible, the ASM must be contacted in order to resolve the issue with the client.

9.5 Unextracted Quality Control Sample (QCS) (ICV, etc in TALS) – At a minimum, a QCS at mid-level calibration concentration must be run as part of the IDC, and at least quarterly. Calculate the percent recovery using the equation in Section 13.1. The QCS percent recovery for each analyte must be within 70-130% of the true value.

9.5.1 If the criteria are not met, then all calibration, QC, and field sample data must be considered invalid. Corrective actions must be taken as described in Section 10.6. The analyst must reanalyze all the calibration, QC, and field samples after the problems have been resolved.

9.6 Continuing Calibration Checks (CCCs)(CCV,CCCL, CCVIS, etc in TALS) – A low-level CCC (CCL) at 0.5 µg/L must be run immediately after initial calibration curve, prior to running the QCS, any QC and field samples. Subsequent CCCs must be run after every ten field samples and at the end of each analysis batch, and is rotated between a mid-level CCC (CCM) at 7.5µg/L and a high-level CCC (CCH) at 15µg/L. If ten or fewer samples are analyzed, the ending CCC is rotated between CCM and CCH with each batch. Calculate the percent recovery using the equation in Section 13.1. CCL recoveries must be within 50-150% of the true value, CCM and CCH recoveries must be within 70-130% of the true value for each analyte. Mathematically, for 0.5µg/L to 20µg/L calibration range, the midpoint concentration is 10µg/L. UCMR 4 defines midpoint as ±20% of mathematical midpoint. In this case, ±20% of 10µg/L is 8µg/L to 12µg/L. For ease in standard preparation, CCM was assigned a concentration of 7.5µg/L.

9.6.1 If the CCC criteria are not met, then all data since the last passing CCC must be considered invalid. Corrective actions must be taken as described in Section 10.6. Any field samples analyzed since the last acceptable CCC must be reanalyzed as part of the same or a new analysis batch after the problems have been resolved with the following exception. For a particular target analyte, if the CCC fails because the recoveries are greater than the upper recovery limits, and field samples show no detection, non-detects may be reported without reanalysis. **Note:** there may be further requirements from ASMs for flagged data, however this is not a concern for operations staff.

9.7 Laboratory Reagent Blanks (LRB)(MB, MBL, etc in TALS) – Include at least one LRB with each extraction batch including a set of up to 20 samples. The target analytes must be less than 1/3 the MRL. Any time a new lot of SPE cartridges is received, an LRB must be performed. If a target analyte is detected in the LRB at concentrations greater than or equal to 1/3 the MRL, then all data for the analyte must be considered invalid for all associated samples in the extraction batch. Corrective actions must be taken as described in Section 10.6. The analyst must reanalyze the LRB as part of the same or a new analysis batch after the problems have been resolved. **Note:** Subtracting blank values from sample results is not permitted.

9.7.1 If the reanalysis meets the LRB acceptance criteria, report only the reanalysis data.

9.7.2 If the reanalysis does not meet the LRB criteria, re-extraction of all samples with results above MRL must be performed. If re-extraction is not possible, the ASM must be contacted in order to resolve the issue with the client.

9.8 Laboratory Fortified Blank (LFB/LCS) – Include one low level (2 ng/L) LFB/LCS with each extraction batch including a set of up to 20 samples. Then, mid (30 ng/L) and high (60 ng/L) LFB/LCS levels must be rotated between extraction batches. This is illustrated in the following table:

Batch 1: LFB low/MRL Check and LFB/LCS-Mid
Batch 2: LFB low/MRL Check and LFB/LCS-High
Batch 3: LFB low/MRL Check and LFB/LCS-Mid
Batch 4: LFB/LCS-low and LFB/LCS-High
Batch 5: LFB/LCS-Mid and LFB/LCS-Mid
Batch 6: LFB/LCS-High and LFB/LCS-High
etc.

9.8.1 Calculate the percent recovery using the equation in Section 13.1. The LFB/LCS percent recovery for each analyte must be within 50-150% of the true value for the low level, and 70-130% of the true value for the medium and high levels.

9.8.2 If the criteria are not met for the LFB/LCS, corrective actions must be taken. LFB/LCS2 if analyzed immediately after the LFB/LCS may serve as the corrective action. Additional corrective actions are described in section 10.6. The analyst must reanalyze the LFB as part of the same or a new analysis batch after the problems have been resolved.

9.8.3 If the reanalysis meets the LFB/LCS criteria, report only the reanalysis data.

9.8.4 If the reanalysis does not meet the LFB criteria and LFB/LCS2 failed to meet criteria, re-extraction of the entire extraction batch must be performed. If re-extraction is not possible, the ASM must be contacted in order to resolve the issue with the client to resample.

9.8.5 Any time a new lot of SPE cartridges is received, an LRB and low level LFB/MRL Check must be performed. The target analytes must be less than 1/3 the MRL in the LRB. Calculate the percent recovery in the low level LFB/MRL Check using the equation in Section 13.1. The low level LFB/MRL Check percent recovery for each analyte must be within 50-150% of the true value.

9.10 Laboratory Fortified Sample Matrix (LFSM/MS) – Include at least one LFSM/MS with each extraction batch including a set of up to 20 samples. The fortification concentrations must be alternated between a low, mid, and high-level concentration. The low-level (LFSM/MS-Low) fortification concentration must be at or below the MRL for each analyte. The mid-level (LFSM/MS-Mid) fortification concentration must be near the mid-level calibration standard for each analyte. The high-level (LFSM/MS-High) fortification concentration must be near the high-level calibration standard for each analyte. LFSM/MS-Low is at 2 ng/L, LFSM/MS-Mid is at 30 ng/L, and LFSM/MS-High is at 60 ng/L.

9.10.1 Calculate the percent recovery using the equation in Section 13.2. For fortification levels at the MRL, the percent recovery must be within 50-150% of the true value. Recoveries for samples fortified at all other concentrations must be within 70-130% of the true value.

9.10.2 If the results are outside of the designated recovery range, and the recovery for that analyte is shown to be in control in the CCCs and LFB/LCSs, the recovery problem for the LFSM/MS is judged to be matrix biased. The result for that analyte in the unfortified sample is labeled suspect based on poor recoveries in the matrix spike to inform the data user that the results are suspect.

9.11 Laboratory Fortified Sample Matrix Duplicate (LFSMD/MSD) or Field Duplicate (FD/DUP) – Include at least one LFSMD/MSD or FD/DUP with each extraction batch including a set of up to 20 samples. Like LFSM/MS, the fortification concentrations must be alternated between a low, mid, and high-level concentration. Use the same LFSM/MS fortification concentrations and recovery criteria. An extraction batch that contains an LFSMD/MSD does not require a FD/DUP.

9.11.1 Calculate the percent recovery using the equation at Section 13.2 to calculate percent recovery. For analyte concentrations within two times the MRL, the percent recovery must be within 50-150% of the true value. Recoveries for all other concentrations must be within 70-130% of the true value.

9.11.2 If the results are outside of the designated recovery range, and the recovery for that analyte is shown to be in control in the CCCs and LFB/LCSs, the recovery problem for the LFSMD/MSD is judged to be matrix biased. The result for that analyte in the unfortified sample is labeled suspect based on poor recoveries in the matrix spike to inform the data user that the results are suspect.

9.11.3 Calculate the relative percent difference (RPD) for sample duplicate measurements using the equation in Section 13.3. For analyte concentrations within a factor of two times the MRL, the RPDs must be less than or equal to 50%. For analyte concentrations greater than two times the MRL, the RPDs must be less than or equal to 30%. If the RPD falls outside the designated range, and the laboratory performance for that analyte is shown to be in control in the CCCs and LFB/LCSs, the recovery is judged matrix biased. The results for the sample are labeled as suspect based on poor RPD values to inform the data user that the results are suspect. For Arizona samples, if RPD failed, then run LFB/LFBD (LCS/LCSD) to obtain precision and accuracy

9.12 Field Reagent Blank (FRB) – Analysis of the FRB is required only if an associated Field Sample contains an analyte greater than or equal to the MRL. The FRB target analytes must be less than or equal to 1/3 the MRL. If a target analyte is detected in the FRB at concentrations greater than 1/3 the MRL, verify that the source of the contamination is due to sample collection/handling or carryover of samples containing high concentrations of analytes. Then reanalyze the FRB in a separate analytical batch or in the same analytical batch following an LIB, must either confirm or not confirm the analyte's presence in the FRB. If the reanalysis passes the FRB requirement, all samples collected with the passing FRB are valid. If the reanalysis still fails the FRB requirement, all samples with results greater than MRL collected with the failed FRB are invalid, recollected, and reanalyzed, and the ASM must be contacted in order to resolve the issue with the client.

9.13 Evaluation of Background When Analytes Exceed the Calibration Range – After analysis of a sample in which method analytes exceed the calibration range, one or more LRBs must be analyzed (to detect potential carryover) until the system meets the LRB acceptance criteria. If this occurs during an automated sequence, examine the results of samples analyzed following the sample that exceeded the calibration range. If the analytes that exceeded the calibration range in the previous sample are detected at, or above, the

MRL, these samples are invalid. If the affected analytes do not exceed the MRL, these subsequent samples may be reported.

9.14 Lab Instrument Blank (LIB) Optional – Analysis of the LIB may be done at the beginning of the run and after the highest calibration point to demonstrate that there is no carryover. If analyzed, the target analytes must be less than 1/3 the MRL. If a target analyte is detected in the LIB at concentrations greater than or equal to 1/3 the MRL, Analysis run must be repeated.

9.15 Qualitative/Semi-Quantitative PFOA Mixed Standard – Analyze a technical grade PFOA at a mid to high concentration as a retention time check prior to running initial demonstration of capability (IDC). Repeat anytime chromatographic changes occur that alter analyte retention time. Both the linear and branched isomers must be within the acquisition window. This standard is not used for quantitation, but for identification of the branched and linear isomers for integration of QC and field samples.

9.16 QC exception reports are generated for failing QC, and QC tracking will be performed at that time.

10) PREVENTIVE MAINTENANCE & TROUBLESHOOTING

10.1 Mechanical Pump Ballasting – At least once per week, ballast the mechanical pump by opening (turn counterclockwise to full open) the gas ballast knob on top of the pump. Leave the knob in this position for 15-20 min, then close tightly. This allows oil in the pump filter reservoir to drain back into the pump body. **Note:** DO NOT perform any data acquisitions during this time. Also, do not ballast the pump for longer than 30 minutes.

10.2 Mechanical Pump Oil Check and Change – Periodically check the oil level in the roughing pump when performing ballasting. The oil should be relatively clear and pale yellow in color and in the working range. Change the pump oil as needed using high grade Edwards or Alcatel pump oil.

10.3 Curtain plate Cleaning – It may become necessary to clean the curtain plate (the outer plate which is at atmospheric pressure). The cone needs to be cleaned when sensitivity drops or there appears to be a lot of noise in the baseline.

10.3.1 Gently clean the curtain plate with aluminum oxide grit. Rinse with reagent water then methanol. Dry with nitrogen gas and re-install. **IMPORTANT:** Do not set the conical part side down as this may damage the orifice.

10.4 Purging the Pump – The LC pumps need to be purged whenever eluents are replaced. Open the purge valve and select "Purge" on the keypad.

10.5 Guard Column Replacement – Replace the guard column as necessary. Particulates resulting from SPE sorbent breakthrough and the mobile phase may easily clog the guard column and the analytical column.

10.6 Corrective Action – Preparation and reanalysis must be performed after problems have been resolved. In order to locate problems, analysts should check:

- 1) calculations to locate possible errors,
- 2) contamination,
- 3) standard solutions for degradation, and

4) instrument performance problems.

Corrective actions may include:

- 1) reprepare all samples,
- 2) prepare and use new stock and substock standard solutions,
- 3) prepare and use a new mobile phase,
- 4) install a new analytical column,
- 5) retune the instrument to optimize the performance, and
- 6) check and perform instrument maintenance.

10.6.1 ESI/MS instrument maintenance may include:

- 1) clean and repair the ESI probe,
- 2) clean the ion source and sampling cone,
- 3) perform mass calibration and retune the instrument,
- 4) clean hexapole and quadrupole,
- 5) change pump oil, and
- 6) other maintenances.

10.6.2 LC instrument maintenance may include:

- 1) prime seal wash and needle wash,
- 2) perform compression check and adjust seals,
- 3) purge the sample management system,
- 4) replace solvent inlet filters and the in-line filter element,
- 5) replace needle and seal pack,
- 6) replace pump check valves,
- 7) maintenance of LC pump components, and
- 8) other maintenances.

11) CALIBRATION & STANDARDIZATION

12) PROCEDURE

13) DATA PROCESSING, DATA EVALUATION, & CALCULATIONS

13.1 Calculation of Percent Recovery - Use the following equation to calculate percent recovery:

$$\% R = \left(\frac{A}{B} \right) \times 100$$

where:

A = measured concentration in the fortified sample

B = fortification concentration.

13.2 Calculation of Percent Recovery for Lab Fortified Sample Matrices:

$$\% R = \frac{(A - B)}{C} \times 100$$

where:

A = measured concentration in the fortified sample,

B = measured concentration in the unfortified sample at or above the MRL.

C = fortification concentration.

13.3 Calculation of Relative Percent Difference (RPD):

13.3.1 For duplicate analysis of field sample:

$$RPD = \frac{|FD1 - FD2|}{(FD1 + FD2) \div 2} \times 100$$

13.3.2 For duplicate analysis of lab fortified matrices:

$$RPD = \frac{|LFSM - LFSMD|}{(LFSM + LFSMD) \div 2} \times 100$$

13.4. Because environmental samples may contain both branched and linear isomers for method analytes, but quantitative standards that contain the linear and branched isomers do not exist for all method analytes, integration and quantitation of the PFAS is dependent on type of standard available for each PFAS. It is recognized that some of the procedures described below for integration of standards, QC samples and Field Samples may cause a small amount of unavoidable bias in the quantitation of the method analytes due to the current state of the commercially available standards.

13.4.1 Multiple chromatographic peaks may be observed for standards of PFHxS and PFOS due to chromatographic resolution of the linear and branched isomers of these compounds. All the chromatographic peaks observed in these standards must be integrated and the areas summed. Chromatographic peaks in all Field Samples and QC samples must be integrated in the same way as the calibration standards for analytes with quantitative standards containing the branched and linear isomers.

13.4.2 For PFOA, identify the branched isomers by analyzing a qualitative standard that includes both linear and branched isomers and compare retention times and tandem mass spectrometry transitions. Quantitate Field Samples and QC samples by integrating the total response (i.e., accounting for peaks that are identified as linear and branched isomers) and relying on the initial calibration with a linear-isomer quantitative PFOA standard.

13.4.3 If standards containing the branched and linear isomers cannot be purchased (i.e., only linear isomer is available), only the linear isomer can be identified and quantitated in Field Samples and QC samples using the linear standard because the retention time of the branched isomers cannot be confirmed.

13.4 Process the QC and samples using the appropriate quantitation method. Update the retention times of the analytes and make any adjustments to the peak detection parameters if necessary. If changes are made, reprocess the samples before continuing.

13.5 Review data file for any incorrect peak identification and integration. Manual integrations are performed when the retention time or baseline designated by the software is incorrect.

13.6 Save the data and print out reports as noted below. Export the data to the LIMS and report the results through Data Entry.

14) METHOD PERFORMANCE

An acceptable analyte calibration must be demonstrated and documented before performing the IDC.

14.1 Initial Demonstration of Capability (IDC) – All requirements of the IDC must be successfully performed prior to analyzing any Field Samples and for each analyst performing this method.

14.1.1 Initial Demonstration of Branched vs. Linear Isomer Profile for PFOA in a Qualitative Standard – Prepare and analyze a mid to high concentration technical grade PFOA and identify retention times of branch isomers of PFOA. This qualitative standard is NOT used for quantitation, but for identification of the branched and linear isomers for integration of QC and field samples. Repeat anytime chromatographic changes occur that alter analyte retention time.

14.1.2 Initial Demonstration of Low System Background – Prepare, extract and analyze an LRB immediately after injecting the highest calibration standard. If an automated SPE system is used, an LRB must be extracted on each port to fulfill this requirement. The LRBs are rotated among the ports to ensure that all the valves and tubing meet the LRB requirements. The target analytes for the LRB must be less than 1/3 the MRL.

14.1.3 Initial Demonstration of Accuracy – Prepare, extract, and analyze 7 replicate LFBs (containing sample preservatives) fortified at 30 ng/L. The acceptable average recovery is 70-130% for all target analytes.

14.1.4 Initial Demonstration of Precision – Using the same set of replicate data generated for Section 14.1.3, calculate the relative standard deviation (RSD). The RSD must be less than 20% for all target analytes.

14.1.5 Minimum Reporting Limit Confirmation (PIR Study)– This procedure must be completed initially on each instrument performing this method. PIR is required for new instrument and new analyst.

14.1.5.1 Prepare, extract, and analyze 7 replicate LFBs at the estimated MRL. Determine the Half Range for the Prediction Interval of Results (HRPIR) using the equation below:

$$HR_{PIR} = 3.963 * SD$$

where:

SD = the standard deviation, 3.963 is a constant value for seven replicates.

14.1.5.2 Confirm that the upper and lower limits for the Prediction Interval of Results (PIR = Mean $\pm$ HR_{PIR}) meet the upper and lower recovery limits as shown below

The Upper PIR Limit must be $\leq$ 150%.

$$\frac{\text{Mean} + \text{HR}_{\text{PIR}}}{\text{Fortified Concentration}} \times 100 \leq 150\%$$

The Lower PIR Limit must be $\geq 50\%$.

$$\frac{\text{Mean} - \text{HR}_{\text{PIR}}}{\text{Fortified Concentration}} \times 100 \geq 50\%$$

14.1.5.3 The MRL is validated if both the Upper and Lower PIR Limits meet the criteria. If these criteria are not met, the MRL has been set too low and must be determined again at a higher concentration.

14.1.6 Method Detection Limit (MDL) – The MDL is not required by this method, however state certification may require MDL study. If annual MDL studies are needed, follow the detailed instructions in Method Detection Limit (MDL) Procedure, Document number W125066. Data should be entered into the MDL Study spread sheet. The method detection limit (MDL) procedure and calculation is based on EPA's procedure "Definition and Procedure for the Determination of the Method Detection Limit, Revision 2".

14.2 Continuing DOC must be performed on an annual basis according to form 06-QA-F0400.

15) POLLUTION PREVENTION

15.1 This method utilizes SPE to extract analytes from water. It requires the use of very small volumes of organic solvent and very small quantities of pure analytes, thereby minimizing the potential hazards to both the analyst and the environment as compared to the use of large volumes of organic solvents in conventional liquid-liquid extractions.

15.2 For information about pollution prevention that may be applicable to laboratory operations, consult "Less is Better: Laboratory Chemical Management for Waste Reduction" available from the American Chemical Society's Department of Government Relations and Science Policy, 1155 16th Street N.W., Washington, D.C., 20036.

16) WASTE MANAGEMENT

16.1 It is the responsibility of the laboratory to determine whether its wastes are hazardous and to assure safe handling and disposal. The laboratory works closely with the Treatment, Storage, and Disposal Facility to ensure that certain wastes are recycled where possible, that the source of waste is reduced to the lowest possible level and that stringent land disposal restrictions are followed.

16.2 Refer to the following documents for additional information regarding waste management:

16.2.1 Hazardous Waste Management and Sample Disposal Procedures

16.2.2 Resource Conservation and Recovery Act (RCRA)-Title 40 of the Code of Federal Regulations, Parts 260 through 270 (40 CFR 260-270)

16.2.3 California Hazardous Waste Control Law (HWCL)-CCR Title 22 where 40 CFR was duplicated into CCR Title 22, Parts 66260-66270.

17) REFERENCES

17.1 U.S. Environmental Protection Agency (USEPA) Method 533 Determination of Per- and Polyfluoroalkyl Substances in Drinking Water by Isotope Dilution Anion Exchange Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry, November, 2019, EPA Document No. 815-B-19-020

18) QC TABLE

See Attachment VI

19) Revision History

v2.0

Y7BM – 01/2021

1. Updated GenX MRL to 2ng/L
2. Changed Methanol chemical grade from P&T grade to LCMS grade
3. Removed Argon from Section 6) Equipment and Supplies
4. Updated part numbers Section 6) Equipment and Supplies
5. Updated standard preparation in Section 7.11 thru 7.14
6. Added minimum scan requirement for each peak in section 11.2.4. A minimum of 10 scans across the chromatographic peak is needed to ensure adequate precision
7. Updated extraction Procedure in section 12.2.4 adding LFB/LCS—Mid/High. Corrected typo error on "Field"
8. Added Revision History in section 19
9. Updated Sections 9.4, 9.7.2, 9.8.2-9.8.4 with clearer statements on dealing with failing data and reporting
10. Updated 12.2.4 to match attachment sequence
11. Updated Attachment IV B.1 to note Genpure water (brand of system)
12. Updated Attachment IV E2, Holding time of standards from 14 days to 28 days to match extracts when CoA of vendor does not state expiration date (method does not define and only mentions stability).
13. Updated Attachment VII to note that LCSD/LFB Dup is not optional when there are AZ samples in the batch, added Cal Standard 8, and removed duplicate Cal Standard 1
14. Updated Attachment IV. Standard mix are now available from commercial vendor thus neat standards are no longer used in standard preparation. Removed standard preparation procedure from neat standards i.e. details on the weight of each neat standard is dissolved in a certain amount of MeOH

URED – 02/10/21

15. Updated Section 5 with current H&S related information
16. Updated Sections 6 and 7 with regards to critical instrumentation/supplies being available

v3.0

URED – 6/21/21

1. Corrected a typo in Section 14.1.4 to correctly reference 14.1.3
2. Updated 9.6.1 to note that CCVs are acceptable but there may be other criteria on ASM side (PA requires flagged data approval, for instance).

V4.0

URED – 11/7/22

1. Removed references to STARLIMS and webforms and replaced with TALS.
2. Removed notations on standard labeling as this is all handled within TALS.
3. Updated Attachments I, II, and III with more recent information.
4. Noted QC synonyms within TALS.

UNCONTROLLED

ATTACHMENTS

- A. Attachment I, Documentation of Minimum Reporting Limit Study
- B. Attachment II, Documentation of Method Detection Limit Study
- C. Attachment III, Documentation of Demonstration of Precision and Accuracy
- C. Attachment IV, Standard Preparation
- D. Attachment V, Instrument Conditions
- E. Attachment VI, QC Summary
- F. Attachment VII, Analytical Sequence
- G. Attachment VIII, Procedure for pH and Free Chlorine Checks
- H. Attachment IX, Extraction Procedure

Attachment I, Documentation of Minimum Reporting Limit Study

Method: 6533
Instrument ID: LCMS11
Date Reported: 03/23/22
Units: PPT
SOP ID: 30814
SOP Revision: 1

PIR Study													
Analysis Date/Time	3/23/2022 22:18	3/23/2022 22:27	3/23/2022 22:37	3/23/2022 22:46	3/23/2022 22:56	3/23/2022 23:06	3/23/2022 23:15						
Extraction Date	03/22/22	03/22/22	03/22/22	03/22/22	03/22/22	03/22/22	03/22/22						
Extraction Batch	1393921	1393921	1393921	1393921	1393921	1393921	1393921						
LCMS Analyst	KAM	KAM	KAM	KAM	KAM	KAM	KAM						
Extraction Analyst	TBS	TBS	TBS	TBS	TBS	TBS	TBS						
Sample Name	202203220112	202203220113	202203220114	202203220115	202203220116	202203220117	202203220118						
								50%		150%			
Compound Name	SPIKED VALUE	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	ave	%rec	stdev	%rsd	MDL ₉₅
11CL-PF3OUdS	2	2.019	2.085	2.02	2.031	2.031	2.031	2.074	2.041571429	102.1%	0.027	1.3%	0.084
9Cl-PF3ONS	2	2.109	2.183	2.144	2.18	2.257	2.121	2.225	2.174142857	108.7%	0.054	2.5%	0.170
ADONA	2	2.287	2.26	2.2	2.223	2.175	2.343	2.413	2.271571429	113.6%	0.084	3.7%	0.264
GenX	2	2.398	2.548	2.207	2.369	2.414	2.286	2.283	2.357857143	117.9%	0.111	4.7%	0.350
PFBS	2	2.248	2.065	2.316	2.209	2.116	2.208	2.329	2.213	110.7%	0.097	4.4%	0.305
PFDA	2	2.414	2.449	2.278	2.323	2.305	2.28	2.338	2.341	117.1%	0.066	2.8%	0.208
PFDoA	2	2.192	2.284	2.221	2.229	2.328	2.131	2.288	2.239	112.0%	0.067	3.0%	0.209
PFHpA	2	2.333	2.252	2.149	2.366	2.382	2.39	2.395	2.323857143	116.2%	0.092	3.9%	0.288
PFHxA	2	2.314	2.346	2.15	2.535	2.457	2.323	2.335	2.351428571	117.6%	0.121	5.1%	0.380
PFHxS	2	2.138	2.26	2.128	2.193	2.225	2.196	2.203	2.191857143	109.6%	0.046	2.1%	0.146
PFNA	2	2.256	2.326	2.354	2.365	2.245	2.256	2.402	2.314857143	115.7%	0.083	2.7%	0.197
PFOA	2	2.503	2.404	2.241	2.418	2.404	2.312	2.471	2.393285714	119.7%	0.090	3.8%	0.283
PFOS	2	2.225	2.274	2.203	2.152	2.316	2.205	2.385	2.251428571	112.6%	0.079	3.5%	0.249
PFUnA	2	2.259	2.289	2.298	2.229	2.335	2.358	2.261	2.289857143	114.5%	0.045	2.0%	0.142
PFMBA	2	2.13	2.105	2.136	2.32	2.137	2.054	2.202	2.154857143	107.7%	0.085	3.9%	0.267
PFMPA	2	2.24	2.148	2.262	2.304	2.351	2.245	2.295	2.263571429	113.2%	0.064	2.8%	0.201
PFEESA	2	2.154	2.117	2.096	2.144	2.184	2.027	2.183	2.129285714	106.5%	0.055	2.6%	0.174
NFDHA	2	2.181	2.292	1.977	2.145	2.233	2.037	2.254	2.159857143	108.0%	0.116	5.4%	0.365
PFBA	2	2.399	2.294	2.373	2.374	2.355	2.315	2.339	2.349857143	117.5%	0.037	1.6%	0.115
PFPeA	2	2.303	2.478	2.313	2.393	2.304	2.255	2.341	2.341	117.1%	0.074	3.1%	0.231
PFPeS	2	2.219	2.155	2.105	2.272	2.177	2.099	2.293	2.188571429	109.4%	0.076	3.5%	0.240
PFHpS	2	2.218	2.395	2.229	2.185	2.329	2.121	2.338	2.259285714	113.0%	0.097	4.3%	0.306
4:2 FTS	2	2.232	2.285	2.313	2.433	2.521	2.325	2.473	2.368857143	118.4%	0.107	4.5%	0.337
6:2 FTS	2	2.282	2.486	2.348	2.427	2.409	2.251	2.482	2.383571429	119.2%	0.093	3.9%	0.292
8:2 FTS	2	2.425	2.414	2.17	2.298	2.278	2.158	2.378	2.303	115.2%	0.110	4.8%	0.345
									#DIV/0!	#DIV/0!	#DIV/0!	#DIV/0!	#DIV/0!
									#DIV/0!	#DIV/0!	#DIV/0!	#DIV/0!	#DIV/0!
									#DIV/0!	#DIV/0!	#DIV/0!	#DIV/0!	#DIV/0!
									#DIV/0!	#DIV/0!	#DIV/0!	#DIV/0!	#DIV/0!
									#DIV/0!	#DIV/0!	#DIV/0!	#DIV/0!	#DIV/0!
									#DIV/0!	#DIV/0!	#DIV/0!	#DIV/0!	#DIV/0!

LCMS-FO-FRM20133
Ver 4.0 (11/12/18)

1 of 3

Attachment I, Documentation of Minimum Reporting Limit Study (con't)

Method: @533
Instrument ID: LCMS11
Date Reported: 03/23/22
Units: PPT
SOP ID: 30814
SOP Revision: 1

Compound Name	SPIKED VALUE
11CL-PF3OudS	2
9CL-PF3ONS	2
ADONA	2
GenX	2
PFBS	2
PFDA	2
PFDoA	2
PFHpA	2
PFHxA	2
PFHxS	2
PFNA	2
PFOA	2
PFOS	2
PFUnA	2
PFMBA	2
PFMPA	2
PFEESA	2
NFDHA	2
PFBA	2
PFPeA	2
PFPeS	2
PFHpS	2
4:2 FTS	2
6:2 FTS	2
8:2 FTS	2
0	0
0	0
0	0
0	0
0	0

Criteria:	Must be +	Must be <150	Must be >50
	MRL-MDL	HR (PIR)	Upper PIR Limit Lower PIR Limit
	1.916	0.105	107.4 96.8
	1.830	0.214	119.4 98.0
	1.736	0.333	130.2 96.9
	1.650	0.442	140.0 95.8
	1.695	0.385	129.9 91.4
	1.792	0.262	130.2 103.9
	1.791	0.264	125.2 98.7
	1.712	0.363	134.4 98.0
	1.620	0.480	141.6 93.6
	1.854	0.183	118.8 100.4
	1.803	0.248	128.2 103.3
	1.717	0.357	137.5 101.8
	1.751	0.314	128.3 96.9
	1.858	0.179	123.4 105.5
	1.733	0.337	124.6 90.9
	1.799	0.253	125.8 100.5
	1.826	0.220	117.4 95.5
	1.635	0.460	131.0 85.0
	1.885	0.145	124.7 110.3
	1.769	0.292	131.6 102.5
	1.760	0.303	124.6 94.3
	1.694	0.385	132.2 93.7
	1.663	0.425	139.7 97.2
	1.708	0.368	137.6 100.8
	1.655	0.435	136.9 93.4
	#DIV/0!	#DIV/0!	#DIV/0! #DIV/0!
	#DIV/0!	#DIV/0!	#DIV/0! #DIV/0!
	#DIV/0!	#DIV/0!	#DIV/0! #DIV/0!
	#DIV/0!	#DIV/0!	#DIV/0! #DIV/0!
	#DIV/0!	#DIV/0!	#DIV/0! #DIV/0!

LCMS-FO-FRM20133
Ver 4.0 [11/12/18]

2 of 3

Attachment II, Documentation of Method Detection Limit Study

Method: @633
Instrument ID: LCMS11
Date Reported: 03/15/22
Units: ng/L
SOP ID: 30814
SOP Revision: 1

MDL Study														
Analysis Date/Time		3/12/2022 5:18	3/12/2022 5:28	3/15/2022 7:16	3/15/2022 7:25	3/23/2022 22:18	3/23/2022 22:27	3/23/2022 22:37						
Extraction Date		03/10/22	03/10/22	03/11/22	03/11/22	03/22/22	03/22/22	03/22/22						
Extraction Batch		1392667	1392667	1392970	1392970	1393921	1393921	1393921						
LCMS Analyst		KAM	KAM	KAM	KAM	KAM	KAM	KAM						
Extraction Analyst		TBS	TBS	TBS	TBS	TBS	TBS	TBS						
Sample Name		202203100262	202209100265	202209110445	202209110444	202209320112	202209320113	202209320114						
Compound Name	Spike Value	StarLims MRL	Rep 1	Rep 2	Rep 1	Rep 2	Rep 5	Rep 6	Rep 7	ave	%rec	stdev	%rsd	MDL ₂
11CL-PF3OUdS	2	2	2.04	2.109	2.204	2.125	2.019	2.085	2.02	2.086	104.3%	0.067	3.2%	0.211
9Cl-PF3ONS	2	2	2.072	2.082	2.251	2.087	2.109	2.183	2.144	2.13257	106.6%	0.065	3.1%	0.205
ADONA	2	2	2.241	2.334	2.493	2.279	2.287	2.26	2.2	2.29914	115.0%	0.095	4.1%	0.298
GenX	2	5	2.379	2.289	2.677	2.251	2.398	2.548	2.207	2.39271	119.6%	0.169	7.0%	0.530
PFBS	2	2	2.372	2.292	2.387	2.297	2.248	2.065	2.316	2.28243	114.1%	0.107	4.7%	0.337
PFDA	2	2	2.225	2.197	2.348	2.207	2.414	2.449	2.278	2.30257	115.1%	0.102	4.4%	0.322
PFDoA	2	2	2.238	2.283	2.579	2.351	2.192	2.284	2.221	2.30698	115.3%	0.131	5.7%	0.411
PFHpA	2	2	2.279	2.363	2.677	2.163	2.335	2.252	2.149	2.286	114.3%	0.111	4.9%	0.350
PFHxA	2	2	2.505	2.245	2.47	2.54	2.314	2.346	2.15	2.36714	118.4%	0.144	6.1%	0.453
PFHxS	2	2	2.214	2.248	2.392	2.162	2.138	2.26	2.128	2.22029	111.0%	0.092	4.1%	0.289
PFNA	2	2	2.396	2.314	2.367	2.338	2.256	2.326	2.354	2.33586	116.8%	0.044	1.9%	0.140
PFOA	2	2	2.55	2.413	2.328	2.453	2.503	2.404	2.241	2.41314	120.7%	0.104	4.3%	0.328
PFOS	2	2	2.156	2.277	2.366	2.343	2.225	2.274	2.209	2.26343	113.2%	0.075	3.3%	0.236
PFUnA	2	2	2.225	2.282	2.326	2.18	2.259	2.289	2.298	2.26557	113.3%	0.049	2.2%	0.155
PFMBA	2	2	2.327	2.17	2.48	2.171	2.13	2.105	2.136	2.217	110.9%	0.137	6.2%	0.430
PFMPA	2	2	2.494	2.334	2.566	2.298	2.24	2.348	2.262	2.33457	116.7%	0.147	6.3%	0.461
PFEEaA	2	2	2.396	2.239	2.447	2.251	2.154	2.117	2.096	2.24286	112.1%	0.136	6.1%	0.427
NFDHA	2	2	2.064	2.273	2.231	2.425	2.181	2.292	1.977	2.29614	110.3%	0.149	6.8%	0.469
PFBA	2	2	2.371	2.404	2.666	2.457	2.399	2.294	2.379	2.42343	121.2%	0.118	4.9%	0.370
PFPeA	2	2	2.439	2.39	2.56	2.614	2.503	2.478	2.313	2.44243	122.1%	0.118	4.8%	0.371
PFPeS	2	2	2.171	2.234	2.355	2.267	2.219	2.155	2.105	2.21514	110.8%	0.082	3.7%	0.258
PFHpS	2	2	2.334	2.32	2.405	2.226	2.218	2.395	2.229	2.30386	115.2%	0.080	3.5%	0.253
4:2 FTS	2	2	2.334	2.311	2.43	2.248	2.232	2.285	2.313	2.30757	115.4%	0.065	2.8%	0.205
6:2 FTS	2	2	2.423	2.323	2.458	2.252	2.282	2.486	2.348	2.36743	118.4%	0.090	3.8%	0.282
8:2 FTS	2	2	2.133	2.324	2.628	2.399	2.425	2.434	2.117	2.35643	117.8%	0.167	7.1%	0.524

LCMS-FO-FRM25416
Ver 3.0 (01/27/20)

1 of 6

Attachment II, Documentation of Method Detection Limit Study (con't)

Method: @533
Instrument ID: LCMS11
Date Reported: 03/15/22
Units: ng/L
SOP ID: 30814
SOP Revision: 1
PFBA
PFPeA

PFPeA		Blank Results											
Analysis Date/Time	3/11/2022 16:35	3/11/2022 16:44	3/11/2022 16:54	3/12/2022 4:58	3/12/2022 5:09	3/15/2022 7:06	3/15/2022 7:35	MUST USE THE HIGHER OF MDL _g (above) and MDL _b (below)					
Extraction Date	03/09/22	03/09/22	03/09/22	03/10/22	03/10/22	03/11/22	03/11/22						
Extraction Batch	1391889	1391889	1391889	1392667	1392667	1392970	1392970						
LCMS Analyst	KAM	KAM	KAM	KAM	KAM	KAM	KAM						
Extraction Analyst	TBS	TBS	TBS	TBS	TBS	TBS	TBS						
Sample Name	202203090494	202203090495	202203090496	202203100260	202203100261	202203110441	202203110442						
Compound Name	StarLims MRL	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	ave	stdev	%cv	MDL _{calc}	MDL _g
11CL-PF3OUdS	2	0.03	0.025	0.011	0.01	0.007	0.018	0.012	0.01614	0.008	53.0%	0.027	0.043
9Cl-PF3ONS	2	0.026	0.021	0.009	0.004	0.011	0.012	0.008	0.013	0.008	59.6%	0.024	0.037
ADONA	2	0.065	0.038	0.023	0.02	0.01	0.013	0.011	0.02571	0.020	77.1%	0.062	0.088
GenX	2	0.126	0.077	0.056	0.051	0.043	0.049	0.04	0.06314	0.030	47.9%	0.095	0.158
PFBS	2	0.117	0.097	0.048	0.034	0.025	0.05	0.098	0.067	0.036	54.0%	0.114	0.181
PFDA	2	0.079	0.07	0.049	0.049	0.062	0.067	0.058	0.06529	0.010	15.2%	0.031	0.096
PFDoA	2	0.126	0.08	0.068	0.063	0.051	0.072	0.063	0.07471	0.024	32.5%	0.076	0.151
PFHpA	2	0.152	0.108	0.102	0.075	0.113	0.057	0.101	0.10114	0.030	29.7%	0.094	0.195
PFHxA	2	0.197	0.179	0.127	0.095	0.152	0.091	0.104	0.135	0.042	31.2%	0.132	0.267
PFHxS	2	0.1	0.07	0.048	0.024	0.014	0.023	0.036	0.045	0.031	68.0%	0.096	0.141
PFNA	2	0.095	0.048	0.026	0.006	0.045	0.08	0.091	0.05588	0.034	60.7%	0.107	0.162
PFOA	2	0.292	0.245	0.235	0.245	0.108	0.011	0.236	0.196	0.099	50.6%	0.312	0.508
PFOS	2	0.085	0.115	0.107	0.078	0.073	0.098	0.093	0.09271	0.015	16.4%	0.048	0.141
PFUnA	2	0.094	0.081	0.042	0.065	0.08	0.034	0.058	0.06486	0.022	33.7%	0.069	0.133
PFMBA	2	0.07	0.052	0.029	0.036	0.018	0.04	0.053	0.04257	0.017	40.5%	0.054	0.097
PFMPA	2	0.067	0.046	0.027	0.023	0.017	0.026	0.029	0.03357	0.017	51.3%	0.054	0.088
PFEESA	2	0.115	0.087	0.04	0.024	0.02	0.015	0.019	0.04571	0.039	86.2%	0.124	0.170
NFDHA	2	0.063	0.049	0.022	0.011	0.055	0.042	0.098	0.04	0.018	45.7%	0.057	0.028
PFBA	2	0.316	0.175	0.12	0.237	0.148	0.235	0.297	0.21829	0.074	33.9%	0.233	0.451
PFPeA	2	0.414	0.257	0.289	0.312	0.257	0.202	0.32	0.293	0.067	22.7%	0.209	0.502
PFPeS	2	0.1	0.069	0.04	0.008	0.006	0.008	0.005	0.03371	0.038	112.3%	0.012	0.153
PFHpS	2	0.005	0.029	0.02	0.004	0.006	0.008	0.021	0.01329	0.010	74.5%	0.031	0.044
4:2 FTS	2	0.135	0.111	0.084	0.051	0.074	0.04	0.054	0.07843	0.034	44.0%	0.108	0.100
6:2 FTS	2	0.127	0.109	0.1	0.066	0.075	0.079	0.087	0.09188	0.021	23.3%	0.067	0.159
8:2 FTS	2	0.046	0.058	0.039	0.022	0.037	0.06	0.053	0.04357	0.015	33.7%	0.046	0.039

If BLK1 to BLK7 are all non-zero numbers, then use MDL_{calc} value as the blank MDL (MDL_b). If BLK1 to BLK7 are a mix of 0s and non-zero numbers, then use the highest MBLK as the blank MDL value.
If BLK1 to BLK7 are all 0, the MDL_b does not apply (can not calculate MDL_b with zero values)
True analyte MDL is the higher of MDL_g or MDL_b

LCMS-FO-FRM25416
Ver 3.0 (01/27/20)

2 of 6

Attachment III, Documentation of Demonstration of Precision and Accuracy

Method: @533
Instrument ID: LCMS11
Date Reported: 03/16/22
Units: ng/L
SOP ID: 30814
SOP Revision: 1

IDOC

Analysis Date/Time	3/12/2022 5:48	3/12/2022 5:59	3/12/2022 6:09	3/12/2022 6:18	3/12/2022 6:28	3/12/2022 6:38	3/12/2022 6:47
Extraction Date	03/10/22	03/10/22	03/10/22	03/10/22	03/10/22	03/10/22	03/10/22
Extraction Batch	1392667	1392667	1392667	1392667	1392667	1392667	1392667
LCMS Analyst	KAM	KAM	KAM	KAM	KAM	KAM	KAM
Extraction Analyst	TBS	TBS	TBS	TBS	TBS	TBS	TBS
Sample Name	202203100264	202203100265	202203100266	202203100267	202203100268	202203100269	202203100270

Compound Name	True Value	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	AVE	AVE %Rec	Std Dev	%RSD
11CL-PP3OUDS	60	57.294	57.065	56.139	65.569	60.365	55.723	56.898	58.4361	97.39%	3.48297	5.06%
BCL-PP3ONS	60	59.453	60.244	58.788	68.192	62.296	57.941	60.611	61.075	101.79%	3.43345	5.62%
ADONA	60	61.55	61.404	58.976	69.327	64.142	63.594	61.71	62.9576	104.93%	3.27062	5.19%
GenX	60	63.155	60.268	60.47	63.536	63.613	57.26	63.406	61.6726	102.79%	2.42658	3.93%
PFBS	60	57.064	61.808	58.957	70.073	64.773	62.847	61.541	62.4376	104.08%	4.20538	6.74%
PFDA	60	59.971	61.584	61.777	67.592	61.074	59.309	61.141	61.7783	102.96%	2.71118	4.39%
PFDAa	60	61.436	62.753	58.745	66.219	63.626	58.894	62.14	61.9733	103.29%	2.63267	4.26%
PFHpA	60	60.461	59.158	61.598	68.519	64.922	60.843	62.75	62.6073	104.35%	3.18753	5.09%
PFHxA	60	63.928	62.968	61.376	71.189	64.244	60.852	65.745	64.2289	107.05%	3.49443	5.44%
PFHxS	60	60.888	62.172	60.79	67.054	62.729	60.079	61.667	62.0827	103.47%	2.41444	3.89%
PFNA	60	60.923	60.356	58.526	70.282	61.91	62.284	62.796	62.4124	104.02%	3.7474	6.00%
PFOA	60	59.89	60.048	61.37	64.735	63.235	62.179	62.479	61.9909	103.32%	1.72847	2.79%
PFOG	60	60.237	61.979	59.292	70.017	64.093	58.919	61.181	62.1587	103.60%	3.86145	6.21%
PFUnA	60	63.688	62.883	62.055	69.509	63.305	61.682	65.328	64.0643	106.77%	2.67918	4.16%
PFMBA	60	62.569	62.62	60.705	62.786	62.502	60.005	59.948	61.5937	102.65%	1.3085	2.12%
PFMPA	60	62.487	64.072	61.6	69.353	64.263	63.787	63.459	64.143	106.91%	2.48311	3.87%
PFEESA	60	60.25	59.807	60.09	70.609	65.806	61.834	66.599	63.5707	105.95%	4.16391	6.55%
NFDHA	60	55.149	67.206	56.663	67.659	51.241	63.241	60.709	60.2669	100.44%	5.22472	10.33%
PFBA	60	58.512	62.616	60.601	67.637	63.824	61.025	62.502	62.3881	103.98%	2.87963	4.62%
PFPeA	60	64.221	64.79	62.55	70.215	64.928	61.939	61.445	64.2983	107.16%	2.95567	4.60%
PFPeS	60	62.907	59.235	60.716	70.257	65.573	60.85	60.952	62.9271	104.88%	3.81804	6.07%
PFHpS	60	61.103	60.521	59.492	68.702	65.014	59.848	61.696	62.3394	103.90%	3.34664	5.37%
4:2 FTS	60	61.949	58.662	59.313	71.595	65.857	64.252	62.621	63.4641	105.77%	4.39235	6.92%
6:2 FTS	60	60.824	63.215	60.943	67.057	64.056	61.49	63.654	63.0341	105.06%	2.20932	3.50%
8:2 FTS	60	58.982	63.13	59.096	69.135	63.222	62.103	62.116	62.541	104.24%	3.39613	5.43%

Criteria	Low %Rec Criteria	High %Rec Criteria	RSD Criteria (S)
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	130%	20%	20%
70%	1		

Analyzed by:

Extracted by:

Approved by:

Date:

Date:

Date:

LCMS-FO-FRM25416
Ver 3.0 (05/27/20)

3 of 6

Attachment IV: Standard Preparation

A. Sodium Hydroxide Solution (0.08M NaOH)

The NaOH solution is used during primary dilution of purchased neat standards to prevent esterification.

- A.1 Weigh and dissolve 1.6g NaOH in 500mL Genpure water
- A.2 Transfer the prepared NaOH solution into a polypropylene bottle, store at room temperature

B. Methanol: Water Solvent Mixture

An 80% Methanol/20% Water solvent mixture is used for preparing secondary dilution standards.

- B.1 Combine 400mL LCMS Grade methanol and 100mL Genpure water
- B.2 Transfer the prepared MeOH/Water solution into a polypropylene bottle, store at room temperature

C. High Working Standard (HWS), 250µg/L

The high working standard is prepared diluting a purchased stock standard with 80:20 Methanol/Water solution. Sodium Hydroxide is added to prevent esterification. Bring all standards to room temperature prior to use.

- C.1 Add 7mL of @533-MeOH/Water Solvent Mixture, from Attachment III.B, to a VOA vial. Remove 3.2µL of the solvent mixture using a gastight syringe
- C.2 To the same VOA vial, add 3.2µL of 0.08M NaOH solution from Attachment III.A.
- C.3 Add 1mL of 2µg/mL EPA 533 PFC stock standard. Cap and invert several times to mix well
- C.4 Transfer the prepared standard into 2mL polypropylene vial
- C.5 Store refrigerated 4±2°C for up to 6 months or stock standard expiration whichever comes first

D. Low Working Standard (LWS), 25µg/L

The low working standard (LWS) is prepared by diluting the high working standard (HWS) with 80:20 methanol/water. Bring all standards to room temperature prior to use.

- D.1 Add about 8mL of @533-MeOH/Water Solvent Mixture, from Attachment III.B, to a 10mL volumetric flask.
- D.2 To the same volumetric flask, 1mL of 250µg/L HWS
- D.3 Add @533-MeOH/Water Solvent Mixture to 10mL mark. Cap and invert several times to mix well
- D.4 Transfer the prepared standard into 2mL polypropylene vial
- D.5 Store refrigerated 4±2°C for up to 6 months or HWS standard expiration whichever comes first

E. Calibration/Continuing Calibration Standards

E.1 The calibration standard is prepared by combining LWS (or HWS) with IDA, IPS, and Methanol/Water solvent mixture to a 2mL final volume in a propylene vial. The volume of each standard and solvent mixture used is as follows:

Calibration/Continuing Calibration Standard						
Working Concentration*, (µg/L)	Equivalent ppt conc in extract*, (ng/L)	Vol of 25µg/L @533-LWS	Vol of 250µg/L @533-HWS	Vol of 1000µg/L IPS	Vol of 500µg/L IDA	Vol of 80:20 MeOH/Water,
0	0	0 µL	—	20 µL	40 µL	1940 µL
0.5 (MRL)	2 (MRL)	40 µL	—	20 µL	40 µL	1900 µL
1.25	5	—	10 µL	20 µL	40 µL	1930 µL
2.5	10	—	20 µL	20 µL	40 µL	1920 µL
5	20	—	40 µL	20 µL	40 µL	1900 µL
10	40	—	80 µL	20 µL	40 µL	1860 µL
15	60	—	120 µL	20 µL	40 µL	1820 µL
17.5	70	—	140 µL	20 µL	40 µL	1800 µL
20	80	—	160 µL	20 µL	40 µL	1780 µL
0.5 (CCL)	2 (CCL)	40 µL	—	20 µL	40 µL	1900 µL
7.5 (CCM)	30 (CCM)	—	60 µL	20 µL	40 µL	1880 µL
15 (CCH)	60 (CCH)	—	120 µL	20 µL	40 µL	1820 µL
7.5 (QCS)	30 (QCS)	—	60 µL	20 µL	40 µL	1880 µL

Notes: * Concentrations are in terms of PFOA. All IPS and IDA are at 10 µg/L (40 ng/L in extracts) except IPS-PFOS-¹³C₄ at 30 µg/L (120 ng/L in extracts) and IDA-4:2 FTS-¹³C₂, IDA-6:2 FTS-¹³C₂ and IDA-8:2 FTS-¹³C₂ at 40 µg/L (160 ng/L in extracts).

E.2 Store refrigerated 4±2°C for up to 28 days or LWS/HWS/IPS/IDA standard expiration whichever comes first

Mathematically, mid concentration is 10µg/L. However, UCMR allows mid CCV (CCM) to be within 20% of mathematical mid concentration. 80% of 10µg/L is 8µg/L. For ease of measuring standards, CCM is prepared at 7.5µg/L.

F. Concentration conversion from salt form to acid form

If an analyte used was purchased in salt form, and the concentration must be corrected for salt content using the equation:

$$\text{Acid concentration} = \text{salt concentration} \times \frac{\text{MW acid}}{\text{MW salt}}$$

Sample calculation:

PFHpS is purchased as the sodium salt.

	CAS #	Chem Formula	MW-salt (from CoA), g/mol	MW-Acid - by Calc, g/mol	CoA conc As Salt, ug/mL	Conc As Acid by calc, ug/mL
PFHpS	21934-50-9	C7F15SO3Na	472.1	450.118	50	47.67

$$\text{PFHpS Acid concentration} = 472.1\mu\text{g/mL} \times \frac{450.118\text{g/mol}}{472.1\text{g/mol}}$$

$$\text{PFHpS Acid concentration} = 47.67\mu\text{g/mL}$$

Attachment V: LCMS Operating Condition

Gradient

Total Time (min)	Flow Rate (mL/min)	A (%) 20mM Ammonium Acetate	B (%) Methanol
0	0.6	95	5
0.6	0.6	95	5
0.7	0.6	45	55
4.3	0.6	10	90
4.5	0.6	1	99
6.9	0.6	1	99
7.00	0.6	95	5
9.00	0.6	95	5

Notes: Injection Volume = 2µL (LCMS-11) and 5µL (LCMS-10), Column Temperature = 40°C, Cooler Temp = 15°C Rinse Solvent = 80% Water/20% MeOH/0.1% Formic Acid

MS/MS Parameters

Q1 Mass	Q3 Mass	RT, min	Compound	MRM Window	Dwell Weight	DP (volts)	Entrance Potential	CE (volts)	CXP (volts)
631	451	4.62	11CL-PF3OUdS		1	-100	-10	-42	-12
531	351	3.95	9Cl-PF3ONS		1	-60	-10	-40	-12
377	251	2.96	ADONA		1	-30	-10	-16	-12
285	169	2.65	GenX		1	-35	-10	-14	-12
299	80	2.24	PFBS		1	-55	-10	-58	-12
513	469	4.11	PFDA		1	-30	-10	-16	-15
613	569	4.77	PFDoA		1	-25	-10	-18	-15
363	319	2.91	PFHpA		1	-25	-10	-12	-15
313	268.8	2.53	PFHxA		1	-25	-10	-12	-15
399	80	2.92	PFHxS	75	1	-60	-10	-74	-12
463	419	3.73	PFNA		1	-25	-10	-14	-15
413	369	3.32	PFOA		1	-25	-10	-14	-15
499	80	3.72	PFOS	75	1	-65	-10	-108	-12
563	519	4.46	PFUnA		1	-25	-10	-16	-12
279	85	2.3	PFMBA		1	-20	-10	-22	-11
229	85	2.01	PFMPA		1	-15	-10	-22	-11
315	135	2.37	PFEESA		1	-75	-10	-30	-11

Q1 Mass	Q3 Mass	RT, min	Compound	MRM Window	Dwell Weight	DP (volts)	Entrance Potential	CE (volts)	CXP (volts)
295	201	2.47	NFDHA		1	-10	-10	-14	-11
213	169	1.89	PFBA		1	-25	-10	-12	-11
263	219	2.2	PFPeA		1	-20	-10	-12	-11
349	80	2.55	PFPeS		1	-100	-10	-74	-11
449	80	3.32	PFHpS		1	-65	-10	-88	-11
327	307	2.49	4:2 FTS		1	-75	-10	-28	-11
427	407	3.29	6:2 FTS		1	-50	-10	-32	-11
527	507	4.1	8:2 FTS		1	-50	-10	-40	-11
287	169	2.65	Surr-13C3-GenX		1	-35	-10	-14	-11
519	474	4.11	Surr-13C6-PFDA		1	-25	-10	-16	-11
318	273	2.53	Surr-13C5-PFHxA		1	-25	-10	-12	-11
217	172	1.89	Surr-13C4-PFBA		1	-25	-10	-12	-11
268	223	2.2	Surr-13C5-PFPeA		1	-20	-10	-12	-11
367	322	2.9	Surr-13C4-PFHpA		1	-25	-10	-12	-15
421	376	3.31	Surr-13C8-PFOA		1	-25	-10	-14	-15
472	427	3.72	Surr-13C9-PFNA		1	-25	-10	-14	-15
570	525	4.45	Surr-13C7-PFUnA		1	-25	-10	-16	-12
615	570	4.77	Surr-13C2-PFDoA		1	-25	-10	-18	-15
302	80	2.24	Surr-13C3-PFBS		1	-55	-10	-58	-12
402	80	2.91	Surr-13C3-PFHxS		1	-60	-10	-74	-12
507	80	3.72	Surr-13C8-PFOS		1	-65	-10	-108	-12
329	309	2.49	Surr-13C2-4:2 FTS		1	-75	-10	-28	-11
429	409	3.28	Surr-13C2-6:2 FTS		1	-50	-10	-32	-11
529	509	4.1	Surr-13C2-8:2 FTS		1	-50	-10	-40	-11
216	172	1.89	IS-13C3-PFBA		1	-25	-10	-12	-11
415	370	3.31	IS-13C2-PFOA		1	-25	-10	-14	-11
503	80	3.72	IS-13C4-PFOS		1	-65	-10	-108	-11

Q1 Mass	Q3 Mass	RT, min	Compound	MRM Window	Dwell Weight	DP (volts)	Entrance Potential	CE (volts)	CXP (volts)
584	419	4.39	N-EtFOSAA	75	1	-70	-10	-28	-12
570	419	4.21	N-MeTFOSAA	75	1	-70	-10	-30	-12
713	669	5.2	PFTA		1	-25	-10	-22	-15
663	619	4.96	PFTDA		1	-25	-10	-20	-15
589.1	418.8	4.38	Surr-d5-NEtFOSAA		1	-70	-10	-30	-12
573	419	4.2	IS-d3-NMeFOSAA		1	-70	-10	-30	-12
515	470	4.04	Surr-13C2-PFDA		1	-25	-10	-16	-12
315	270	2.49	Surr-13C2-PFHxA		1	-25	-10	-12	-15
417	372	3.26	IS-13C4-PFOA		1	-25	-10	-14	-15

Period 1 Experiment 1 Parameter Table

Source/Gas Compound

Decustering Potential (DP) -100.0

Entrance Potential (EP) -10.0

Collision Energy (CE) -42.0

Collision Cell Exit Potential (CXP) -12.0

Apply the following parameters to all other experiments of the same polarity:
☐ Source/Gas ☐ Compound

OK Cancel Help

Period 1 Experiment 1 Parameter Table

Source/Gas Compound

Ion Source: Turbo Spray

Curtain Gas (CUR) 35.0

Collision Gas (CAD) 9

IonSpray Voltage (IS) -4500.0

Temperature (TEM) 450.0

Ion Source Gas 1 (GS1) 40.0

Ion Source Gas 2 (GS2) 55.0

Apply the following parameters to all other experiments of the same polarity:
☐ Source/Gas ☐ Compound

OK Cancel Help

IPS and IDA References

No.	IPS/IDA/Analyte	Precursor Ion>Product Ion (m/z)	Reference
1	IPS-PFBA- ¹³ C ₃	216>172	None
2	IPS-PFOA- ¹³ C ₂	415>370	None
3	IPS-PFOS- ¹³ C ₄	503>80	None
4	IDA-PFBA- ¹³ C ₄	217>172	1
5	IDA-PFPeA- ¹³ C ₅	268>223	1
6	IDA-PFHxA- ¹³ C ₅	318>273	2
7	IDA-PFHpA- ¹³ C ₄	367>322	2
8	IDA-PFOA- ¹³ C ₈	421>376	2
9	IDA-PFNA- ¹³ C ₉	472>427	2
10	IDA-PFDA- ¹³ C ₆	519>474	2
11	IDA-PFUnA- ¹³ C ₇	570>525	2
12	IDA-PFDoA- ¹³ C ₂	615>570	2
13	IDA-HFPO-DA- ¹³ C ₃	287>169	2
14	IDA-PFBS- ¹³ C ₃	302>80	3
15	IDA-PFHxS- ¹³ C ₃	402>80	3
16	IDA-PFOS- ¹³ C ₈	507>80	3
17	IDA-4:2 FTS- ¹³ C ₂	329>309	3
18	IDA-6:2 FTS- ¹³ C ₂	429>409	3
19	IDA-8:2 FTS- ¹³ C ₂	529>509	3
20	PFBA	213>169	4
21	PFPeA	263>219	5
22	PFHxA	313>269	6
23	PFHpA	363>319	7
24	PFOA	413>369	8
25	PFNA	463>419	9
26	PFDA	513>469	10
27	PFUnA	563>519	11
28	PFDoA	613>569	12
29	HFPO-DA	285>169	13
30	PFBS	299>80	14
31	PFPeS	349>80	15
32	PFHxS	399>80	15
33	PFHpS	449>80	16
34	PFOS	499>80	16
35	4:2 FTS	327>307	17
36	6:2 FTS	427>407	18
37	8:2 FTS	527>507	19
38	PFMPA/PFMOPrA	229>85	4
39	PFMBA/PFMOBA	279>85	5
40	ADONA	377>251	7
41	PFEESA	315>135	14
42	NFDHA /PFMOEOAA	295>201	6
43	9CI-PF3ONS	531>351	16
44	11CI-PF3OUdS	631>451	16

Attachment VI, QC Summary

QC TYPE	CONCENTRATION LEVEL	FREQUENCY	ACCEPTANCE CRITERIA
Analysis Batch		Begins and ends with the appropriate CCCs. Maximum of 20 extracted field samples.	
Extraction Batch		No more than 20 field samples by the same person during a work day using the same lots of supplies.	
Initial Calibration	A minimum of 5 (linear) calibration points, the lowest must be at or lower than the MRL. Minimum 6 points required for Quadratic regression	Run during initial method set up. Run appropriate CCCs for subsequent batches, if CCCs do not meet recovery criteria, run initial calibration	Back calculate the analyte concentration and recoveries must be within 50-150% for points at or below the MRL and within 70-130% for other points.
Lab Instrument Blank (LIB)	Unextracted blank.	Optional. If analyzed, run at the beginning of the run and after the highest calibration point.	Results must be less than 1/3 the MRL.
Continuing Calibration Check (CCC)	CCL (2.0 ng/L), CCM (30 ng/L), or CCH (60 ng/L).	A CCL must be analyzed immediately after the initial calibration curve, prior to any QC or samples. Rotate CCM and CCH after every tenth field sample and at the end of each analysis batch.	Recovery must be within 50-150% of the true value for CCL and within 70-130% for CCM and CCH.
Unextracted Mid-level QCS (QCS)	30 ng/L.	Analyze as part of the IDC, at least quarterly, or with each new calibration prep.	Recovery must be within 70-130% of the true value.
Isotope Performance Standards (IPS)	40 ng/L in terms of IPS-PFOA- ¹³ C ₂ .	Introduce into every field sample, calibration, and QC sample.	IPS area counts must be 50-150% of the average IS area count from the initial calibration.
Isotope Dilution Analogues (IDA)	40 ng/L in terms of IDA-PFOA- ¹³ C ₈ .	Introduce into every field sample, calibration, and QC sample.	Recovery must be within 50-200% of the target value.

QC TYPE	CONCENTRATION LEVEL	FREQUENCY	ACCEPTANCE CRITERIA
Lab Reagent Blank (LRB/MBLK)	Reagent Water prepared, and analyzed as a sample.	Include one LRB with each extraction batch. If an automated system is used, the LRBs are rotated among the ports	Results must be less than 1/3 the MRL.
Lab Fortified Blank (LFB/LCS)	LFB-L (2.0 ng/L), LFB-M (30 ng/L), or LFB-H (60 ng/L).	LFB-L with each extraction batch, as well as LFB-M or LFB-H rotated.	70-130% recovery for mid- and high-levels. 50-150% recovery for low-level.
Lab Fortified Sample Matrix (LFSM/MS)	Rotate between low (2.0 ng/L), mid (30 ng/L), and high (30 ng/L) between extraction batches.	Include one LFSM/MS per extraction batch (20 samples or less).	70-130% recovery for mid- and high-levels. 50-150% recovery for low-level.
Lab Fortified Sample Matrix Duplicate (LFSMD/MSD)/Field Duplicate (FD/DUP)	For LFSMD, rotate between low (2.0 ng/L), mid (30 ng/L), and high (30 ng/L) between extraction batches.	Include at least one FD/DUP or LFSMD/MSD with each extraction batch.	RPD must be less than or equal to 50% for analytes within 2x the MRL. RPDs less than or equal to 30% for analytes higher than 2x the MRL. For LFSMD, 70-130% recovery for mid- and high-levels. 50-150% recovery for low-level.
Field Reagent Blank (FRB)	Reagent Water prepared, and analyzed as a sample.	Process only if an associated field sample contains an analyte at or above the MRL.	Results must be less than or equal to 1/3 the MRL.
Initial Demonstration of Low System Background	Prepare, extract, and analyze as a sample.	Analyze an extracted LRB prior to any other IDC steps. If an automated system is used, the LRBs must be extracted from each port. After IDC, LRB are rotated among the ports	Results must be less than 1/3 the MRL.
Initial Demonstration of Precision	Prepare, extract and analyze 7 replicate LFB/LCSs fortified at 30 ng/L.	When beginning the use of this method, with each new analyst, and after a major change in instrumentation.	RSD must be less than 20% for the analytes.

QC TYPE	CONCENTRATION LEVEL	FREQUENCY	ACCEPTANCE CRITERIA
Initial Demonstration of Accuracy	Use the same results obtained from the Initial Demonstration of Precision.	When beginning the use of this method, with each new analyst, and after a major change in instrumentation.	Mean recovery must be within 70-130%.
Continuing Demonstration of Capability		MDL and Precision and Accuracy performed annually for each analyst or extractionist.	RSD must be less than 20% for the analytes. Mean recovery must be within 70-130%.
MRL Confirmation	Prepare, extract and analyze 7 replicate LFB/LCSs fortified at the proposed MRL.	Initially on each instrument and whenever there is a major change in instrumentation. Each new analyst and extractionist, performing this method.	The upper PIR limits must be less than or equal to 150%. The lower PIR limits must be greater than or equal to 50%.
LFB/LFBDup (LCS/LCSDup)	Rotate between mid (30 ng/L), and high (30 ng/L) between extraction batches.	For Arizona samples when MS/MSD failed acceptance for accuracy and precision	RPD must be less than or equal to 50% for analytes within 2x the MRL. RPDs less than or equal to 30% for analytes higher than 2x the MRL. 70-130% recovery for mid- and high-levels. 50-150% recovery for low-level.
Establish RT for both linear and branched isomers	Run at mid to high level concentration	Run as part of IDC Repeat anytime chromatographic changes occur that alter analyte retention time.	All isomers of each analyte must elute with the same MRM window

Attachment VII, Analytical Sequence

Cal Standard 1

Cal Standard 2

Cal Standard 3

Cal Standard 4

Cal Standard 5

Cal Standard 6

Cal Standard 7

Cal Standard 8

CCC at MRL Level

QCS at Mid Level

LRB/MBLK

MRL Check

LCS/LFD

LCSD/LFB Dup (If needed, however this is not optional when there are AZ samples in the batch)

Field Sample 1

LFSM/MS on Field Sample 1

LFSMD/MSD on Field Sample 1

Field Sample 2 – 10

CCC at Mid Level

Field Sample 11 – 20

CCC at High Level

Attachment VIII, Procedure for pH and Free Chlorine Check

- 1) pH Check using Fisher brand pH Strips. (pH range 5 to 9)
 - a) Check the pH of the samples before checking the free chlorine.
 - b) Uncap the sample bottle.
 - c) Using a KimWipe, wipe the mouth of the bottle every time before and after pouring out the sample. Pour 20 mL of the sample into a clean 1 ounce graduated medicine cup.
 - d) Dip a pH test strip into the sample aliquot. DO NOT dip the pH test strip in the sample.
 - e) Read the strip after the manufacturer's allotted development time (immediately for the Fisher brand strips). Compare the color of the paper to the corresponding color on the chart furnished with the paper. (The pH must be between 6.5 and 7.5.)
 - f) Record the pH first in TALS as the original observation, and then record the pH on the sample bottle label. Mark with an "X" and set aside samples that failed to meet pH acceptance criteria for later pH verification using VWR pH strips. (pH range 6 to 10)
- 2) Free Chlorine Check by using SenSafe Strips
 - a) Using the same 20mL aliquot that is already in the medicine cup that was used for pH as in step 1, take 1 SenSafe Strip and stroke it 40 times for 20 seconds in the sample, making 1 back stroke and 1 forth stroke per second. Make sure to keep the indicator window portion of the strip completely submerged in the sample during the 20 seconds.
 - b) Record the residual chlorine result first in TALS as the original observation, and then record the residual chlorine result on the sample bottle label.If free chlorine is $\geq 0.1\text{mg/L}$, analyst must notify ASM and generate NCM.

Attachment IX. Extraction Procedure

12) PROCEDURE

A. This procedure shall be performed using an SPE delivery apparatus device. Extraction and/or elution steps must not be changed or omitted to accommodate the use of an automated system. If an automated system is used, the LRBs must be rotated among the ports during routine analysis to ensure that all the valves and tubing meet the LRB requirements. Acceptable performance for the LRB must be met for each port during the IDC.

B. Some of the PFAS adsorb to surfaces, including polypropylene. Therefore, the aqueous sample bottles must be rinsed with the elution solvent whether extractions are performed manually or by automation. The bottle rinse is passed through the cartridge to elute the method analytes and is then collected.

C. The SPE cartridges and sample bottles described in this section are designed as single use items and shall be discarded after use. They may not be refurbished for reuse in subsequent analyses.

D. SAMPLE PREPARATION – Within 3 days of sample receipt, verify free chlorine. Verify pH prior to extraction

1. Verify and record that the sample pH is between 6.0 and 8.0. Acetic acid may be added as needed to lower the pH. If the pH is adjusted, generate a NCM to document the addition of acetic acid to the sample.
2. Verify and record that the sample has free chlorine less than 0.10 ppm. Generate a NCM if free chlorine is greater than 0.10 ppm. Notify ASM but proceed with extraction unless advised to cancel analysis.
3. Weigh and record the initial sample bottle weight to the nearest 0.5 g. After extraction, the empty bottle is also weighed. The weights are used for initial sample volume determination. Some of the PFAS adsorb to surfaces, thus the sample volume may NOT be transferred to a graduated cylinder for volume measurement.
4. The QC samples may be prepared by measuring 250 mL of reagent water with a polypropylene graduated cylinder or filling a 250 mL sample bottle to near the top. Verify and record the pH and free chlorine of all the QC samples
5. Record the spiking standards used for each batch.
6. Fortify the QC and samples with 10 µL of 1ppm isotope dilution analogue spiking standard. Cap and invert to mix. Each extraction batch must have an MRL spiked at the low level. The LCS/LCSD must be spiked at mid and high levels, which are alternated between batches. The MS/MSD must be spiked at low, mid and high levels, which are also alternated between batches. See table below for spiking amounts and spiking standards.

QC Sample	25 ppb Low Working Standard	250 ppb High Working Standard	Final Concentration in 250 mL sample
MRL	20 µL		2 ng/L
LCS/LCSD – mid level		30 µL	30 ng/L
LCS/LCSD – high level		60 µL	60 ng/L
MS/MSD – low level	20 µL		2 ng/L
MS/MSD – mid level		30 µL	30 ng/L
MS/MSD – high level		60 µL	60 ng/L

Spiking Standard	Amount Spiked	Final Concentration
0.5 ppm IDA	20 µL	40ng/L (160ng/L of telomere sulfonates) in 250 mL sample
1 ppm IPS	10 µL	10µg/L (30µg/L 13C4-PFOS) in 1 mL extract

MANUAL EXTRACTION –

APPARATUS

- A. Solid Phase Extraction (SPE) Apparatus
 1. Vacuum Extraction Manifold: Equipped with flow/vacuum control
 2. Sample Delivery System: Use of polypropylene transfer lines, which transfers the sample directly from the sample container to the SPE cartridge, is recommended
 3. Laboratory Vacuum System: Sufficient capacity to maintain a vacuum of approximately 10 to 15 inches of mercury

CARTRIDGE SPE PROCEDURE

- A. Rinse the polypropylene transfer lines with methanol and then reagent water.
- B. CARTRIDGE CLEAN-UP and CONDITIONING – DO NOT allow cartridge packing material to go dry during any of the conditioning steps. If the cartridge goes dry during the conditioning phase, the conditioning must be started over.
 1. Rinse each cartridge with three 5 mL aliquots of methanol. A total 15 mL of methanol.

2. Rinse each cartridge with one 5 mL aliquot of aqueous 0.10 M phosphate buffer pH 7.
3. Condition each cartridge with two 5 mL aliquots of aqueous 0.10 M phosphate buffer pH 7, without allowing the buffer solution to drop below the top edge of the packaging. A total of 10 mL of phosphate buffer solution.
4. Add 3 mL of phosphate buffer to each cartridge and fill the remaining volume with reagent water, approximately 2mL.

C. SAMPLE EXTRACTION

1. Attach a polypropylene transfer line to each cartridge.
2. Turn on the vacuum and adjust it to approximately -5 inches Hg.
3. Begin transferring the sample to the cartridge at a flow rate of about 5 mL/min. Do not allow the cartridge to go dry before all the sample has passed through. The transferring step should take approximately 50 minutes. Flow rates above 5 mL/min during transferring may cause low analyte recovery.

D. SAMPLE BOTTLE and CARTRIDGE RINSE

1. After the entire sample has passed through the cartridge, rinse the sample bottles with a 10 mL aliquot of 1 g/L ammonium acetate in reagent water. Draw the rinsate through the sample transfer lines and the cartridges.
2. Add 1 mL of methanol to the sample bottle and draw through the sample transfer lines and the cartridges. This step is designed to remove the water from the transfer lines and cartridges to reduce the salt and water present in the eluate. The methanol rinse can also help to reduce interferences by removing weakly retained organic material prior to elution.
3. Draw air through the cartridge for 10 minutes at high vacuum (15 – 20 in. Hg).

E. SAMPLE BOTTLE and CARTRIDGE ELUTION

1. Turn off and release the vacuum.
2. Lift the extraction manifold top and insert a rack with labeled collection tubes into the extraction tank to collect the extracts as they are eluted from the cartridges.
3. Rinse the sample bottles with 5 mL of methanol with 2% ammonium hydroxide (v/v)
4. Elute the analytes from the cartridges by pulling the 5 mL of 2% ammonium hydroxide in methanol (v/v) through the sample transfer lines and the cartridges. Use a low vacuum such that the solvent exits the cartridge in a dropwise fashion.
5. Repeat the sample bottle rinse and cartridge elution with a second 5 mL aliquot of methanol with 2% ammonium hydroxide (v/v)
6. Remove transfer lines

7. If needed, add additional 2% ammonium hydroxide in methanol (v/v) directly into cartridge and elute in a dropwise fashion. Final volume should be approximately 10 mL.
8. After elution, store samples at $\leq 6^{\circ}\text{C}$ until ready to concentrate.

F. EXTRACT CONCENTRATION and FINAL VOLUME

1. Concentrate the extract to dryness under a gentle stream of nitrogen in a heated water bath ($55 - 60^{\circ}\text{C}$).
2. Reconstitute the extract with 990 μL of 20% reagent water in methanol (v/v)
3. Add 10 μL of the internal standard to the extract and vortex.
4. Transfer the final extract to a polypropylene autosampler vial and label with the appropriate sample number or QC.
5. Store extracts at room temperature until ready for analysis.

G. SAMPLE VOLUME DETERMINATION – Weigh and record the empty bottle to the nearest 0.5 g. This value is subtracted from the initial sample bottle weight to determine the initial sample volume. Assume a sample density of 1.0 g/L.

AUTOMATED EXTRACTION – This procedure is in development

APPARATUS

A. Solid Phase Extraction (SPE) Apparatus

1. An automatic/robotic sample preparation system (Thermo Scientific Dionex Autotrace 280 or equivalent) designed for use with SPE cartridges.
2. Extraction and/or elution steps may not be changed or omitted to accommodate the use of an automated system.
3. Care must be taken with automated SPE systems to ensure the PTFE commonly used in these systems does not contribute to unacceptable analyte concentrations in the LRB .

CARTRIDGE SPE PROCEDURE – The following steps are the procedure at which the autotrace is programmed for this method.

No.	Method
1	Process 6 Samples using the following method steps:
2	Condition Cartridge with 2.0 mL of P & T Methanol into solvent waste
3	Dry Cartridge with gas for 1.0 minutes
4	Condition Cartridge with 10.0 mL of P & T Methanol into solvent waste
5	Condition Cartridge with 10.0 mL of 0.1M phosphate buffer into aqueous waste
6	Condition Cartridge with 3.0 mL of 0.1M phosphate buffer into aqueous waste

7	Condition Cartridge with 5.0 mL of reagent water into aqueous waste
8 ^a	Load 340 mL of sample onto Cartridge
9	Pause and Alert operator, resume when CONTinue is pressed
10 ^{a, b}	Load 20.0 mL of sample onto Cartridge
11	Pause and Alert operator, resume when CONTinue is pressed
12 ^{a, c}	Load 10.0 mL of sample onto Cartridge
13	Dry Cartridge with gas for 15.0 minutes
14 ^{a, d}	Manually rinse Sample Container with 10.0 mL to collect
15 ^{a, d}	Manually rinse Sample Container with 10.0 mL to collect
16	Concentrate Sample with gas for 2.0 minutes
17	End

Notes: Nitrogen gas pressure is set at 10 psi.

^a Ensure that the sample distribution line is properly placed in the sample bottle

to withdraw the entire sample volume from the bottle.

^b During Step 10, rinse the sample bottle with 10 mL of 1 g/L ammonium acetate/reagent water.

^c During Step 12, rinse the sample bottle with 1 mL methanol.

^d During Steps 14 and 15, rinse the sample bottle with 5 mL of 2% ammonium hydroxide (v/v)/methanol

Flow Rates	Instrument Parameters
Cond Flow: 5.0 mL/min	Max. Elution Vol.: 12.0 mL
Load Flow: 5.0 mL/min	Exhaust Fan On: Yes
Rinse Flow: 20.0 mL/min	Beeper On: Yes
Elute Flow: 5.0 mL/min	Solvent Set
Cond Air Push: 15.0 mL/min	Solvent 1: 0.1M phosphate buffer
Rinse Air Push: 20.0 mL/min	Solvent 2: P & T Methanol
Elute Air Push: 5.0 mL/min	Solvent 3: Reagent Water
SPE Parameters	Solvent 4: Not Used
Push Delay: 5 sec	Solvent 5: Not Used
Air Factor: 1.0	
Autowash Vol.: 1.00 mL	

EXTRACT CONCENTRATION and FINAL VOLUME

- Concentrate the extract to dryness under a gentle stream of nitrogen in a heated water bath (55 – 60° C).
- Reconstitute the extract with 990 µL of 20% reagent water in methanol (v/v)
- Add 10 µL of the internal standard to the extract and vortex.

- D. Transfer the final extract to a polypropylene autosampler vial and label with the appropriate sample number or QC.
- E. Store extracts at room temperature until ready for analysis.

SAMPLE VOLUME DETERMINATION – Weigh and record the empty bottle to the nearest 0.5 g. This value is subtracted from the initial sample bottle weight to determine the initial sample volume. Assume a sample density of 1.0 g/L.

UNCONTROLLED

 Document number: LCMS-SOP24130 Old Reference:	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Level:  Standard Operating Procedure
Version: 6		Organisation level: 4-Laboratory Site
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Responsible: 6_QAD4

EUROFINS EATON ANALYTICAL, LLC
Standard Operating Procedure

EPA Method 537.1 Version 2.0 March 2020

Confidential

- 01) TITLE
- 02) SCOPE AND APPLICATION
- 03) METHOD SUMMARY
- 04) INTERFERENCES
- 05) SAFETY CONSIDERATIONS
- 06) INSTRUMENTATION/APPARATUS
- 07) REAGENTS & STANDARDS
- 08) SAMPLE COLLECTION, PRESERVATION and HANDLING
- 09) CALIBRATION PROCEDURE
- 10) ANALYTICAL PROCEDURE
- 11) QUALITY CONTROL REQUIREMENTS
- 12) CALCULATIONS
- 13) METHOD PERFORMANCE
- 14) REFERENCES
- 15) DEVIATIONS FROM REFERENCED METHODOLOGY
- 16) METHOD DETECTION LIMIT
- 17) DEMONSTRATION OF CAPABILITY
- 18) DEFINITIONS
- 19) POLLUTION PREVENTION
- 20) WASTE MANAGEMENT
- 21) REVISIONS
- 22) ATTACHMENTS

01) TITLE

Determination of selected per-and polyfluorinated alkyl acids in drinking water by solid phase extraction and liquid chromatography/tandem mass spectrometry (LC/MS/MS).

02) SCOPE AND APPLICATION

A. This is a liquid chromatography/tandem mass spectrometry (LC/MS/MS) method for the determination of selected perfluorinated alkyl acids (PFAS) in drinking water. Accuracy and precision data have been generated in reagent water, and finished ground and surface waters for the compounds listed in the table below.

Table 1. This method is applicable to the following analytes:

Analyte	Acronym	CAS #	MRL, ng/L
Method Analytes			
N-ethyl Perfluorooctanesulfonamidoacetic acid	NEtFOSAA	2991-50-6	2

	Always check on-line for validity.		Level: 
	EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)		Standard Operating Procedure
Document number: LCMS-SOP24130			Organisation level: 4-Laboratory Site
Old Reference:			Responsible: 6_QAD4
Version: 6			
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract		

N-methyl Perfluorooctanesulfonamidoacetic acid	NMeFOSAA	2355-31-9	2
Perfluorobutanesulfonic acid	PFBS	375-73-5	2
Perfluorodecanoic acid	PFDA	335-76-2	2
Perfluorododecanoic acid	PFDoA	307-55-1	2
Perfluoroheptanoic acid	PFHpA	375-85-9	2
Perfluorohexanesulfonic acid	PFHxS	355-46-4	2
Perfluorohexanoic acid	PFHxA	307-24-4	2
Perfluorononanoic acid	PFNA	375-95-1	2
Perfluorooctanesulfonic acid	PFOS	1763-23-1	2
Perfluorooctanoic acid	PFOA	335-67-1	2
Perfluorotetradecanoic acid	PFTeDA	376-06-7	2
Perfluorotridecanoic acid	PFTrDA	72629-94-8	2
Perfluoroundecanoic acid	PFUnA	2058-94-8	2
Hexafluoropropylene oxide dimer acid	HFPO-DA / Gen-X	13252-13-6 ^a	2
4, 8-dioxo-3H-Perfluorononanoic acid	ADONA	919005-14-4 ^b	2
9-chlorohexadecafluoro-3-oxanone-1-sulfonic acid	9CI-PF3ONS	756426-58-1 ^c	2
11-chloroeicosafluoro-3-oxaundecane-1-sulfonic acid	11CI-PF3OUdS	763051-92-9 ^d	2

- a. HFPO-DA is one component of the GenX processing aid technology.
- b. ADONA is available as the sodium salt (no CASRN) and the ammonium salt (CASRN is 958445-448).
- c. 9CI-PF3ONS analyte is available in salt form (e.g. CASRN of potassium salt is 73606-19-6).
- d. 11CI-PF3OUdS is available in salt form (e.g. CASRN of potassium salt is 83329-89-9).

B. The Minimum Reporting Level (MRL) is the lowest analyte concentration that meets Data Quality Objectives (DQOs) that are developed based on the intended use of this method. The single laboratory lowest concentration MRL (LCMRL) is the lowest true concentration for which the future recovery is predicted to fall, with high confidence (99%), between 50 and 150% recovery. EPA's single laboratory LCMRLs for analytes in this method range from 0.53-6.3 ng/L. The procedure used to determine the LCMRL is described in section 14a of the SOP. Eurofins Eaton Analytical at Monrovia MRL check confirmation is done at 2ppt.

C. Eurofins Eaton Analytical (EEA-P) according to the EPA method will not be required to determine the LCMRL for this method, but will need to demonstrate that the MRL meets the requirements described in Section 9.2.6 of the referenced method EPA 537.1; Minimum Reporting Level Confirmation.

1. MDL studies are required annually by EEA-P following 40 CFR 136 MDL Revision 2. Detection limit is defined as the statistically calculated minimum concentration that can be measured with 99% confidence that the reported value is greater than zero.
2. The DL is compound dependent and is dependent on extraction efficiency, sample matrix, fortification concentration, and instrument performance.

 Document number: LCMS-SOP24130 Old Reference:	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Level:  Standard Operating Procedure
Version: 6		Organisation level: 4-Laboratory Site
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Responsible: 6_QAD4

3. This method is intended for use by analysts skilled in solid phase extractions, the operation of LC/MS/MS instruments, and the interpretation of the associated data.

D. METHOD FLEXIBILITY – In recognition of technological advances in analytical systems and techniques, the laboratory is permitted to modify the evaporation technique, separation technique, LC column, mobile phase composition, LC conditions and MS and MS/MS conditions. Changes may not be made to sample collection and preservation, the sample extraction steps, or to the quality control requirements (Section 11) of the referenced method EPA 537.1. Method modifications should be considered only to improve method performance. Modifications that are introduced in the interest of reducing cost or sample processing time, but result in poorer method performance, should not be used. **Analytes must be adequately resolved chromatographically in order to permit the mass spectrometer to dwell on a minimum number of compounds eluting within a retention time window. Instrumental sensitivity (or signal-to-noise) will decrease if too many compounds are permitted to elute within a retention time window.** In all cases where method modifications are proposed, the analyst must perform the procedures outlined in the initial demonstration of capability (IDC), verify that all Quality Control (QC) acceptance criteria in this method (Section 9) of the referenced method EPA 537.1 are met, and that acceptable method performance can be verified in a real sample matrix.

E. California State Water Resources Control Board Division of Drinking Water is requiring public drinking water systems approving USEPA Method 537.1 to analyze Perfluorinated Alkyl Substances (PFAs) in drinking water matrix.

03) METHOD SUMMARY

250-mL water sample is fortified with surrogates and passed through a solid phase extraction (SPE) cartridge containing polystyrenedivinylbenzene (SDVB) to extract the method analytes and surrogates. The compounds are eluted from the solid phase with a small amount of methanol. The extract is concentrated to dryness with nitrogen in a heated water bath, and then adjusted to a 1-mL volume with IS in 96:4% (vol/vol) methanol:water. A 2-5-µL injection is made into an LC equipped with a C₁₈ column that is interfaced to an MS/MS. The analytes are separated and identified by comparing the acquired mass spectra and retention times to reference spectra and retention times for calibration standards acquired under identical LC/MS/MS conditions. The concentration of each analyte is determined by using the internal standard technique. Surrogate analytes are added to all Field and QC Samples to monitor the extraction efficiency of the method analytes.

04) INTERFERENCES

A. All glassware must be meticulously cleaned. Wash glassware with detergent and tap water, rinse with tap water, followed by a reagent water rinse. Non-volumetric glassware can be heated in a muffle furnace at 400 °C for 2 hours or solvent rinsed. Volumetric glassware should be solvent rinsed and not be heated in an oven above 120 °C. Store clean glassware inverted or capped. **Do not cover with aluminum foil because PFAAs can be potentially transferred from the aluminum foil to the glassware.**

a. NOTE: PFAS standards, extracts and samples should not come in contact with any glass containers or pipettes as these analytes can potentially adsorb to glass surfaces. PFAA analyte, IS and SUR standards commercially purchased in glass ampoules are acceptable; however, all subsequent transfers or dilutions performed by the analyst must be prepared and stored in polypropylene containers.

 Document number: LCMS-SOP24130 Old Reference: Version: 6 Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS) Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Level:  Standard Operating Procedure Organisation level: 4-Laboratory Site Responsible: 6_QAD4

B. Method interferences may be caused by contaminants in solvents, reagents (including reagent water), sample bottles and caps, and other sample processing hardware that lead to discrete artifacts and/or elevated baselines in the chromatograms. The method analytes in this method can also be found in many common laboratory supplies and equipment, such as PTFE (polytetrafluoroethylene) products, LC solvent lines, methanol, aluminum foil, SPE sample transfer lines, etc. All items such as these must be routinely demonstrated to be free from interferences (less than 1/3 the MRL for each method analyte) under the conditions of the analysis by analyzing laboratory reagent blanks as described in Section 11 of the referenced method EPA 537.1.

Subtracting blank values from sample results is not permitted.

C. Matrix interferences may be caused by contaminants that are co-extracted from the sample. The extent of matrix interferences will vary considerably from source to source, depending upon the nature of the water. Humic and/or fulvic material can be co-extracted during SPE and high levels can cause enhancement and/or suppression in the electrospray ionization source or low recoveries on the SPE sorbent. Total organic carbon (TOC) is a good indicator of humic content of the sample. Under the LC conditions used during method development, matrix effects due to total organic carbon (TOC) were not observed.

D. Relatively large quantities of the preservative (Section 8) are added to sample bottles. The potential exists for trace-level organic contaminants in these reagents. Interferences from these sources should be monitored by analysis of laboratory reagent blanks (Section 11) of the referenced method EPA 537.1, particularly when new lots of reagents are acquired.

E. SPE cartridges can be a source of interferences. The analysis of field and laboratory reagent blanks can provide important information regarding the presence or absence of such interferences. Brands and lots of SPE devices should be tested to ensure that contamination does not preclude analyte identification and quantitation.

05) SAFETY CONSIDERATIONS

A. **Safety Data Sheets (SDSs)** must be reviewed for information pertaining to the proper treatment and precautionary measure prior to handling any reagents. They are located in red binders in the **Safety Officer's office** and **online**.

B. Refer to the SOP "**Hazardous Waste Management and Sample Disposal Procedures, Chemical Hygiene Plan**" and OSHA Standard 29 CFR 1910.1450 Occupational Exposure to Hazardous Chemicals in Laboratories; Final Rule for additional safety information

C. All samples, standards, and solvents used in this SOP should be treated as potential health hazards and handled with care. Chemists should consult relevant SDSs and follow laboratory safety procedures when doing extractions, preparing standard solutions, and performing the analysis.

D. The toxicity or carcinogenicity of each reagent used in this method has not been precisely defined. Each chemical should be treated as a potential health hazard, and exposure to these chemicals should be minimized. Each laboratory is responsible for maintaining an awareness of OSHA regulations regarding safe handling of chemicals used in this method.

E. PFOA has been described as likely to be carcinogenic to humans. Pure standard materials and stock standard solutions of these method analytes should be handled with suitable protection to skin and eyes, and care should be taken not to breathe the vapors or ingest the materials.

 Document number: LCMS-SOP24130 Old Reference: Version: 6	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Level:  Standard Operating Procedure Organisation level: 4-Laboratory Site
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Responsible: 6_QAD4

06) INSTRUMENTATION/APPARATUS



UNCONTROLLED

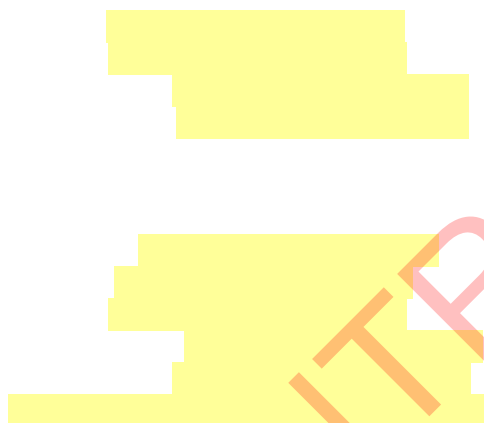
 Document number: LCMS-SOP24130 Old Reference: Version: 6	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Level:  Standard Operating Procedure Organisation level: 4-Laboratory Site
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Responsible: 6_QAD4

UNCONTROLLED

 Document number: LCMS-SOP24130 Old Reference: Version: 6	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Level:  Standard Operating Procedure Organisation level: 4-Laboratory Site
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Responsible: 6_QAD4

M.

7) REAGENTS & STANDARDS



UNCONTROLLED

 Document number: LCMS-SOP24130 Old Reference: Version: 6	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Level:  Standard Operating Procedure Organisation level: 4-Laboratory Site
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Responsible: 6_QAD4

UNCONTROLLED

 Document number: LCMS-SOP24130 Old Reference: Version: 6	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Level:  Standard Operating Procedure Organisation level: 4-Laboratory Site
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Responsible: 6_QAD4

UNCONTROLLED

 Document number: LCMS-SOP24130 Old Reference: Version: 6	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Level:  Standard Operating Procedure Organisation level: 4-Laboratory Site
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Responsible: 6_QAD4

UNCONTROLLED

 Document number: LCMS-SOP24130 Old Reference:	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Level:  Standard Operating Procedure
Version: 6		Organisation level: 4-Laboratory Site
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Responsible: 6_QAD4

08) SAMPLE COLLECTION, PRESERVATION and HANDLING

A. SAMPLE BOTTLE PREPARATION

1. Samples must be collected in a 275-mL polypropylene bottle fitted with a polypropylene screw-cap (vendors refer to this as the 250-mL bottle). 250 ml sample is extracted as the extra volume is used for free chlorine and pH check.
2. The preservation reagent, listed in the table below, is added to each sample bottle as a solid prior to shipment to the field (or prior to sample collection).

Compound	Amount	Purpose
Trizma®	5.0 g/L	buffering reagent and removes free chlorine

B. SAMPLE COLLECTION

1. The sample handler must wash their hands before sampling and wear nitrile gloves while filling and sealing the sample bottles. PFAS contamination during sampling can occur from a number of common sources, such as food packaging and certain foods and beverages. Proper hand washing and wearing nitrile gloves will aid in minimizing this type of accidental contamination of the samples.
2. Open the tap and allow the system to flush until the water temperature has stabilized (approximately 3 to 5 min). Collect samples from the flowing system.
3. Fill sample bottles, taking care not to flush out the sample preservation reagent. Samples do not need to be collected headspace free.
4. After collecting the sample, cap the bottle and agitate by hand until preservative is dissolved. Keep the sample sealed from time of collection until extraction.

C. FIELD REAGENT BLANKS (FRB)

1. A FRB must be handled along with each sample set. The sample set is composed of samples collected from the same sample site and at the same time. At the laboratory, fill the field blank sample bottle with reagent water and preservatives, seal, and ship to the sampling site along with the sample bottles. For each FRB shipped, an empty sample bottle (no preservatives) must also be shipped. At the sampling site, the sampler must open the shipped FRB and pour the preserved reagent water into the empty shipped sample bottle, seal and label this bottle as the FRB. The FRB is shipped back to the laboratory along with the samples and analyzed to ensure that PFAS were not introduced into the sample during sample collection/handling.
2. The same batch of preservative must be used for the FRBs as for the field samples.
3. The reagent water used for the FRBs must be initially analyzed for method analytes as a LRB/MBLK and must meet the LRB/MBLK criteria prior to use. This requirement will ensure samples are not being discarded due to contaminated reagent water rather than contamination during sampling.

D. SAMPLE SHIPMENT AND STORAGE – Samples must be chilled during shipment and must not exceed 10 °C during the first 48 hours after collection. Sample temperature must be confirmed to be

 Document number: LCMS-SOP24130 Old Reference:	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Level:  Standard Operating Procedure
Version: 6		Organisation level: 4-Laboratory Site
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Responsible: 6_QAD4

at or below 6 °C when the samples are received at the laboratory 48 hours after sample collection. Samples stored in the lab must be held at or below 6 °C until extraction, but should not be frozen.

NOTE: Samples that are significantly above 10° C, at the time of collection, may need to be iced or refrigerated for a period of time, in order to chill them prior to shipping. This will allow them to be shipped with sufficient ice to meet the above requirements.

E. SAMPLE DECHLORINATION AND PRESERVATION – Trizma as a buffer should bring sample pH to near 7.0 at room temperature. The pH of samples must be verified using pH paper, and the pH should be between 6.5 and 7.5. If pH does not meet acceptance criteria, sample is rejected. Free Chlorine is checked by using SenSafe Strips. If free chlorine is >0.1 mg/L, there is no need to confirm with DPD method. There is no need to add more Trizma Buffer if sample free chlorine is > 0.1 mg/L (See Attachment XI)

F. SAMPLE AND EXTRACT HOLDING TIMES – Results of the sample storage stability study indicated that all compounds listed in this method have adequate stability for 14 days when collected, preserved, shipped and stored as described in Sections 8.D. Therefore, water samples should be extracted as soon as possible but must be extracted within 14 days. Extracts must be stored at room temperature and analyzed within 28 days after extraction.

9) CALIBRATION PROCEDURE

 Document number: LCMS-SOP24130 Old Reference: Version: 6	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Level:  Standard Operating Procedure Organisation level: 4-Laboratory Site
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Responsible: 6_QAD4

UNCONTROLLED

 Document number: LCMS-SOP24130 Old Reference: Version: 6	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Level:  Standard Operating Procedure Organisation level: 4-Laboratory Site
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Responsible: 6_QAD4

UNCONTROLLED

 Document number: LCMS-SOP24130 Old Reference: Version: 6	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Level:  Standard Operating Procedure Organisation level: 4-Laboratory Site
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Responsible: 6_QAD4

UNCONTROLLED

 Document number: LCMS-SOP24130 Old Reference: Version: 6	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Level:  Standard Operating Procedure Organisation level: 4-Laboratory Site
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Responsible: 6_QAD4

10) ANALYTICAL PROCEDURE

UNCONTROLLED

 Document number: LCMS-SOP24130 Old Reference: Version: 6	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Level:  Standard Operating Procedure Organisation level: 4-Laboratory Site
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Responsible: 6_QAD4

UNCONTROLLED

 Document number: LCMS-SOP24130 Old Reference: Version: 6	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Level:  Standard Operating Procedure Organisation level: 4-Laboratory Site
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Responsible: 6_QAD4

UNCONTROLLED

 Document number: LCMS-SOP24130 Old Reference: Version: 6	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Level:  Standard Operating Procedure Organisation level: 4-Laboratory Site
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Responsible: 6_QAD4

F.

12) QUALITY CONTROL REQUIREMENTS

A. QC requirements include the Initial Demonstration of Capability (IDC) and ongoing QC requirements that must be met when preparing and analyzing Field Samples. This section describes the QC parameters, their required frequencies, and the performance criteria that must be met in order to meet EPA quality objectives. The QC criteria discussed in the following sections are summarized in Attachment VI. These QC requirements are considered the minimum acceptable QC criteria. Laboratories are encouraged to institute additional QC practices to meet their specific needs.

1. METHOD MODIFICATIONS – The analyst is permitted to modify LC columns, LC conditions, evaporation techniques, internal standards or surrogate standards, and MS and MS/MS conditions. Each time such method modifications are made, the analyst must repeat the procedures of the IDC. **Modifications to LC conditions should still produce conditions such that co-elution of the method analytes is minimized to reduce the probability of suppression/enhancement effects.**

B. INITIAL DEMONSTRATION OF CAPABILITY – The IDC must be successfully performed prior to analyzing any Field Samples. Prior to conducting the IDC, the analyst must first generate an acceptable Initial Calibration following the procedure outlined below.

 Document number: LCMS-SOP24130 Old Reference: Version: 6 Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS) Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Level:  Standard Operating Procedure Organisation level: 4-Laboratory Site Responsible: 6_QAD4

1. INITIAL DEMONSTRATION OF LOW SYSTEM BACKGROUND – Any time a new lot of SPE cartridges, solvents, centrifuge tubes, disposable pipets, and autosampler vials are used, it must be demonstrated that an LRB/MBLK is reasonably free of contamination and that the criteria are met. If an automated extraction system is used, an LRB/MBLK should be extracted on each port to ensure that all the valves and tubing are free from potential PFAS contamination. Acceptance limit is < 1/3 MRL.

2. INITIAL DEMONSTRATION OF PRECISION (IDP) – Prepare, extract, and analyze four to seven replicate LFB/LCSs fortified near the mid range of the initial calibration curve. Sample preservatives as described in Section 8 must be added to these samples. The relative standard deviation (RSD) of the results of the replicate analyses must be less than 20%.

3. INITIAL DEMONSTRATION OF ACCURACY (IDA) – Using the same set of replicate data generated calculate average recovery. The average recovery of the replicate values must be within ± 30% of the true value.

4. INITIAL DEMONSTRATION OF PEAK ASYMMETRY FACTOR – Peak asymmetry factors must be calculated using the equation in Section 11 for the first two eluting peaks (if only two analytes are being analyzed, both must be evaluated) in a mid-level CAL standard. The peak asymmetry factors must fall in the range of 0.8 to 1.5.

5. MINIMUM REPORTING LEVEL (MRL) CONFIRMATION – 2ppt is established as an MRL for all compounds for this method based on the intended use of the method. The MRL may be established by a laboratory for their specific purpose or may be set by a regulatory agency. Establish an Initial Calibration following the procedure outlined in Section 11. The lowest CAL standard used to establish the Initial Calibration must be at or below the concentration of the MRL. Establishing the MRL concentration too low may cause repeated failure of ongoing QC requirements. Confirm the MRL following the procedure outlined below.

- a. Fortify, extract, and analyze seven replicate LFB/LCSs at the proposed MRL concentration. These LFBs must contain all method preservatives described in Section 8. Calculate the mean measured concentration (*Mean*) and standard deviation for these replicates. Determine the Half Range for the prediction interval of results (HR_{PIR}) using the equation below

$$HR_{PIR} = 3.963s$$

where

s = the standard deviation

3.963 = a constant value for seven replicates.¹

- b. Confirm that the upper and lower limits for the Prediction Interval of Result ($PIR = Mean \pm HR_{PIR}$) meet the upper and lower recovery limits as shown below

The Upper PIR Limit must be =150% recovery.

$$\frac{Mean + HR_{PIR}}{FortifiedConcentration} \times 100\% \leq 150\%$$

 Document number: LCMS-SOP24130 Old Reference: Version: 6 Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS) Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Level:  Standard Operating Procedure Organisation level: 4-Laboratory Site Responsible: 6_QAD4

The Lower PIR Limit must be =50% recovery.

$$\frac{Mean - HR_{PIR}}{FortifiedConcentration} \times 100\% \geq 50\%$$

c. The MRL is validated if both the Upper and Lower PIR Limits meet the criteria described above (Sect. 11). If these criteria are not met, the MRL has been set too low and must be determined again at a higher concentration.

6. CALIBRATION CONFIRMATION – Analyze a QCS as described in Section 11 to confirm the accuracy of the standards/calibration curve.

7. DETECTION LIMIT DETERMINATION – *It is required by various regulatory bodies associated with compliance monitoring. See Section 16 for 40 CFR, part 136, Appendix B, Rev 2 MDL Procedure.*

a. Replicate analyses for this procedure must be done over at least three days (i.e., both the sample extraction and the LC/MS/MS analyses must be done over at least three days). Prepare at least seven replicate LFB/LCSs at a concentration estimated to be near the DL. This concentration may be estimated by selecting a concentration at 2.5 times the noise level. The DLs in Attachment I were calculated from LFB/LCSs fortified at various concentrations as indicated in the table. The appropriate fortification concentrations will be dependent upon the sensitivity of the LC/MS/MS system used. All preservation reagents listed in Section 8 must also be added to these samples. Analyze the seven replicates through all steps.

NOTE: If an MRL confirmation data set meets these requirements, a DL may be calculated from the MRL confirmation data, and no additional analyses are necessary.

Calculate the MDL using the following equation

$$DL = s \times t_{(n-1, 1-\alpha=0.99)}$$

Where:

s = standard deviation of replicate analyses

$t_{(n-1, 1-\alpha=0.99)}$ = Student's t value for the 99% confidence level with $n-1$ degrees of freedom; 3.143 for $n = 7$.

n = number of replicates.

NOTE: Do not subtract blank values when performing DL calculations. The MDL is a statistical determination of precision only. If the DL replicates are fortified at a low enough concentration, it is likely that they will not meet the precision and accuracy criteria for CCCs, and may result in a calculated MDL that is higher than the fortified concentration. Therefore, no precision and accuracy criteria are specified.

8. If a laboratory is establishing their own MRL, the calculated MDLs should not be used as the MRL for analytes that commonly occur as background contaminants. Method analytes that are seen in the background should be reported as present in Field

 Document number: LCMS-SOP24130 Old Reference: Version: 6 Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS) Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Level:  Standard Operating Procedure Organisation level: 4-Laboratory Site Responsible: 6_QAD4

Samples, only after careful evaluation of the background levels. It is recommended that a MRL be established at the mean LRB concentrations + 3s or 3 times the mean LRB/MBLK concentration, whichever is greater. This value should be calculated over a period of time, to reflect variability in the blank measurements. It is recommended that this value be used as an MRL in order to avoid reporting false positive results.

C. ONGOING QC REQUIREMENTS – This section summarizes the ongoing QC criteria that must be followed when processing and analyzing Field Samples.

1. LABORATORY REAGENT BLANK (LRB/MBLK)(MB, MBL, etc in TALS) – An LRB/MBLK is required with each extraction batch to confirm that potential background contaminants are not interfering with the identification or quantitation of method analytes. If more than 20 Field Samples are included in a batch, analyze an LRB/MBLK for every 20 samples. If the LRB/MBLK produces a peak within the retention time window of any analyte that would prevent the determination of that analyte, determine the source of contamination and eliminate the interference before processing samples. Background contamination must be reduced to an acceptable level before proceeding. Background from method analytes or other contaminants that interfere with the measurement of method analytes must be below 1/3 of the MRL. Blank contamination is estimated by extrapolation, if the concentration is below the lowest CAL standard. This extrapolation procedure is not allowed for sample results as it may not meet data quality objectives. If the method analytes are detected in the LRB at concentrations equal to or greater than this level, then all data for the problem analyte(s) must be considered invalid for all samples in the extraction batch. Because background contamination is a significant problem for several method analytes, it is highly recommended that the analyst maintain a historical record of LRB data.

2. CONTINUING CALIBRATION CHECK (CCC)(CCV, CCVIS, etc in TALS) – Low-level CCC Standard is analyzed at the beginning of each analysis batch before field samples, and the recoveries shall fall within 50-150%. Mid-level CCC and high-level CCC are analyzed after every 10 Field Samples and at the end of the analysis batch. Their recoveries shall fall within 70-130%.

3. LABORATORY FORTIFIED BLANK (LFB/LCS)(LLCS, etc in TALS) – An LFB/LCS is required with each extraction batch. The fortification concentration of the LFB/LCS must be rotated between medium and high concentration from batch to batch. In addition, one LFB/LCS at low concentration must also be extracted per day. The low concentration LFB/LCS must be as near as practical to, but no more than two times, the MRL. Similarly, the high concentration LFB/LCS should be near the high end of the calibration range established during the initial calibration. Results of the low-level LFB/LCS analyses must be 50-150% of the true value. Results of the medium and high-level LFB/LCS analyses must be 70-130% of the true value. If the LFB results do not meet these criteria for method analytes, then all data for the problem analyte(s) must be considered invalid for all samples in the extraction batch.

4. INTERNAL STANDARDS (IS) – The analyst must monitor the peak areas of the IS(s) in all injections during each analysis day. The IS responses (peak areas) in any chromatographic run must be within 70-140% of the response in the most recent CCC and must not deviate by more than 50% from the average area measured during initial analyte calibration. If the IS areas in a chromatographic run do not meet these criteria, inject a second aliquot of that extract aliquotted into a new capped autosampler vial.

 Document number: LCMS-SOP24130 Old Reference: Version: 6 Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS) Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Level:  Standard Operating Procedure Organisation level: 4-Laboratory Site Responsible: 6_QAD4

Random evaporation losses have been observed with the polypropylene caps causing high IS(s) areas.

- a. If the reinjected aliquot produces an acceptable IS response, report results for that aliquot.
- b. If the reinjected extract fails again, the analyst should check the calibration by reanalyzing the most recently acceptable CAL standard. If the CAL standard fails the criteria of Section 9, recalibration is in order per Section 9. If the CAL standard is acceptable, extraction of the sample may need to be repeated provided the sample is still within the holding time. Otherwise, report results obtained from the reinjected extract, but annotate as suspect. Alternatively, collect a new sample and re-analyze.

5. SURROGATE RECOVERY – The SUR standard is fortified into all samples, CCCs, LRBs, LFBs, LFSMs, LFSMDs, FD, and FRB prior to extraction see attachment for concentration. It is also added to the CAL standards. The SUR is a means of assessing method performance from extraction to final chromatographic measurement. Calculate the recovery (%R) for the SUR using the following equation

$$\%R = \left(\frac{A}{B} \right) \times 100$$

Where:

A = calculated surrogate concentration for the QC or field sample, and

B = fortified concentration of the surrogate.

SUR recovery must be in the range of 70-130%. When SUR recovery from a sample, blank, or CCC is less than 70% or greater than 130%, check 1) calculations to locate possible errors, 2) standard solutions for degradation, 3) contamination, and 4) instrument performance. Correct the problem and reanalyze the extract.

- a. If the extract reanalysis meets the SUR recovery criterion, report only data for the reanalyzed extract.
- b. If the extract reanalysis fails the 70-130% recovery criterion, the analyst should check the calibration by injecting the last CAL standard that passed. If the CAL standard fails the criteria of this SOP, recalibration is in order. If the CAL standard is acceptable, extraction of the sample should be repeated provided the sample is still within the holding time. If the re-extracted sample also fails the recovery criterion, report all data for that sample as suspect/SUR recovery to inform the data user that the results are suspect due to SUR recovery. Alternatively, collect a new sample and re-analyze.

6. LABORATORY FORTIFIED SAMPLE MATRIX (LFSM/MS) – Analysis of an LFSM/MS is required in each extraction batch and is used to determine that the sample matrix does not adversely affect method accuracy. Assessment of method precision is accomplished by analysis of a Field Duplicate (FD); however, infrequent occurrence of method analytes

	Always check on-line for validity.	Level:  Standard Operating Procedure
Document number: LCMS-SOP24130	EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Organisation level: 4-Laboratory Site
Old Reference:		Responsible: 6_QAD4
Version: 6		
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	

would hinder this assessment. If the occurrence of method analytes in the samples is infrequent, or if historical trends are unavailable, a second LFSM/MS, or LFSMD/MSD, must be prepared, extracted, and analyzed from a duplicate of the Field Sample. Extraction batches that contain LFSMD/MSDs will not require the extraction of a FD/DUP. If a variety of different sample matrices are analyzed regularly, for example, drinking water from groundwater and surface water sources, method performance should be established for each. Over time, LFSM/MS data should be documented by the laboratory for all routine sample sources.

a. Within each extraction batch, a minimum of one Field Sample (FS) is fortified as an LFSM/MS. The LFSM/MS is prepared by spiking a sample with an appropriate amount of the Analyte PDS. Select a spiking concentration that is greater than or equal to the matrix background concentration, if known. Use historical data and rotate through the low, mid and high concentrations when selecting a fortifying concentration.

b. Calculate the percent recovery (%R) for each analyte using the equation

$$R = \frac{(A - B)}{C} \times 100$$

where :

A = measured concentration in the fortified sample

B = measured concentration in the unfortified sample

C = fortification concentration.

c. Analyte recoveries may exhibit matrix bias. For samples fortified at or above their native concentration, recoveries should range between 70 - 130%, except for low-level fortification near or at the MRL (within a factor of 2 times the MRL concentration) where 50-150% recoveries are acceptable. If the accuracy of any analyte falls outside the designated range, and the laboratory performance for that analyte is shown to be in control in the CCCs, the recovery is judged to be matrix biased. The result for that analyte in the unfortified sample is labeled suspect/matrix to inform the data user that the results are suspect due to matrix effects.

7. FIELD DUPLICATE OR LABORATORY FORTIFIED SAMPLE MATRIX DUPLICATE (FD/DUP or LFSMD/MSD) – Within each extraction batch, a minimum of one FD/DUP or LFSMD/MSD must be analyzed. Duplicates check the precision associated with sample collection, preservation, storage, and laboratory procedures. If method analytes are not routinely observed in Field Samples, an LFSMD/MSD should be analyzed rather than an FD. Note that field duplicates must be reported for CA samples.

a. Calculate the relative percent difference (RPD) for duplicate measurements (FD1 and FD2) using the equation

$$RPD = \frac{|FD1 - FD2|}{(FD1 + FD2)/2} \times 100$$

 Document number: LCMS-SOP24130 Old Reference: Version: 6 Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS) Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Level:  Standard Operating Procedure Organisation level: 4-Laboratory Site Responsible: 6_QAD4

b. RPDs for FDs should be ≤ 30%. Greater variability may be observed when FDs have analyte concentrations that are within a factor of 2 of the MRL. At these concentrations, FDs should have RPDs that are ≤ 50%. If the RPD of any analyte falls outside the designated range, and the laboratory performance for that analyte is shown to be in control in the CCC, the recovery is judged to be matrix biased. The result for that analyte in the unfortified sample is labeled suspect/matrix to inform the data user that the results are suspect due to matrix effects.

c. If an LFSMD is analyzed instead of a FD, calculate the relative percent difference (RPD) for duplicate LFSMs (LFSM and LFSMD) using the equation

$$RPD = \left(\frac{|LFSM - LFSMD|}{\frac{(LFSM + LFSMD)}{2}} \right) \times 100\%$$

d. RPDs for duplicate LFSMs should be ≤ 30% for samples fortified at or above their native concentration. Greater variability may be observed when LFSMs are fortified at analyte concentrations that are within a factor of 2 of the MRL. LFSMs fortified at these concentrations should have RPDs that are ≤ 50% for samples fortified at or above their native concentration. If the RPD of any analyte falls outside the designated range, and the laboratory performance for that analyte is shown to be in control in the CCC, the recovery is judged to be matrix biased. The result for that analyte in the unfortified sample is labeled suspect/matrix to inform the data user that the results are suspect due to matrix effects.

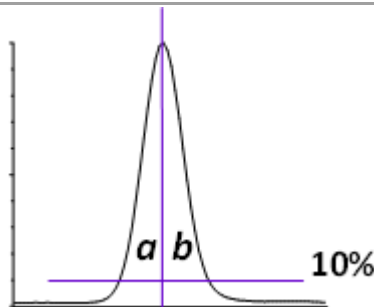
8. FIELD REAGENT BLANK (FRB) or FIELD BLANK – FRB acceptance criteria is < 1/3 MRL. The purpose of the FRB is to ensure that PFAAs measured in the Field Samples were not inadvertently introduced into the sample during sample collection/handling. Analysis of the FRB is required only if a Field Sample contains a method analyte or analytes at or above the MRL. The FRB is processed, extracted and analyzed in exactly the same manner as a Field Sample. A FRB must have hits ≤ to the MRL to be considered acceptable. Samples with hits in a FRB over the MRL will invalidate the associated sample and a resample must be requested. Other QC requirements (Surrogate, internal) are identical to a typical sample. Note that, when analyzed, Field Reagent Blanks must be reported for CA samples.

TRIP BLANK - Trip blank acceptance criteria is < 1/3 MRL. This sample is to evaluate potential contamination from sample shipping and handling procedures. See section 8.G of the SOP for more details.

9. PEAK ASYMMETRY FACTOR – A peak asymmetry factor must be calculated using the equation below during the IDL and every time chromatographic changes are made that may affect peak shape. The peak asymmetry factor for the first two eluting peaks in a mid-level CAL standard (if only two analytes are being analyzed, both must be evaluated) must fall in the range of 0.8 to 1.5. **Modifying the standard or extract composition to more aqueous content to prevent poor shape is not permitted.**

 Document number: LCMS-SOP24130 Old Reference: Version: 6	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Level:  Standard Operating Procedure Organisation level: 4-Laboratory Site
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Responsible: 6_QAD4

$$A_s = \frac{b}{a}$$



where:

A_s = peak asymmetry factor

b = width of the back half of the peak measured (at 10% peak height) from the trailing edge of the peak to a line dropped perpendicularly from the peak apex

a = the width of the front half of the peak measured (at 10% peak height) from the leading edge of the peak to a line dropped perpendicularly from the apex.

10. QUALITY CONTROL SAMPLES (QCS) (ICV, etc in TALS) – As part of the IDC each time a new Analyte PDS is prepared, and at least quarterly, analyze a QCS sample from a source different from the source of the CAL standards. If a second vendor is not available, then a different lot of the standard should be used. The QCS should be prepared and analyzed just like a CCC. Acceptance criteria for the QCS are identical to the CCCs; the calculated amount for each analyte must be $\pm 30\%$ of the expected value. If measured analyte concentrations are not of acceptable accuracy, check the entire analytical procedure to locate and correct the problem.

D. Contingencies for handling out of control data or unacceptable data.

1. Submit a **Non-Conformance Memo (NCM)** for QC out of specification for samples that cannot be re-extracted. Generate JIRA CAR when there is a pattern or trend in the failed batch or instrument QC.
2. Details regarding use of the **NCM** can be found in the **Absorb Training database**.
3. See **Policy-QA-QP3861** for a list of data qualifiers and their definitions.

E. For each batch complete a [537.1 QC Checklist](#)

12) CALCULATIONS

A. DATA ANALYSIS AND CALCULATION

1. Complete chromatographic resolution is not necessary for accurate and precise measurements of analyte concentrations using MS/MS. In validating this method, concentrations were calculated by measuring the product ions. Other ions may be selected at the discretion of the analyst.
2. Calculate analyte and SUR concentrations using the multipoint calibration established. Do not use daily calibration verification data to quantitate analytes in samples. Adjust

	Always check on-line for validity.	Level:  Standard Operating Procedure
Document number: LCMS-SOP24130	EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Organisation level: 4-Laboratory Site
Old Reference:		Responsible: 6_QAD4
Version: 6		
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	

final analyte concentrations to reflect the actual sample volume used for extraction.

3. Prior to reporting the data, the chromatogram should be reviewed for any incorrect peak identification or poor integration.

4. Calculations must utilize all available digits of precision, but final reported concentrations should be rounded to an appropriate number of significant figures (one digit of uncertainty), typically two, and not more than three significant figures.

13) METHOD PERFORMANCE

A. Attachment I, Documentation of Demonstration of Precision and Accuracy

B. Attachment II, Documentation of Method Detection Limit Study

C. The above Attachments are current studies at the time of SOP approval only to demonstrate method performance. A more recent study is available for review upon request.

14) REFERENCES

A. EPA Method 537.1: Determination of Selected Per- and Polyfluorinated Alkyl substances in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS), Version 2.0, EPA Document No. EPA/600/R-18/352, Office of Research and Development, March 2020 .

B. EPA, EPA/815-R-05-004, Manual for the Certification of Laboratory Analyzing Drinking Water, 5th edition, January, 2005.

C. CA State Water Resources Control Board Division of Drinking Water, Drinking Water Sample Collection for Per and Polyfluorinated Alkyl Substances (PFAS) Sampling Guidance, March 2019.

D. Quality Systems for Chemical Testing. Volume 1 Module 4. 2016 TNI Standards, November 1, 2017.

E. Management and Technical Requirements for Environmental Laboratories. Volume 1 Module 2. 2016 TNI Standards, Revision 2.1, December 6, 2016.

F. US Code of Federal Regulations 40 CFR 136 MUR 2017 Appendix B-Definition and Procedure For The Determination of the Method Detection Limit Revision 2 December 2017.

15) DEVIATIONS FROM REFERENCED METHODOLOGY

A. No major deviation - using online WAX SPE as trap column for HPLC contamination control.

B. Use of a Teflon transfer tube system instead of polypropylene tube system.

C. Section 06 - Lab uses polypropylene insert in 1.8 ml amber vials instead of PP vials and caps. The method blank contamination, if any, is very negligent less than 1/3 of MRL 2 ppt.

	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Level:  Standard Operating Procedure
Document number: LCMS-SOP24130		
Old Reference:		
Version: 6		Organisation level: 4-Laboratory Site
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Responsible: 6_QAD4

D. For FRB, a preserved reagent water and unpreserved/empty bottle is sent to sampling site instead of Unpreserved reagent water/preserved bottle. Therefore EEA is technically following 537.1 rev 1 for FRB preparation due to a typo in rev 2. See Attachment XII for documentation of this issue from EPA.

16) METHOD DETECTION LIMIT

A. See section 11.B.7 of the SOP for Detection Limit Determination method requirements.

B. For general MDL procedure and requirements from 40 CFR 136, Appendix B Revision 2, see Work instructions [QA-WI25066](#).

17) DEMONSTRATION OF CAPABILITY

A. For general Chemistry Initial and Continuing Demonstration of Capability requirements from the 2016 TNI Standard, see Work instructions [QA-WI25084](#).

B. Refer to method specific requirements in section 11.B of the SOP for Initial Demonstration of Precision (IDP) and Initial Demonstration of Accuracy (IDA). Acceptance limits 70-130% recovery for accuracy and <20% RSD for Precision.

18) DEFINITIONS

Refer to the [WI26472](#), Glossary for a complete list of terms and definitions.

19) POLLUTION PREVENTION

A. This method utilizes SPE to extract analytes from water. It requires the use of very small volumes of organic solvent and very small quantities of pure analytes, thereby minimizing the potential hazards to both the analyst and the environment as compared to the use of large volumes of organic solvents in conventional liquid-liquid extractions.

B. For information about pollution prevention that may be applicable to laboratory operations, consult "Less is Better: Laboratory Chemical Management for Waste Reduction" available from the American Chemical Society's Department of Government Relations and Science Policy, 1155 16th Street N.W., Washington, D.C., 20036.

20) WASTE MANAGEMENT

A. It is the responsibility of the laboratory to determine whether its wastes are hazardous and to assure safe handling and disposal. The laboratory works closely with the Treatment, Storage, and Disposal Facility to ensure that certain wastes are recycled where possible, that the source of waste is reduced to the lowest possible level and that stringent land disposal restrictions are followed.

B. Refer to the following documents for additional information regarding waste management:

1. Hazardous Waste Management and Sample Disposal Procedures
2. Resource Conservation and Recovery Act (RCRA)-Title 40 of the Code of Federal Regulations, Parts 260 through 270 (40 CFR 260-270)
3. California Hazardous Waste Control Law (HWCL)-CCR Title 22 where 40 CFR was duplicated into CCR Title 22, Parts 66260-66270.

	Always check on-line for validity.	Level:  Standard Operating Procedure
Document number: LCMS-SOP24130	EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Organisation level: 4-Laboratory Site
Old Reference:		Responsible: 6_QAD4
Version: 6		
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	

21) REVISIONS

A. Revision 1.0
New SOP

- B. Revision 2.0 (EY3P 05/02/19, 05/07/19, and 05/10/19)
1. Section 8.E. - Added pH does not meet acceptance criteria, sample is rejected and updated Free Chlorine criteria.
 2. Section 8.G - Removed trip blank since not required by CA
 3. Section 10.C - added manual extract procedure
 4. Section 11.D - Updated from a calibration curve is generated to chromatographic changes are made that may affect peak shape.
 5. Section 16 - Added link to MDL requirements Work Instructions.
 6. Section 17 - Added link to IDC requirements Work Instructions.
 7. Added Attachment X, Procedure for pH and Free Chlorine Checks
 8. All attachments - Updated header with current SOP information.
 9. Section 2.C.1 - changed is to are
 10. Section 9.B.1 - changed ? to to
 11. Section 10.C.3 - added Automated SPE program setting
 12. Section 11.B.7.a - changed ? to a

- C. Revision 3.0 (KAM 07/09/19)
1. Section 9B.3 - Changed from minimum of five points calibration to six points.
 2. Section 22C - update calibration table to include 60ppt in the curve.
 3. Section 22C - update calibration table to include levels for CCCM and CCCH.

(ARH 12/09/19)

1. Table 1 - Corrected the name and the concentration of the following analytes - PFBS, PFHxS, Gen X, 11CI-PF3ONS, 11CI-PF3OUdS and PFOS.
2. Added line 7.N.4.

(Revised 12/10/19 - MFR)

1. Table 1 - Revised analyte name to match reference method. Updated MRL to 2ng/L
2. Revised Section 7 for standard preparation
3. Revised attachment table of contents, add most recent MDL and PIR study, detailed standard preparation procedure

(12/17/19 RDL)

4. Section 7.B - Replaced purified with nano-pure
5. Section 7.N.3 - Deleted "During method development, the concentrations of the SUR(s) were 10pg/μL in the standard (10ng/L in the sample) and the IS(s) were 10pg/μL. The lowest concentration CAL standard must be at or below the MRL, which may depend on system sensitivity."
6. Sections 10 -11 - Renumbered several items for clarity.

(12/17/19 MFR)

1. Section 2.C.2 Deleted "DL detection was not performed at EEA for this method"

 Document number: LCMS-SOP24130 Old Reference: Version: 6 Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Always check on-line for validity. EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS) Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	Level:  Standard Operating Procedure Organisation level: 4-Laboratory Site Responsible: 6_QAD4

(12/18/19 - UAON)

1. multiple sections - update from EEA to EEA-M
2. Section 6 - added " All instrumentation/apparatus listed are critical consumables for this method. Stock is kept on hand to complete analysis." as per ISO requirement
3. Section 7 - added "All reagents and standards listed are critical consumables for this method. Stock is kept on hand to complete analysis" as per ISO requirement.

Revision 4.0

(11/03/2020)

1. Changed reference to 537.1 version 2.0
2. Section 2.A table 1. Updated GenX MRL to 2ng/L
3. Added link to MSDS online
4. Updated catalog numbers on sections 6 and 7
5. Updated instrument list
6. Updated expiration of 20mM Ammonium acetate (Section 7.F)
7. Updated attachments with most recent MDL/DOC as well as added attachments XI and XII

Revision 5.0 (6/21/21 URED)

1. Updated Section 7) J. K.1., L.2., M.2., to note PDSs are good for one month.

Revision 6.0 (11/7/22 URED)

1. Updated reference links throughout the document to the new server location
2. Updated QC notations to note TALS equivalents
3. Removed STARLIMS and Webforms notations to TALS
4. Updated Attachments I, II, and III with most recent examples
5. Updated Section 9.B.2.b to note proper integration of branched isomers.
6. Updated Section 11. D. with regards to NCM use, removing QIR notations.
7. Removed references to EEA-M in Section 05)B., simply noting the SOP with a link.

22) ATTACHMENTS

- A. Attachment I, Documentation of Demonstration of Method Detection Limit Study
- B. Attachment II, Documentation of Demonstration of Precision and Accuracy
- C. Attachment III, Documentation of Minimum Reporting Limit Study
- D. Attachment IV, Standard Preparation
- E. Attachment V, Instrument Conditions
- F. Attachment VI, Method QC Summary
- G. Attachment VII, Analytical Sequence
- H. Attachment VIII, QA/QC Requirements for Method 537.1
- I. Attachment IX, Caliper Lifesciences Automated SPE Workstation Conditions/Parameters
- J. Attachment X, Procedure for pH and Free Chlorine Checks

	Always check on-line for validity.	Level: 
	EPA 537.1 - Determination of Selected Per- and polyfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)	Standard Operating Procedure
		Organisation level: 4-Laboratory Site
		Responsible: 6_QAD4
Document number: LCMS-SOP24130 Old Reference:		
Version: 6		
Approved by: AZV7, UNCO, Y7BM Effective Date: 23-JAN-2023	Document users: 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_LCMS, 4_Eurofins_EUUSMO_Eaton_Analytical_Monrovia/6_Extract	

K. Attachment XI, EPA email with regards to Trizma preservation

L. Attachment XII, EPA email with regards to field blanks

Attachment:
[24130_537.1_attachement-v6..11.7.22.pdf \(.pdf\)](#)

End of document

Version history

Version	Approval	Revision information	
4	29.DEC.2020		
5	22.JUN.2021		
6	18.JAN.2023		

ATTACHMENTS

- A. Attachment I, Documentation of Method Detection Limit Study
- B. Attachment II, Documentation of Demonstration of Precision and Accuracy
- C. Attachment III, Documentation of Minimum Reporting Limit
- D. Attachment IV, Standard Preparation
- E. Attachment V, Instrument Conditions
- F. Attachment VI, Method QC Summary
- G. Attachment VII, Analytical Sequence
- H. Attachment VIII, QA/QC Requirements for Method 537.1
- I. Attachment IX, Caliper Lifesciences Automated SPE Workstation Conditions/Parameters
- J. Attachment X, Procedure for pH and Free Chlorine Checks
- K. Attachment XI, EPA email with regards to Trizma preservation
- L. Attachment XII, EPA email with regards to Field Blanks

Attachment I, Documentation of Method Detection Limit Study

Method: @537.1
Instrument ID: LCMS11
Date Reported: 01/26/22
Units: ng/L
SOP ID: 24130
SOP Revision: 2

MDL Study

Compound Name	Spike Value	Analysis Date/Time	1/25/2022 18:14	1/25/2022 18:34	3/15/2022 18:33	1/26/2022 7:47	1/26/2022 8:06	1/27/2022 17:06	1/27/2022 17:17
11CL-PP3OUBS	1.884	2	1.80	1.77	1.94	1.80	1.88	1.97	1.98
9CL-PP3OUBS	1.864	2	1.77	1.67	1.91	1.87	1.86	1.92	2.03
ADONA	1.88	2	2.00	2.06	2.06	2.00	2.10	2.00	1.91
GenX	2	2	1.89	2.09	2.22	1.99	2.16	2.12	2.19
N-ElFOSAA	2	2	1.87	1.99	2.19	1.94	2.11	2.08	2.27
N-MeFOSAA	2	2	1.85	1.97	2.04	1.95	2.13	2.11	2.16
PFBS	1.77	2	1.88	1.88	1.94	1.94	1.93	1.93	1.98
PFDA	2	2	1.98	2.03	2.07	2.04	2.07	2.11	2.16
PFDA	2	2	1.80	1.93	2.05	2.04	2.03	2.04	2.09
PFHBA	2	2	2.30	2.24	2.38	2.19	2.24	2.25	2.39
PFHBA	2	2	2.23	2.34	2.26	2.17	2.14	2.27	2.30
PFHAs	1.824	2	1.99	1.94	1.94	1.90	2.07	2.05	2.13
PFNA	2	2	1.96	2.14	2.15	2.21	2.28	2.16	2.34
PFDA	2	2	2.18	2.21	2.44	2.26	2.27	2.20	2.32
PFOS	1.851	8	1.78	1.90	1.96	1.84	1.97	1.94	1.91
PFTA	2	2	1.68	1.76	1.86	1.84	2.31	2.30	2.37
PFTA	2	2	1.87	1.91	1.98	2.04	2.11	2.06	2.12
PFUAA	2	2	1.79	1.87	2.00	1.97	1.99	1.98	2.03

ave %rec %stdv %MDL

1.87714 98.6% 0.086 4.7% 0.277

1.89429 100.0% 0.115 6.2% 0.362

2.02143 107.5% 0.064 3.2% 0.202

2.09429 104.7% 0.117 5.8% 0.369

2.07286 103.6% 0.146 7.0% 0.459

2.04429 102.2% 0.090 4.4% 0.282

1.92286 100.6% 0.040 2.1% 0.127

2.00571 100.3% 0.008 2.8% 0.152

1.99714 99.9% 0.100 5.0% 0.313

2.28429 114.2% 0.076 3.3% 0.239

2.24429 112.2% 0.070 3.1% 0.221

2.01714 110.6% 0.072 3.5% 0.225

2.17714 108.9% 0.121 5.8% 0.380

2.27714 113.9% 0.085 3.7% 0.266

1.9 102.6% 0.009 9.6% 0.215

2.03429 101.7% 0.287 14.1% 0.901

2.01286 100.6% 0.097 4.8% 0.303

1.94714 97.4% 0.085 4.4% 0.268

autofrac
1/26/22

LCMS-FD-FRM05416
Ver 3.0 (01/27/20)

Attachment I, Documentation of Method Detection Limit Study (cont)

Method: 8537.1
Instrument ID: LCM511
Date Reported: 01/20/22
Units: ng/L
SOP ID: 24130
SOP Revision: 2

Blank Results

Analysis Date/Time	1/25/2022 17:44	1/25/2022 17:55	1/25/2022 18:04	1/26/2022 7:17	1/26/2022 7:28	1/27/2022 16:47	1/27/2022 16:58
Extraction Date	01/19/22	01/19/22	01/19/22	01/21/22	01/21/22	01/25/22	01/25/22
Extraction Batch	1380424	1380424	1380424	1381124	1381124	1381617	1381617
LCMS Analyst	KAM	KAM	KAM	KAM	KAM	KAM	KAM
Extraction Method	SV4R	SV4R	SV4R	SV4R	SV4R	SV4R	SV4R
Sample Name	202201180391	202201180390	202201180391	202201180392	202201180393	202201180395	202201180396
Compound Name	StartLine MRL	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6
11CL PF3OLMS	2	0	0	0	0	0	0
11CL PF3OLMS	2	0	0	0	0	0	0
ADONA	2	0.02	0.08	0.03	0.05	0.02	0.02
GenX	2	0	0	0	0	0	0
N-ethyl-OSAA	2	0.09	0	0	0	0	0
N-methyl-OSAA	2	0.11	0	0	0	0	0
PFBS	2	0	0	0	0	0	0
PFTA	2	0.03	0.02	0	0	0.03	0.03
PFTDA	2	0	0	0	0	0	0
PFTBA	2	0.07	0.06	0.05	0.07	0.04	0.04
PFTHA	2	0.07	0.07	0.06	0.1	0.06	0.07
PFTAS	2	0	0	0	0	0	0
PFTNA	2	0.02	0	0	0	0	0
PFOA	2	0.13	0.12	0.14	0.13	0.12	0.11
PFOS	2	0.06	0	0	0	0	0
PFTA	2	0.14	0.11	0.13	0.18	0.17	0.22
PFTDA	2	0	0	0	0	0	0
PFTHA	2	0	0	0	0	0	0

MUST USE THE HIGHER OF MDL_{ave} (above) and MDL₉ (below)

ave	stddev	%RSD	MDL _{ave}	MDL ₉
0	0.000	0.00%	0.000	0.000
0	0.000	0.00%	0.000	0.000
0.02743	0.011	40.2%	0.300	0.000
0	0.000	0.00%	0.460	0.000
0.01286	0.034	264.6%	0.000	0.446
0.01071	0.042	384.6%	0.000	0.000
0	0.000	0.00%	0.390	0.000
0.01857	0.020	105.1%	0.000	0.000
0	0.000	0.00%	0.000	0.000
0.056971	0.011	20.4%	0.370	0.040
0.07743	0.013	16.8%	0.500	0.114
0	0.000	0.00%	0.230	0.000
0.00286	0.008	284.6%	0.150	0.000
0.12571	0.010	7.8%	0.250	0.156
0.00857	0.023	264.6%	0.120	0.000
0.16857	0.053	31.7%	0.160	0.336
0	0.000	0.00%	0.000	0.000
0	0.000	0.00%	0.000	0.000

Handwritten notes: 0.03, 0.09, 0.05, 0.02, 0.06

LCMS-EO-FRM21416
Ver 3.0 (01/27/20)

2 of 3

Attachment II, Documentation of Demonstration of Precision and Accuracy

Method: 0537.1
 Instrument ID: LCM511
 Date Reported: 01/23/22
 Units: ng/L
 SOP ID: 24130
 SOP Revision: 2

		IDOC									
Analysis Date	1/24/2022 7:38	1/24/2022 8:15	1/24/2022 8:38	1/24/2022 8:57	1/24/2022 9:07						
Extraction Date	01/21/22	01/21/22	01/21/22	01/21/22	01/21/22						
Extraction Batch	1381124	1381124	1381124	1381124	1381124						
LCMS Analyst	KAM	KAM	KAM	KAM	KAM						
Extraction Analyst	SV4R	SV4R	SV4R	SV4R	SV4R						
Sample Name	202201230386	202201230387	202201230388	202201230389	202201230390	202201230391	202201230392				
Compound Name	True Value	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Ave	Ave %RSD	%RSD
1,1,1-TCF3C005	23.35	17.77	22.37	21.62	21.98	22.04	22.39	22.24	21.4871	91.34%	1.66006
1,1,1-TCF3C005	23.3	17.07	22.42	22.5	21.76	22.19	21.3	22.34	21.4643	92.08%	1.72192
1,1,1-TCF3C005	23.5	18.72	23.51	23.84	23.62	24.1	23.05	23.68	22.9443	97.64%	1.89182
1,1,1-TCF3C005	25	19.17	24.63	25.18	24.05	25.23	24.38	24.62	23.6943	95.58%	2.12462
1,1,1-TCF3C005	25	19.34	22.87	22.63	22.49	22.72	22.76	23.69	22.2571	89.09%	1.78781
1,1,1-TCF3C005	25	19.48	23.98	24.53	23.87	23.2	24.12	24.58	23.3957	93.46%	1.76688
1,1,1-TCF3C005	22.125	18.23	22.23	20.88	21.97	20.95	22.62	22.24	21.3171	94.33%	1.92485
1,1,1-TCF3C005	25	18.87	23.28	24.45	24.06	24.61	23.88	23.79	23.28	93.12%	1.95957
1,1,1-TCF3C005	25	18.42	23.16	23.82	23.58	22.68	22.98	23.89	22.6486	93.59%	1.91637
1,1,1-TCF3C005	25	19.58	25.13	25.72	24.99	26.42	25.38	25.6	24.69	94.70%	2.301
1,1,1-TCF3C005	25	19.88	25.37	25.47	25.3	24.76	25.14	25.47	24.9857	94.39%	2.1847
1,1,1-TCF3C005	22.8	18.53	24.04	23.59	23.26	23.65	23.98	23.67	22.8866	100.67%	1.81383
1,1,1-TCF3C005	25	19.3	24.77	25.17	24.9	25.51	24.54	24.95	24.1914	96.77%	2.09123
1,1,1-TCF3C005	25	19.24	24.02	25.4	25.11	26.13	24.74	24.54	24.2971	97.19%	2.28862
1,1,1-TCF3C005	23.138	18.1	22.47	22.49	22.07	22.14	22.94	22.78	21.8557	94.48%	1.68532
1,1,1-TCF3C005	25	19.01	23.4	24.05	24.2	24.13	23.27	23.75	23.1157	92.48%	1.84677
1,1,1-TCF3C005	25	18.98	23	24.29	23.24	24.82	22.96	23.02	23.0157	92.08%	1.86603
1,1,1-TCF3C005	25	18.4	22.38	23.29	22.84	23.81	23.09	23.87	22.46	89.84%	1.84032

Analyzed by: [Signature] Date: 2/1/22
 Extracted by: [Signature] Date: 2/1/22
 Approved by: [Signature] Date: 02/01/22

LCM5-FC-FRM25A1E
 Ver 3.0 (01/27/20)

Method:
Instrument ID:
Date Reported
Units
SOP ID:
SOP Revision:

Date: 2/1/22

Date: 2/1/22

Date: 02/01/22

Analyzed by: 
Extracted by: 
Approved by: 

LCMS-FO-FRM20133
Ver 4.0 (11/12/18)

6. *Principles of Mathematics*

[illegible]

Method: LCMS11
Instrument ID: 012622
Date Reported: PPT
Units: 24110
SOPID: 2
SOP Revision:

[illegible]

LCN5-F3-FRM20133
Ver 4.0(11/12/16)

Attachment IV, Standard Preparation**A. 537 Analyte Primary Dilution Standard (PDS), high concentration**

The high concentration analyte PDS solution is prepared by diluting purchased stock standard

Analyte PDS High Concentration				
Name	Stock Standard Conc, µg/ml	Spike Vol	Final Conc, µg/ml	Final Vol, mL**
Custom Analyte Mix	2	3.125mL	0.25*	25

* Refer to certificate of analysis; a few analytes are less than 2µg/mL.

** A solution of 96:4 methanol/water is used to make up the remaining volume.

Bring all stock standards to room temperature before use

Transfer the prepared standard into 2mL polypropylene vials and store in the refrigerator

Record standard preparation ~~and label vials as follows @537 Analyte PDS STD(H) 250PPB (DATE PREPARED) (ANALYSTS INITIALS) (vial number) in TALS~~

B. 537 Analyte Primary Dilution Standard (PDS), low concentration

The low concentration analyte PDS solution is prepared by diluting the high concentration analyte PDS with 96:4 methanol/water

Analyte PDS Low Concentration				
Name	PDS Standard Conc, µg/ml	Spike Vol, mL	Final Conc, µg/ml	Final Vol, mL*
PDS High Concentration	0.25	1	0.025	10

* A solution of 96:4 methanol/water is used to make up the remaining volume.

Bring all PDS standards to room temperature before use

Transfer the prepared standard into 2mL polypropylene vials and store in the refrigerator

Record standard preparation ~~in TALS and label vials as follows @537 Analyte PDS STD(L) 25PPB- (DATE PREPARED) (ANALYSTS INITIALS) (vial number)~~

Attachment IV, Standard Preparation (cont'd)**C. Calibration standard**

The calibration standard is prepared as follows:

Working Conc, ug/L	Equivalent ppt Conc in Extract*	Volume of 96:4 (Methanol/water) Mixture, µL	volume 537-IS-PDS, µL	Vol 537-SURR-PDS, µL	Vol 537 PDS STD low Conc, µL	Vol 537 PDS STD High Conc, µL
0	0	1960	20	20	0	-
0.5	2	1920	20	20	40	-
1.25	5	1950	20	20	-	10
2.5	10	1940	20	20	-	20
5	20	1920	20	20	-	40
10	40	1880	20	20	-	80
15	60	1840	20	20	-	120
20	80	1800	20	20	-	160
0.5	2 = CCCL	1920	20	20	40	-
7.5	30 = CCCM	1900	20	20	-	60
12.5	50 = CCCH	1860	20	20	-	100

*This concentration incorporates sample prep factor. A 250mL sample is extracted with an extract final volume of 1mL

Bring all standards to room temperature before use

Record standard preparation and label vials as follows @537-CAL-(DATE PREPARED)-(ANALYSTS INITIALS)in TALS

Attachment IV, Standard Preparation (cont'd)**D. Second Source Primary Dilution Standard**

The second source PDS solution is prepared by diluting a stock standard 96:4 methanol/water. The second source stock standard should be purchased from a vendor different from what was used in analyte PDS standard (attachment III, section A and B)

Analyte PDS Low Concentration				
Name	PDS Standard Conc, µg/ml	Spike Vol, mL	Final Conc, µg/ml	Final Vol, mL*
2 nd Source custom Mix	2	0.625	0.25	5

* A solution of 96:4 methanol/water is used to make up the remaining volume.

Bring stock standards to room temperature before use

Transfer the prepared standard into 2mL polypropylene vials and store in the refrigerator

Record standard preparation ~~and label vials as follows @537-QCS-PDS-(DATE PREPARED)-(ANALYSTS INITIALS)-(vial number)in TALS.~~

E. Second source QCS

The second source QCS is analyzed after every calibration and is prepared by combining several standards

Working Conc, ug/L	Equivalent ppt Conc in Extract*	Volume of 96:4 (Methanol/water) Mixture, µL	volume 537-IS-PDS, µL	Vol 537-SURR-PDS, µL	Vol 2nd Source PDS, µL
5	20	1920	20	20	40

*This concentration incorporates sample prep factor. A 250mL sample is extracted with an extract final volume of 1mL

Bring all standards to room temperature before use

Record standard preparation ~~in TALS and label vials as follows @537-2nd Source-QCS-(DATE PREPARED)-(ANALYSTS INITIALS)~~

Attachment IV, Standard Preparation (cont'd)

F. Internal Standards Primary Dilution Standard (PDS):

The IS PDS solution is prepared by combining 3 separate stock standards into 1 mix

Internal Standard Preparation				
Name	Stock Standard Conc, µg/ml	Spike Vol, mL	Final Conc, µg/ml	Final Vol, mL*
13C4PFOA (MPFOA)	50	0.5	1	25
13C4PFOS (MPFOS)	50	0.5	1	
d3-NMeFOSAA	50	2	4	

* A solution of 96:4 methanol/water is used to make up the remaining volume.

Bring all stock standards to room temperature before use

Transfer the prepared standard into 2mL polypropylene vials and store in the refrigerator

Record standard preparation ~~and label vials as follows @537 IS PDS STANDARD (DATE PREPARED) (ANALYTS INITIALS) (vial number) in TALS.~~

G. 537 IS Solution

The IS solution is used at sample preparation. It is prepared by diluting @537 IS Primary Dilution Standard

- Add 5mL of @537-IS-PDS to a 500mL volumetric flask with about 490mL 96:4 methanol/water solution
- Fill to the mark with 96:4 methanol/water solution
- Cap and invert flask several times to mix
- Transfer into corning centric star vials.
- Record standard preparation ~~and label vials as follows @537 IS SOLUTION (DATE PREPARED) (ANALYTS INITIALS) in TALS.~~
- Store standards in the refrigerator. Bring to room temperature prior to use

Attachment IV, Standard Preparation (cont'd)**H. Surrogate Primary Dilution Standard**

The surrogate PDS solution is prepared by combining 4 separate stock standards into 1 mix

Surrogate Standard Preparation				
Name	Stock Conc. (µg/mL)	Spike Vol. (mL)	Surr Conc. (µg/mL)	Final Vol. (mL)*
13C2-PFHxA (MPHXA)	50	0.5	1	25
13C2-PFDA (MPFDA)	50	0.5	1	
d5-NEtFOSAA	50	2	4	
GenX-13C3 (M3HFPO-DA)	50	0.5	1	

* A solution of 96:4 methanol/water is used to make up the remaining volume.

Bring all stock standards to room temperature before use

Record standard preparation in TALS and label vials as follows @537-SURR-PDS-STD-(DATE PREPARED)-
(ANALYSTS INITIALS)

Attachment V: Instrument Conditions-p1

5334537-1-012320.dam
Wednesday August 12 2020 15:08:47 PM
Friday October 23 2020 08:08:21 AM
M COLUMN 5002 mm 3uV 20nm NH4AC MeOH new gradient
LC Sync
Off
6min30sec
780
1
Acquisition Method
Analyst 1.7.1

Scans in Period: 780
Min. Dwell Time: 3 ms
Max. Dwell Time: 250 ms
Relative Start Time: 0.00 msec
Scheduled Ionization: Off
Experiments in Period: 1
Use target Cycle Time: No
Target Cycle Time: N/A

Period 1: Experiment 1:

Scan Type: MRM (MRM)
Scheduled MRM: Yes
Polarity: Negative
Scan Mode: N/A
Ion Source: Turbo Spray
Dynamic Window Extension: No
Dynamic Background Subtraction: No
sMRM Q1Q3 Resolution: No
MRM detection window: 60 sec
Target Scan Time: 0.5000 sec
Resolution Q1: Unit
Resolution Q3: Unit
Intensity Thres.: 0.00 cps
Settling Time: 0.0000 msec
MR Pause: 5.0070 msec
MCA: No
Step Size: 0.00 Da

Q1 Mass (Da) Q3 Mass (Da) RT (min) Param Start Stop ID

Attachment V: Instrument Conditions (cont'd)-p2

631.000	451.000	5.00	DE	-100.00	-100.00	11Cl-PF301GS
				CE	-42.00	-42.00
				CXF	-12.00	-12.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID
531.000	351.000	4.34	DF	-60.00	-60.00	9Cl-PF301GS
				CE	-40.00	-40.00
				CXF	-12.00	-12.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID
377.000	251.000	3.38	DF	-30.00	-30.00	ADONA
				CE	-16.00	-16.00
				CXF	-12.00	-12.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID
285.000	169.000	3.07	DF	-35.00	-35.00	GenX
				CE	-14.00	-14.00
				CXF	-12.00	-12.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID
299.000	80.000	2.69	DF	-55.00	-55.00	PFBS
				CE	-58.00	-58.00
				CXF	-12.00	-12.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID
513.000	469.000	4.50	DF	-30.00	-30.00	PFPA
				CE	-16.00	-16.00

Attachment V: Instrument Conditions (cont'd)-p3

Window (sec)	0.0	Prim / Sec	1	Trig Threshold	N/A	Dwell Weight	1.0	Group	N/A	CX	-15.00	-15.00
Q1 Mass (Da)	613.000	Q3 Mass (Da)	569.000	RT (min)	5.15	Param	DF	Start	Stop	ID		
								-25.00	-25.00	PFDA		
								CE	-18.00	-18.00		
								CX	-15.00	-15.00		
Window (sec)	0.0	Prim / Sec	1	Trig Threshold	N/A	Dwell Weight	1.0	Group	N/A	CX	-15.00	-15.00
Q1 Mass (Da)	363.000	Q3 Mass (Da)	319.000	RT (min)	3.33	Param	DF	Start	Stop	ID		
								-25.00	-25.00	PFDA		
								CE	-12.00	-12.00		
								CX	-15.00	-15.00		
Window (sec)	0.0	Prim / Sec	1	Trig Threshold	N/A	Dwell Weight	1.0	Group	N/A	CX	-15.00	-15.00
Q1 Mass (Da)	313.000	Q3 Mass (Da)	268.800	RT (min)	2.97	Param	DF	Start	Stop	ID		
								-25.00	-25.00	PFDA		
								CE	-12.00	-12.00		
								CX	-15.00	-15.00		
Window (sec)	0.0	Prim / Sec	1	Trig Threshold	N/A	Dwell Weight	1.0	Group	N/A	CX	-15.00	-15.00
Q1 Mass (Da)	399.000	Q3 Mass (Da)	80.000	RT (min)	3.35	Param	DF	Start	Stop	ID		
								-60.00	-60.00	PFMS		
								CE	-74.00	-74.00		
								CX	-12.00	-12.00		
Window (sec)	75.0	Prim / Sec	1	Trig Threshold	N/A	Dwell Weight	1.0	Group	N/A	CX	-15.00	-15.00
Q1 Mass (Da)	463.000	Q3 Mass (Da)	419.000	RT (min)	4.12	Param	DF	Start	Stop	ID		
								-25.00	-25.00	PFNA		
								CE	-14.00	-14.00		
								CX	-15.00	-15.00		

Attachment V: Instrument Conditions (cont'd)-p4

Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group
0.0	1	N/A	1.0	N/A
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param Start	Stoq ID
413.000	369.000	3.72	DF -25.00	PFQA
			CE -14.00	-14.00-14.00
			CXF -15.00	-15.00-15.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group
0.0	1	N/A	1.0	N/A
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param Start	Stoq ID
499.000	80.000	4.12	DF -65.00	PFQS
			CE -108.00	-108.00
			CXF -12.00	-12.00-12.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group
75.0	1	N/A	1.0	N/A
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param Start	Stoq ID
563.000	519.000	4.84	DF -25.00	PFtA
			CE -16.00	-16.00-16.00
			CXF -12.00	-12.00-12.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group
0.0	1	N/A	1.0	N/A
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param Start	Stoq ID
279.000	85.000	2.75	DF -20.00	PFMEA
			CE -22.00	-22.00-22.00
			CXF -11.00	-11.00-11.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group
0.0	1	N/A	1.0	N/A
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param Start	Stoq ID
229.000	85.000	2.46	DF -15.00	PFMEA
			CE -22.00	-22.00-22.00
			CXF -11.00	-11.00-11.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group
0.0	1	N/A	1.0	N/A

Attachment V: Instrument Conditions (cont'd)-p5

Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Parar	Start	Stop	ID
315.000	135.000	2.82	DF	-75.00	-75.00	PFEESA
				CE	-30.00	-30.00
				CXF	-11.00	-11.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Parar	Start	Stop	ID
295.000	201.000	2.93	DF	-10.00	-10.00	NETHA
				CE	-14.00	-14.00
				CXF	-11.00	-11.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Parar	Start	Stop	ID
213.000	169.000	2.35	DF	-25.00	-25.00	PFFA
				CE	-12.00	-12.00
				CXF	-11.00	-11.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Parar	Start	Stop	ID
263.000	219.000	2.65	DF	-20.00	-20.00	PFFaA
				CE	-12.00	-12.00
				CXF	-11.00	-11.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Parar	Start	Stop	ID
349.000	80.000	3.00	DF	-100.00	-100.00	PFFeS
				CE	-74.00	-74.00
				CXF	-11.00	-11.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		

Attachment V: Instrument Conditions (cont'd)-p6

Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID
449.000	80.000	3.74	DF	-65.00	-65.00	PFHqS
				CE	-88.00	-88.00
				CXF	-11.00	-11.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID
327.000	307.000	2.95	DF	-75.00	-75.00	4:2 FTS
				CE	-28.00	-28.00
				CXF	-11.00	-11.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID
427.000	407.000	3.70	DF	-50.00	-50.00	6:2 FTS
				CE	-32.00	-32.00
				CXF	-11.00	-11.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID
527.000	507.000	4.50	DF	-50.00	-50.00	8:2 FTS
				CE	-40.00	-40.00
				CXF	-11.00	-11.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID
287.000	169.000	3.08	DF	-35.00	-35.00	Surr-13C3-GenX
				CE	-14.00	-14.00
				CXF	-11.00	-11.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID
519.000	474.000	4.50	DF	-25.00	-25.00	Surr-13C6-PFDA

Attachment V: Instrument Conditions (cont'd)-p7

CE -16.00 -16.00 CXF -11.00 -11.00									
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group					
0.0	1	N/A	1.0	N/A					
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID			
318.000	273.000	2.97	DE	-25.00	-25.00	Surr-13C5-FFHxA			
CE -12.00 -12.00 CXF -11.00 -11.00									
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group					
0.0	1	N/A	1.0	N/A					
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID			
217.000	172.000	2.35	DE	-25.00	-25.00	Surr-13C4-FFFeA			
CE -12.00 -12.00 CXF -11.00 -11.00									
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group					
0.0	1	N/A	1.0	N/A					
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID			
268.000	223.000	2.65	DE	-20.00	-20.00	Surr-13C5-FFFeA			
CE -12.00 -12.00 CXF -11.00 -11.00									
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group					
0.0	1	N/A	1.0	N/A					
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID			
367.000	322.000	3.33	DE	-25.00	-25.00	Surr-13C4-FFHpA			
CE -12.00 -12.00 CXF -15.00 -15.00									
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group					
0.0	1	N/A	1.0	N/A					
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID			
421.000	376.000	3.72	DE	-25.00	-25.00	Surr-13C8-FFOA			
CE -14.00 -14.00 CXF -15.00 -15.00									

Attachment V: Instrument Conditions (cont'd)-p8

Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group
0.0	1	N/A	1.0	N/A
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start Stox ID
472.000	427.000	4.12	DF	-25.00 -25.00 Surr-1339-FFNA
			CE	-14.00-14.00
			CXF	-15.00-15.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group
0.0	1	N/A	1.0	N/A
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start Stox ID
570.000	525.000	4.84	DF	-25.00 -25.00 Surr-1337-FFUnA
			CE	-16.00-16.00
			CXF	-12.00-12.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group
0.0	1	N/A	1.0	N/A
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start Stox ID
615.000	570.000	5.15	DF	-25.00 -25.00 Surr-1332-FFDoA
			CE	-18.00-18.00
			CXF	-15.00-15.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group
0.0	1	N/A	1.0	N/A
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start Stox ID
302.000	80.000	2.69	DF	-55.00 -55.00 Surr-1333-FFES
			CE	-58.00-58.00
			CXF	-12.00-12.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group
0.0	1	N/A	1.0	N/A
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start Stox ID
402.000	80.000	3.35	DF	-60.00 -60.00 Surr-1333-FFHS
			CE	-74.00-74.00
			CXF	-12.00-12.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group

0.0	1	N/A	1.0	N/A
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start Stop ID
507.000	80.000	4.10	DF	-65.00 -65.00 Surr-13C8-PFOS
				CE -108.00
				CXF -12.00-12.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group
0.0	1	N/A	1.0	N/A
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start Stop ID
329.000	309.000	2.93	DF	-75.00 -75.00 Surr-13C2-4:2 FTS
				CE -28.00-28.00
				CXF -11.00-11.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group
0.0	1	N/A	1.0	N/A
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start Stop ID
429.000	409.000	3.70	DF	-50.00 -50.00 Surr-13C2-6:2 FTS
				CE -32.00-32.00
				CXF -11.00-11.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group
0.0	1	N/A	1.0	N/A
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start Stop ID
529.000	509.000	4.50	DF	-50.00 -50.00 Surr-13C2-8:2 FTS
				CE -40.00-40.00
				CXF -11.00-11.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group
0.0	1	N/A	1.0	N/A
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start Stop ID
216.000	172.000	2.35	DF	-25.00 -25.00 IS-13C3-PFEA
				CE -12.00-12.00
				CXF -11.00-11.00
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group
0.0	1	N/A	1.0	N/A
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start Stop ID
0.0	0.0	0.0	0.0	0.0

Attachment V: Instrument Conditions (cont'd)-p10

Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID
415.000	370.000	3.72	DE	-25.00	-25.00	IS-13C2-FFOA
			CE	-14.00	-14.00	
			CXF	-11.00	-11.00	
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID
503.000	80.000	4.10	DE	-65.00	-65.00	IS-13C4-FFOS
			CE	-108.00	-108.00	
			CXF	-11.00	-11.00	
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID
584.000	419.000	4.85	DE	-70.00	-70.00	N-EtFOSAA
			CE	-28.00	-28.00	
			CXF	-12.00	-12.00	
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
75.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID
570.000	419.000	4.66	DE	-70.00	-70.00	N-MeFOSAA
			CE	-30.00	-30.00	
			CXF	-12.00	-12.00	
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
75.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID
713.000	669.000	5.66	DE	-25.00	-25.00	FFTA
			CE	-22.00	-22.00	
			CXF	-15.00	-15.00	
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID

Attachment V: Instrument Conditions (cont'd)-p11

663.000	619.000	5.42	DE	-25.00	-25.00	PFTnDA
			CE	-20.00	-20.00	
			CXF	-15.00	-15.00	
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID
589.100	418.800	4.85	DE	-70.00	-70.00	Surr-05-NEtFOSAA
			CE	-30.00	-30.00	
			CXF	-12.00	-12.00	
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID
573.000	419.000	4.66	DE	-70.00	-70.00	IS-d3-NEtFOSAA
			CE	-30.00	-30.00	
			CXF	-12.00	-12.00	
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID
515.000	470.000	4.50	DE	-25.00	-25.00	Surr-13C2-PFtA
			CE	-16.00	-16.00	
			CXF	-12.00	-12.00	
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID
315.000	270.000	2.97	DE	-25.00	-25.00	Surr-13C2-PFtA
			CE	-12.00	-12.00	
			CXF	-15.00	-15.00	
Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group		
0.0	1	N/A	1.0	N/A		
Q1 Mass (Da)	Q3 Mass (Da)	RT (min)	Param	Start	Stop	ID
417.000	372.000	3.72	DE	-25.00	-25.00	IS-13C4-PFOA
			CE	-14.00	-14.00	

Attachment V: Instrument Conditions (cont'd)-p12

UNCONTROLLED

Window (sec)	Prim / Sec	Trig Threshold	Dwell Weight	Group
0.0	1	N/A	1.0	N/A

Parameter Table (Period 1 Experiment 1):

CUR:	35.00
TEM:	450.00
GS1:	40.00
GS2:	55.00
CAD:	9.00
IS:	-4500.00
EF:	-10.00

CXF -15.00 -15.00

Integrated Valve Method Properties

Valve	Diverter	Total Time (min)	Position
1		1.5	B
2		6.2	A

Sciex IC Method Properties

Sciex IC system Equilibration time = 0.00 min
Sciex IC system Injection Volume = 2.00 ul

Binary Gradient

Model:	AC Pump
<General>	
Stop time:	9.00 min
Flow:	0.6000 mL/min
Pressure limits Maximum:	7000 psi
Pressure limits Minimum:	0 psi
B.Conc:	5.0 %
B.Curve:	0

<Gradient>

Time	Flow	B.Conc	B.Curve
min	mL/min	%	
0.60	0.6000	5.0	0
0.70	0.6000	55.0	0
4.30	0.6000	90.0	0
4.50	0.6000	99.0	0
7.49	0.6000	99.0	0
7.50	0.6000	5.0	0
9.00	0.6000	5.0	0

<Solenoid valve>

Pump A: None

Attachment V: Instrument Conditions (cont'd)-p13

UNCONTROLLED

Pump B:	None
<Compressibility settings>	None
<Compressibility settings>	No
<Autopurge settings>	
Purge order Mobile phase name	Purge time min
1st	None
2nd	None
<Init conc-replacement>	
Use Init conc-replacement:	No
Isocratic 1	
Model:	AC Pump
<General>	
Stop time:	0.01 min
Flow:	0.0000 mL/min
Pressure limits Maximum:	1451 psi
Pressure limits Minimum:	0 psi
<Solenoid valve>	
Pump A:	None
<Compressibility settings>	
<Compressibility settings>	No
<Autopurge settings>	
Purge order Mobile phase name	Purge time min
1st	None
<Init conc-replacement>	
Use Init conc-replacement:	No
Autosampler	
Model:	AC Autosampler
<General>	
Use Autosampler:	Yes
<Sample rack settings>	
Specify rack:	No
Rack/Stack	Type Needle stroke mm
Sample rack	Rack 1.5mL 105 vials 50
	Rack 1.5mL 70 vials 52
	Rack 1mL Cool 51
	Rack 4mL Cool 51
	Rack MTP 96 Cool 45
	Rack MTP 384 Cool 45
	Rack Deep Well 96 Cool 40

Attachment V: Instrument Conditions (cont'd)-p14

Changer stack D	Rack Deep Well 384 Cool	40
	MTP 96	45
	DWP 96	45
	1.5mL	44
	MTP 384	45
	DWP 384	45
Changer stack C	MTP 96	45
	DWP 96	45
	1.5mL	44
	MTP 384	45
	DWP 384	45
Changer stack B	MTP 96	45
	DWP 96	45
	1.5mL	44
	MTP 384	45
	DWP 384	45
Changer stack A	MTP 96	45
	DWP 96	45
	1.5mL	44
	MTP 384	45
	DWP 384	45
<Injection settings>		
Sampling speed:		5 µL/s
Use cooler temperature:		Yes
Cooler temperature:		15 °C
Control vial needle stroke:		50 mm
<Acquisition cycle time optimization>		
Pretreatment start timing:		Off
<Rinse settings>		
Rinse mode:	Before and after aspiration	
Rinse dip time:	0 s	
Rinsing speed:	35 µL/s	
Rinsing volume:	500 µL	
<Purge settings>		
Purge time:	25.0 min	
<Autopurge settings>		
Execute sampler purge:	No	
Column Oven		
=====		
Model:	AC Column Oven	
<General>		
Use Column Oven:	Yes	
Oven temperature:	40 °C	
Temperature limit (Maximum):	90 °C	
<Advanced>		
Wait for temperature equilibration before run:	Yes	
<Valve>		

Attachment V: Instrument Conditions (cont'd)-p15

Valve L: Position:1
Valve R: Position:1
System Controller
Model: FCV-12H
Controller: FCV-12H
<General>
Execute autopurge before analysis: No
<External output settings>
Power on: No
Event 1: No
Event 2: No
Event 3: No
Event 4: No
<Autopurge settings>
<Warm up>
Wait time: 0 min
Binary Gradient flow: 0.0000 mL/min
Isocratic flow: 0.0000 mL/min
<Execute after autopurge>
Turn oven(s) on after autopurge: No
Turn pump(s) on after autopurge: No

Attachment VI: Method QC Summary

QC TYPE	CONCENTRATION LEVEL	FREQUENCY	ACCEPTANCE CRITERIA
Analysis Batch		Begins and ends with the appropriate CCCs. Maximum of 20 extracted field samples.	
Extraction Batch		No more than 20 field samples by the same person during a work day using the same lots of supplies.	
Initial Calibration	A minimum of 5 (linear) calibration points, the lowest must be at or lower than the MRL. Minimum 6 points required for Quadratic regression	Run during initial method set up. Run appropriate CCCs for subsequent batches, if CCCs do not meet recovery criteria, run initial calibration	Back calculate the analyte concentration and recoveries must be within 50-150% for points at or below the MRL and within 70-130% for other points.
Continuing Calibration Check (CCC)	CCL (2.0 ng/L), CCM (30 ng/L), or CCH (50 ng/L).	A CCL must be analyzed immediately after the initial calibration curve, prior to any QC or samples. Rotate CCM and CCH after every tenth field sample and at the end of each analysis batch.	Recovery must be within 50-150% of the true value for CCL and within 70-130% for CCM and CCH.
Unextracted Mid-level QCS (QCS)	20 ng/L	Analyze as part of the IDC, at least quarterly, or with each new calibration prep.	Recovery must be within 70-130% of the true value.
Internal Standard	40 ng/L in terms of IS PFOA- ¹³ C ₄ .	Introduce into every field sample, calibration, and QC sample.	IS area counts must be 50-150% of the average IS area count from the initial calibration and 70-140% of the most recent CCC
Surrogate	40 ng/L in terms of ¹³ C ₂ -PFHxA	Introduce into every field sample, calibration, and QC sample.	Recovery must be within 70-130% of the target value.
Lab Reagent Blank (LRB/MBLK)	Reagent Water prepared, and analyzed as a sample.	Include one LRB/MBLK with each extraction batch.	Results must be less than 1/3 the MRL.

QC TYPE	CONCENTRATION LEVEL	FREQUENCY	ACCEPTANCE CRITERIA
		If an automated system is used, the LRBs should be rotated among the ports	
Lab Fortified Blank (LFB/LCS)	LFB-L (2.0 ng/L), LFB-M (25 ng/L), or LFB-H (50 ng/L).	LFB-L per day. As well as LFB-M or LFB-H rotated per batch	70-130% recovery for mid- and high-levels. 50-150% recovery for low-level.
Lab Fortified Sample Matrix (LFSM/MS)	Rotate between low (2.0 ng/L), mid (25 ng/L), and high (50 ng/L) between extraction batches.	Include one LFSM/MS per extraction batch (20 samples or less).	70-130% recovery for mid- and high-levels. 50-150% recovery for low-level.
Lab Fortified Sample Matrix Duplicate (LFSMD/MSD)/Field Duplicate (FD/DUP)	For LFSMD, rotate between low (2.0 ng/L), mid (30 ng/L), and high (30 ng/L) between extraction batches.	Include at least one FD/DUP or LFSMD/MSD with each extraction batch.	RPD must be less than or equal to 50% for analytes within 2x the MRL. RPDs less than or equal to 30% for analytes higher than 2x the MRL. For LFSMD, 70-130% recovery for mid- and high-levels. 50-150% recovery for low-level.
Field Reagent Blank (FRB)	Reagent Water prepared, and analyzed as a sample.	Process only if an associated field sample contains an analyte at or above the MRL.	Results must be less than or equal to 1/3 the MRL.
Initial Demonstration of Low System Background	Prepare, extract, and analyze as a sample.	Analyze an extracted LRB prior to any other IDC steps. If an automated system is used, the LRBs must be extracted from each port. After IDC, LRB should be rotated among the ports	Results must be less than 1/3 the MRL.
Initial Demonstration of Precision	Prepare, extract and analyze 7 replicate LFB/LCSs fortified at 25 ng/L.	When beginning the use of this method, with each new analyst, and after a major change in instrumentation.	RSD must be less than 20% for the analytes.

QC TYPE	CONCENTRATION LEVEL	FREQUENCY	ACCEPTANCE CRITERIA
Initial Demonstration of Accuracy	Use the same results obtained from the Initial Demonstration of Precision.	When beginning the use of this method, with each new analyst, and after a major change in instrumentation.	Mean recovery must be within 70-130%.
Continuing Demonstration of Capability		MDL and Precision and Accuracy performed annually for each analyst or extractionist.	RSD must be less than 20% for the analytes. Mean recovery must be within 70-130%.
MRL Confirmation	Prepare, extract and analyze 7 replicate LFB/LCSs fortified at the proposed MRL.	Initially on each instrument and whenever there is a major change in instrumentation. Each new analyst and extractionist, performing this method.	The upper PIR limits must be less than or equal to 150%. The lower PIR limits must be greater than or equal to 50%.
LFB/LFBDup (LCS/LCSDup)	Rotate between mid (25 ng/L), and high (50 ng/L) between extraction batches.	For Arizona samples when MS/MSD failed acceptance for accuracy and precision	RPDs less than or equal to 30%. 70-130% recovery.
Establish RT for both linear and branched isomers	Run at mid to high level concentration	Run as part of IDC Repeat anytime chromatographic changes occur that alter analyte retention time.	All isomers of each analyte must elute with the same MRM window

Attachment VII: Analytical Sequence

Cal Std1

Cal Std2

Cal Std3

Cal Std4

Cal Std5 - Peak Assymetry Factor calculation

Cal Std6

CCC at MRL Level

QCS at Mid-level

LRB/MBLK

LFB at MRL level

LFB

FS (Field Sample) 1

LFSM/MS on FS 1

LFSMD/MSD on FS 1

FS 2 to FS 10

Mid-level CCC

FS 11 to FS 20

High-level CCC

Attachment IX, Caliper Lifesciences Automated SPE Workstation Conditions/Parameters

AutoTrace SPE Instrument Copyright © 2009 by Dionex Corporation Version 1.00.00 Cartridge Model

File Methods Command Login/Logout Data Windows Help

Methods Command Login Data Help Exit

Set Up Parameters

Last Change 10/3/2018 5:27:08 PM

Flow Rates

Cond Flow: 15.0 mL/min
Load Flow: 6.0 mL/min
Rinse Flow: 20.0 mL/min
Elute Flow: 4.0 mL/min
Cond Air Push: 15.0 mL/min
Rinse Air Push: 20.0 mL/min
Elute Air Push: 4.0 mL/min

SPE Parameters

Push Delay: 5 sec
Air Factor: 1.0
Autowash Vol.: 1.00 mL

Instrument Parameters

Max. Elution Vol.: 20.0 mL
Exhaust Fan On: ☒
Beeper On: ☒

No User logged on NUM

Set Up Methods

Name 537
Samples 6 Solvent Set 537

Condition Cartridge

Min. Max. Volume 0.1 8.0 10.0 [mL]

Waste Position

☒ Solvent Waste
☐ Aqueous Waste

solvents

☐ CH₂Cl₂
☒ CH₃OH
☐ HPLC H₂O
☐ 90% MTBE:10% CH₃OH
☐ 0.05 N HCl

Estimated time 2h : 5min

No.	Method
1	Process 6 Samples using the following method steps:
2	Condition Cartridge with 8.0 mL of MECH into solvent waste
3	Condition Cartridge with 8.0 mL of MECH into solvent waste
4	Condition Cartridge with 9.0 mL of WATER into aqueous waste
5	Condition Cartridge with 9.0 mL of WATER into aqueous waste
6	Load 270.0 mL of sample onto Cartridge
7	Pause and Alert operator, resume when CONTINUE is pressed
8	Load 18.0 mL of sample onto Cartridge

No User logged on NUM

Attachment X Procedure for pH and Free Chlorine Checks

Note: The pH must be verified prior to checking for free chlorine. See Extractions-WI23106, pH and Chlorine Check for Extractions

1. pH Verification
 - a) Check the initial pH of the sample and record it on the backlog, which will later be entered in ~~the webform sheet~~ TALS. The pH must be between 6.5 and 7.5.
 - b) If the sample pH is out of range, verify the pH of the back-up container.
 - c) If the the pH of the back-up container is also out of range, notify the ASM and supervisor of the non-compliant sample.
 - d) If allowed by the client, add approximately 1.25 g of trizma to the sample bottle. Verify the final pH and record it on the backlog.
 - e) If the final pH is within range, extract the sample and generate a QIR. If the final pH is not within range, do not add more trizma. Extract the sample and generate a QIR, while noting that the pH requirement was not met.
2. Free Chlorine Check
 - a) Check and record the initial free chlorine using the DPD free chlorine reagent and approximately 10 mL of sample. Free chlorine must be less than 0.1 ppm.
 - b) If the color of the sample turns pink after addition of the reagent, verify the presence of chlorine using the SenSafe strips.
 - c) If free chlorine is verified, check the back-up container for free chlorine.
 - d) If the back-up container also fails, notify the ASM and supervisor of the non-compliant sample.
 - e) If allowed by the client, add approximately 1.25 g of trizma to the sample bottle. Check and record the final free chlorine.
 - f) If free chlorine is less than 0.1 ppm, extract the sample and generate a QIR. If free chlorine is still present, notify the ASM and request that the client re-sample

Note: Excessive amounts of trizma may result in low surrogate recovery

Attachment XI EPA email with regards to Trizma preservation**Dean, Robert**

To: Ramos, Marnellie
Subject: RE: EPA 537.1 - re ph and residual chlorine from Jody Shoemaker

From: Ali Haghani
Sent: Monday, April 15, 2019 8:10 AM
To: Nilda Cox
Subject: FW: EPA 537

Response to EPA 537 preservation

From: Shoemaker, Jody [<mailto:shoemaker.jody@epa.gov>]
Sent: Monday, April 15, 2019 4:53 AM
To: Ali Haghani
Subject: RE: EPA 537

EXTERNAL EMAIL*

Ali,

Trizma only sequesters free chlorine and reduces it to the weaker oxidant, chloramine. Thus, when measuring free chlorine, it has to be measured very quickly or the chloramine will cause color to form with time.

I know this came up during UCMR 3 so I will ask OGWDW what their guidance was to labs since I don't remember now.

As for adding Trizma at the bench – have you actually determined that the samples in questions are not buffered near pH 7? Just because they have chloramine present doesn't mean they are not buffered.

You should not have a need to add more Trizma to drinking water samples to meet the pH requirements of the method.

Regarding M559-Dan's plan is to finish the edits of the draft method today and send it out by the end of the day. Sorry, but we were both off work on Friday so did not get your email until this morning. Thank you for your willingness to participate in this verification.

Sincerely,
Jody

Dr. Jody Shoemaker
EPA/National Exposure Research Laboratory
26 W. Martin Luther King Drive
MS 587
Cincinnati, OH 45268
513-569-7298 (office)
shoemaker.jody@epa.gov

From: Ali Haghani <AliHaghani@eurofinsUS.com>
Sent: Thursday, April 11, 2019 10:51 PM
To: Shoemaker, Jody <shoemaker.jody@epa.gov>
Subject: EPA 537

Hello Jody;

I am looking forward again to do collaboration for your new method EPA 559 and in order to start preparing for it can I please have a copy of it right now - if possible so that I can go over it over the weekend.

Also I know EPA 537 adds Trizma for making all samples the same pH for extraction reproducibility. However, probably due to a policy or something outside the method itself chlorine check needs to be performed.

My question is that we occasionally receive samples that have still hits for chlorine is it possible to add additional Trizma at the bench to neutralize prior to extraction to omit resampling? My current understanding is that chlorine does not have adverse effect on the targeted compounds.

In addition we have found that DPD chlorine check can have false positives and we use strips to make sure samples are free of dissolved chlorine.

Kindest regards
Ali

Notify us [here](#) to report this email as spam.

* WARNING - EXTERNAL: This email originated from outside of Eurofins. Do not click any links or open any attachments unless you trust the sender and know that the content is safe!

Attachment XII EPA email with regards to Field Blanks
Dean, Robert

To: Cox, Nilda; Ramos, Marnellie; Rodriguez, Eduardo
Subject: RE: 537.1 Rev 2 Clarifications from Jody

From: Shoemaker, Jody [<mailto:shoemaker.jody@epa.gov>]
Sent: Thursday, August 27, 2020 12:32 PM
To: Nilda Cox <NildaCox@eurofinsUS.com>
Subject: RE: 537.1 Rev 2 Clarifications

EXTERNAL EMAIL*

Nilda,

The changes to the FRB were made in order for M537.1 to be consistent with Method 533 which was published after the first version of Method 537.1 came out. There really isn't much scientific difference but we decided we thought it would make it less confusing if M537.1 and M533's FRB were prepared in the same manner.

Hopefully this makes things clearer.

Jody

Dr. Jody Shoemaker
EPA/Center for Environmental Solutions and Emergency Response
26 W. Martin Luther King Drive
MS 689
Cincinnati, OH 45268
513-569-7298 (office)
shoemaker.jody@epa.gov

From: Nilda Cox <NildaCox@eurofinsUS.com>
Sent: Thursday, August 27, 2020 3:01 PM
To: Shoemaker, Jody <shoemaker.jody@epa.gov>
Subject: 537.1 Rev 2 Clarifications

Again – I need your assistance on 537.1 rev 2 . thank you for your previous assistance.

1. Need to clarify section 8.3 .1 changes . Please see below for my comments in blue highlights
2. I think the first paragraph was changed but the second paragraph was left alone coming from the original 537.1 revision 1)
3. Also – 537.1 rev 2 - stated that updates were editorial changes . Is section 8.3.1 changes intended to be editorial changes only ? thank you

8.3. FIELD REAGENT BLANKS (FRB)

8.3.1.

An FRB must be handled along with each sample set. The sample set is composed of samples collected from the same sample site and at the same time. At the laboratory, fill the field blank sample bottle with reagent water, then seal, and ship to the sampling site along with the sample bottles.

(1.Nilda - so the reagent water is not preserved)

For each FRB shipped, a second FRB bottle containing only the preservative must also be shipped. (2.Nilda - The FRB bottle now has preservative).

At the sampling site, the sampler must open the shipped FRB and pour the preserved reagent water (3.Nilda -this one is preserved reagent water -- inconsistent on the above item 1) into the empty shipped sample bottle. (4. Nilda - this is empty - inconsistent with item 2) seal and label this bottle as the FRB.

The FRB is shipped back to the laboratory along with the samples and analyzed to ensure that PFAS were not introduced into the sample during sample collection/handling.

thank you for your assistance .
regards

Nilda Cox
Quality Assurance Manager/Regulatory Consulting Manager

Eurofins Eaton Analytical, LLC
750 Royal Oaks Drive; Suite 100
Monrovia, CA 91016
Phone: +1 626 386 1170
Mobile: +1 626 318 8517

Email: NildaCox@Eurofinsus.com
Website: www.EurofinsUS.com/Env

Please note: In order to continue to provide critical testing services, **Eurofins Environment Testing laboratories in the US are maintaining our courier services and continue to sample, analyze and report all test data as usual.** The situation around COVID-19 continues to be fluid and we are continuing to follow local and government mandates as applicable. For up-to-date business information, visit our website and follow us on Facebook and LinkedIn.

Links to use:

Website: <https://www.eurofinsus.com/environment-testing>
Facebook: <https://www.facebook.com/EurofinsEnvTesting>
LinkedIn: <https://www.linkedin.com/company/eurofins-env-testing-america>

The information transmitted is intended only for the person or entity to which it is addressed and may contain confidential and/or privileged material. Any review, retransmission, dissemination or other use of, or taking of any action in reliance upon this information by persons or entities other than the intended recipient is prohibited. If you receive this in error, please contact the sender and delete the material from any computer. Email transmission cannot be guaranteed to be secure or error free as information could be intercepted, corrupted, lost, destroyed, arrive late or incomplete. The sender therefore is in no way liable for any errors or omissions in the content of this message which may arise as a result of email transmission. If verification is required, please request a hard copy. We take reasonable precautions to ensure our emails are free from viruses. You need, however, to verify that this email and any attachments are free of viruses, as we can take no responsibility for any computer viruses, which might be transferred by way of this email. We may monitor all email communication through our networks. If you contact us by email, we may store your name and address to facilitate communication.

Notify us [here](#) to report this email as spam.

Draft Method 1633

Analysis of Per- and Polyfluoroalkyl Substances (PFAS) in Aqueous, Solid, Biosolids, and Tissue Samples by LC-MS/MS

U.S. Environmental Protection Agency
Office of Water (4303T)
Office of Science and Technology
Engineering and Analysis Division
1200 Pennsylvania Avenue, NW
Washington, DC 20460

EPA 821-D-21-001

Draft Method 1633

Analysis of Per- and Polyfluoroalkyl Substances (PFAS) in Aqueous, Solid, Biosolids, and Tissue Samples by LC-MS/MS

August 2021

Notice

This document represents a draft of a PFAS method currently under development by the EPA Office of Water, Engineering and Analysis Division (EAD), in conjunction with the Department of Defense (DOD). **This method is not required for Clean Water Act compliance monitoring until it has been proposed and promulgated through rulemaking.**

A single-laboratory validation of the procedure has been completed and the report on the results of that study is being prepared. Historically, EAD posts draft methods on the Clean Water Act website after the single-laboratory validation report is completed. However, due to a large number of public and stakeholder requests, this method is being posted on the web before the single-laboratory validation study report is finalized. A revision of this draft method with a later publication date may be issued at that time. No procedural changes are expected as a result of the single-laboratory validation, but some of the performance data (which are presented only as examples) may change once the statistical analysis of the single-laboratory validation data is completed.

This draft method has been subjected to multiple levels of review across several EPA Program Offices. DOD expects to begin a multi-laboratory validation study of the procedure in late 2021, in collaboration with the Office of Water and the Office of Land and Emergency Management.

The Office of Water will use the results of the multi-laboratory validation study to finalize the method and add formal performance criteria. The method validation process may eliminate some of the parameters listed in this draft method.

In the meantime, the Office of Water is releasing this draft on its web site. Laboratories, regulatory authorities, and other interested parties are encouraged to review the method, and where appropriate, utilize it for their own purposes, with the explicit understanding that this is a draft method, subject to revision.

Acknowledgements

This draft method was prepared under the direction of Adrian Hanley of the Engineering and Analysis Division, Office of Science and Technology, within EPA's Office of Water, in collaboration with the Department of Defense.

EPA acknowledges the support of a number of organizations in the development and validation of this draft method, including the developers of the original procedure, the Department of Defense, the members of EPA's workgroup, and EPA's support contractor staff at General Dynamics Information Technology, including:

Adrian Hanley	EPA Office of Water, Office of Science and Technology, Engineering and Analysis Division
S. Bekah Burket	EPA Office of Water, Office of Science and Technology, Engineering and Analysis Division
Troy Strock	EPA Office of Land and Emergency Management
Diane Reese	EPA Region 4
Steve Reimer	EPA Region 10
Janice Willey	Department of Defense, Naval Sea Systems Command
Richard H. Anderson	Department of Defense, Air Force Civil Engineer Center
Allyson Buytendyk	Institute for Defense Analysis
Coreen Hamilton	SGS-AXYS Analytical
Ivona Zysk	SGS-AXYS Analytical
Ting Chen	SGS-AXYS Analytical
Henry Huang	SGS-AXYS Analytical
Mirna Alpizar	General Dynamics Information Technology
Harry McCarty	General Dynamics Information Technology

Disclaimer

See the notice on the title page regarding the status of this method.

Mention of trade names or commercial products does not constitute endorsement or recommendation for use.

Contact

Please address questions, comments, or suggestions to:

CWA Methods Team, Engineering and Analysis Division (4303T)
Office of Science and Technology
U.S. Environmental Protection Agency
1200 Pennsylvania Avenue
Washington, DC 20460

<https://www.epa.gov/cwa-methods>

<https://www.epa.gov/cwa-methods/forms/contact-us-about-cwa-analytical-methods>

Table of Contents

Acknowledgements.....	i
Disclaimer	i
Contact	i
1.0 Scope and Application	1
2.0 Summary of Method	1
3.0 Definitions.....	2
4.0 Contamination and interferences	2
5.0 Safety	3
6.0 Equipment and Supplies.....	5
7.0 Reagents and standards	8
8.0 Sample collection, preservation, storage, and holding times	13
9.0 Quality Control	14
10.0 Calibration and Standardization.....	18
11.0 Sample preparation and extraction.....	25
12.0 Extraction, Cleanup, and Concentration	30
13.0 Instrumental Analysis	32
14.0 Performance Tests during Routine Operations	33
15.0 Data Analysis and Calculations	35
16.0 Method Performance.....	38
17.0 Pollution Prevention.....	38
18.0 Waste Management.....	39
19.0 References.....	39
20.0 Tables, Diagrams, Flowcharts, and Validation Data.....	41
21.0 Glossary	55
 Appendix A - Sample Pre-screening Instructions.....	 59
Appendix B - Aqueous Sample Subsampling Instructions	60

**DRAFT Method 1633 - Analysis of Per- and Polyfluoroalkyl Substances (PFAS)
in Aqueous, Solid, Biosolids, and Tissue Samples by LC-MS/MS**

1.0 Scope and Application

- 1.1** Method 1633 is for use in the Clean Water Act (CWA) for the determination of the per- and polyfluoroalkyl substances (PFAS) in Table 1 in aqueous, solid (soil, biosolids, sediment) and tissue samples by liquid chromatography/mass spectrometry (LC-MS/MS).
- 1.2** The method calibrates and quantifies PFAS analytes using isotopically labeled standards. Where linear and branched isomers are present in the sample and either qualitative or quantitative standards containing branched and linear isomers are commercially available, the PFAS analyte is reported as a single analyte consisting of the sum of the linear and branched isomer concentrations.
- 1.3** The instrumental portion of this method is for use only by analysts experienced with LC-MS/MS or under the close supervision of such qualified persons. Each laboratory that uses this method must demonstrate the ability to generate acceptable results using the procedure in Section 9.2.
- 1.4** By their very nature, many components of PFAS present analytical challenges unique to this class of analytes. For example, PFAS analytes readily adhere to the walls of the sample containers and may also stratify in the container. EPA has included procedures in the method that must be employed to address such challenges (see Section 11.0 and Appendices A and B).
- 1.5** This method is “performance-based,” which means that modifications may be made without additional EPA review to improve performance (e.g., overcome interferences, or improve the sensitivity, accuracy, or precision of the results) *provided that* all performance criteria in this method are met. Requirements for establishing equivalency are in Section 9.1.2 and include 9.1.2.2c. For CWA uses, additional flexibility is described at 40 CFR 136.6. Changes in performance, sensitivity, selectivity, precision, recovery, etc., that result from modifications within the scope of 40 CFR Part 136.6, and Section 9.0 of this method must be documented, as well as how these modifications compare to the specifications in this method. Changes outside the scope of 40 CFR Part 136.6 and Section 9.0 of this method may require prior review or approval.

2.0 Summary of Method

Environmental samples are prepared and extracted using method-specific procedures. Sample extracts are subjected to cleanup procedures designed to remove interferences. Analyses of the sample extracts are conducted by LC-MS/MS in the multiple reaction monitoring (MRM) mode. Sample concentrations are determined by isotope dilution or extracted internal standard quantification (see Section 10.3) using isotopically labeled compounds added to the samples before extraction.

2.1 Extraction

- 2.1.1** Aqueous samples are spiked with isotopically labeled standards, extracted using solid-phase extraction (SPE) cartridges and undergo cleanup using carbon before analysis.
- 2.1.2** Solid samples are spiked with isotopically labeled standards, extracted into basic methanol, and cleaned up by carbon and SPE cartridges before analysis.

- 2.1.3** Tissue samples are spiked with isotopically labeled standards, extracted in potassium hydroxide and acetonitrile followed by basic methanol, and cleaned up by carbon and SPE cartridges before analysis.
- 2.2** This method measures the analytes as either their anions or neutral forms. The default approach for Clean Water Act uses of the method is to report the analytes in their acid or neutral forms, using the equations in Section 15.2, although the differences between the anion and acid form concentrations are minimal (See Table 8). Other project-specific reporting schemes may be used where required.
- 2.3** Individual PFAS analytes are identified through peak analysis of the quantification and confirmation ions, where applicable.
- 2.4** Quantitative determination of target analyte concentrations is made with respect to an isotopically labeled PFAS standard; the concentrations are then used to convert raw peak areas in sample chromatograms to final concentrations.
- 2.5** Results for target analytes are recovery corrected by the method of quantification (i.e., either isotope dilution or extracted internal standard quantification, see Section 10.3). Isotopically labeled compound recoveries are determined by comparison to the responses of one of seven non-extracted internal standards (a.k.a., the “recovery” standards) and are used as general indicators of overall analytical quality.
- 2.6** The quality of the analysis is assured through reproducible calibration and testing of the extraction, cleanup, and LC-MS/MS systems.

3.0 Definitions

Definitions are provided in the glossary at the end of this method.

4.0 Contamination and interferences

- 4.1** Solvents, reagents, glassware, and other sample processing hardware may yield artifacts and elevated baselines causing misinterpretation of chromatograms. Specific selection of reagents and solvents may be required.
- 4.2** Clean all equipment prior to, and after each use to avoid PFAS cross-contamination. Typical cleaning solvents used include water, methanol, and methanolic ammonium hydroxide. The residual PFAS content of disposable plasticware and filters must be verified by batch/lot number and may be used without cleaning if PFAS levels are less than half the Minimum Level (ML, see Table 6).
- 4.2.1** All glass equipment that is used in the preparation or storage of reagents is cleaned by washing with detergent and baking in a kiln or furnace (Section 6.2.2). After detergent washing, glassware should be rinsed immediately with reagent water. Prior to use, baked glassware must be solvent rinsed and then air dried. A solvent rinse procedure using methanolic ammonium hydroxide (1%), toluene, and methanol is recommended.
- 4.2.2** All parts of the SPE manifold must be cleaned between samples by sonicating in methanolic ammonium hydroxide (1%) and air drying prior to use. Smaller parts, like the needles, adapters, reservoirs, and stopcocks associated with the manifold require rinsing

with tap water prior to sonicating in methanolic ammonium hydroxide (1%) and air drying. When in use, after loading the samples but prior to elution procedures, the chamber must be rinsed with methanolic ammonium hydroxide (1%).

- 4.2.3** All equipment used in the filleting, dissecting, shucking, compositing, and homogenization of tissue must be cleaned with detergent and hot water, then rinsed with ultra-pure water followed by a series of solvent rinses. A typical solvent rinse procedure would be acetone, followed by toluene, and then dichloromethane.
- 4.3** All materials used in the analysis must be demonstrated to be free from interferences by running method blanks (Section 9.5) at the beginning and with each sample batch (samples started through the extraction process on a given analytical batch to a maximum of 20 field samples).
 - 4.3.1** The reference matrix must simulate, as closely as possible, the sample matrix being tested. Ideally, the reference matrix should not contain PFAS in detectable amounts but should contain potential interferents in the concentrations expected to be found in the samples to be analyzed.
 - 4.3.2** For tissue, chicken breast or other similar animal tissue (see Section 7.2.3) may be used as the reference matrix. The laboratory must verify that the source product used does not contain PFAS in detectable amounts.
 - 4.3.3** When a reference matrix that simulates the sample matrix under test is not available, reagent water (Section 7.2.1) can be used to simulate water samples and Ottawa sand and/or reagent-grade sand (Section 7.2.2) can be used to simulate soils.
- 4.4** Interferences co-extracted from samples will vary considerably from source to source, depending on the diversity of the site being sampled. Interfering compounds may be present at concentrations several orders of magnitude higher than the native PFAS. Because low levels of PFAS are measured by this method, elimination of interferences is essential. The cleanup steps given in Section 12.0 can be used to reduce or eliminate these interferences and thereby permit reliable determination of the PFAS at the levels shown in Table 6. The most frequently encountered interferences are fluoropolymers; however, when analyzing whole fish samples, bile salts (e.g., Taurodeoxycholic Acid [TDCA]) can interfere in the chromatography. For this reason, analysis of a standard containing TDCA is required as part of establishing the initial chromatographic conditions (see Sections 10.2.2.5 and 10.3.5).
- 4.5** Each piece of reusable glassware may be numbered to associate that glassware with the processing of a particular sample. This may assist the laboratory in tracking possible sources of contamination for individual samples, identifying glassware associated with highly contaminated samples that may require extra cleaning, and determining when glassware should be discarded.

5.0 Safety

- 5.1** The toxicity or carcinogenicity of each chemical used in this method has not been precisely determined; however, each compound should be treated as a potential health hazard. Exposure to these compounds should be reduced to the lowest possible level.
 - 5.1.1** PFOA has been described as likely to be carcinogenic to humans. Pure standards should be handled by trained personnel, with suitable protection to skin and eyes, and care should be taken not to breathe the vapors or ingest the materials.

- 5.1.2** It is recommended that the laboratory purchase dilute standard solutions of the analytes in this method. However, if primary solutions are prepared, they must be prepared in a hood, following universal safety measures.
- 5.2** The laboratory is responsible for maintaining a current awareness file of Occupational Safety and Health Administration (OSHA) regulations regarding the safe handling of the chemicals specified in this method. A reference file of safety data sheets (SDS) should also be made available to all personnel involved in these analyses. It is also suggested that the laboratory perform personal hygiene monitoring of each analyst who uses this method and that the results of this monitoring be made available to the analyst. Additional information on laboratory safety can be found in References 1-4. The references and bibliography at the end of Reference 3 are particularly comprehensive in dealing with the general subject of laboratory safety.
- 5.3** Samples suspected to contain these compounds are handled using essentially the same techniques employed in handling radioactive or infectious materials. Well-ventilated, controlled access laboratories are required. Assistance in evaluating the health hazards of particular laboratory conditions may be obtained from certain consulting laboratories and from State Departments of Health or Labor, many of which have an industrial health service. Each laboratory must develop a strict safety program for handling these compounds.
- 5.3.1** Facility – When finely divided samples (dusts, soils, dry chemicals) are handled, all operations (including removal of samples from sample containers, weighing, transferring, and mixing) should be performed in a glove box demonstrated to be leak tight or in a fume hood demonstrated to have adequate air flow. Gross losses to the laboratory ventilation system must not be allowed. Handling of the dilute solutions normally used in analytical and animal work presents no inhalation hazards except in the case of an accident.
- 5.3.2** Protective equipment – Disposable plastic gloves, apron or lab coat, safety glasses or mask, and a glove box or fume hood adequate for radioactive work should be used. During analytical operations that may give rise to aerosols or dusts, personnel should wear respirators equipped with activated carbon filters. Eye protection (preferably full-face shields) must be worn while working with exposed samples or pure analytical standards. Latex gloves are commonly used to reduce exposure of the hands.
- 5.3.3** Training – Workers must be trained in the proper method of removing contaminated gloves and clothing without contacting the exterior surfaces.
- 5.3.4** Personal hygiene – Hands and forearms should be washed thoroughly after each manipulation and before breaks (coffee, lunch, and shift).
- 5.3.5** Confinement – Isolated work areas posted with signs, segregated glassware and tools, and plastic absorbent paper on bench tops will aid in confining contamination.
- 5.3.6** Waste Handling – Good technique includes minimizing contaminated waste. Plastic bag liners should be used in waste cans. Janitors and other personnel should be trained in the safe handling of waste.
- 5.3.7** Laundry – Clothing known to be contaminated should be collected in plastic bags. Persons that convey the bags and launder the clothing should be advised of the hazard and trained in proper handling. The clothing may be put into a washer without contact if the launderer knows of the potential problem. The washer should be run through a cycle before being used again for other clothing.

- 5.4 Biosolids samples may contain high concentrations of biohazards and must be handled with gloves and opened in a fume hood or biological safety cabinet to prevent exposure. Laboratory staff should know and observe the safety procedures required in a microbiology laboratory that handles pathogenic organisms when handling biosolids samples.

6.0 Equipment and Supplies

Note: Brand names, suppliers, and part numbers are for illustration purposes only and no endorsement is implied. Equivalent performance may be achieved using apparatus and materials other than those specified here. Meeting the performance requirements of this method is the responsibility of the laboratory.

6.1 Sampling equipment for discrete or composite sampling

6.1.1 Sample bottles and caps

Note: Do not use PTFE-lined caps on sample containers.

- 6.1.1.1** Liquid samples (waters, sludges, and similar materials containing < 50 mg solids per sample) – Sample bottle, HDPE, with linerless HDPE or polypropylene caps.
-

Note: At least two aliquots of aqueous samples are collected to allow sufficient volume for the determination of percent solids and for pre-screening analysis. One aliquot should be collected in a 500-mL container while the second aliquot may be collected in a smaller sample container (e.g., 250-mL or 125-mL).

- 6.1.1.2** Solid samples (soils, sediments, and biosolids that contain more than 50 mg solids) – Sample bottle or jar, wide-mouth, HDPE, 500-mL, with linerless HDPE or polypropylene caps.

- 6.1.1.3** Tissue samples – Sample jar, wide-mouth HDPE, 100-mL, with linerless HDPE or polypropylene caps.

- 6.1.2** Compositing equipment – Automatic or manual compositing system incorporating containers cleaned per bottle cleaning procedure above. Only HDPE tubing must be used. If the sampler uses a peristaltic pump, a minimum length of compressible silicone rubber tubing may be used in the pump only. Before use, the tubing must be thoroughly rinsed with methanol, followed by repeated rinsing with reagent water to minimize sample contamination. An integrating flow meter is used to collect proportional composite samples.

6.2 Equipment for glassware cleaning

Note: If blanks from bottles or other glassware, show no detectable PFAS contamination when using fewer cleaning steps than required above, unnecessary cleaning steps and equipment may be eliminated.

6.2.1 Laboratory sink with overhead fume hood

6.2.2 Kiln – Capable of reaching 450 °C within 2 hours and maintaining 450 - 500 °C $\pm$ 10 °C, with temperature controller and safety switch (Cress Manufacturing Co., Santa Fe Springs, CA, B31H, X31TS, or equivalent). For safety, the kiln or furnace should be vented outside the laboratory, or to a trapping system.

6.3 Equipment for sample preparation

6.3.1 Polyethylene gloves

6.3.2 Laboratory fume hood (of sufficient size to contain the sample preparation equipment listed below)

6.3.3 Glove box (optional)

6.3.4 Tissue homogenizer – Pro Scientific PRO400DS homogenizer or equivalent with stainless steel macro-shaft and turbo-shear blade

6.3.5 Meat grinder – Hobart, or equivalent, with 3- to 5-mm holes in inner plate

6.3.6 Equipment for determining percent moisture

6.3.6.1 Oven – Capable of maintaining a temperature of 110 $\pm$ 5 °C

6.3.6.2 Desiccator

6.3.7 Balances

6.3.7.1 Analytical – Capable of weighing 0.1 mg

6.3.7.2 Top loading – Capable of weighing 10 mg

6.3.8 Aluminum foil

6.3.9 Disposable spoons, 10 mg, polypropylene or stainless steel

6.3.10 Ultrasonic mixer (sonicator)

6.3.11 HDPE bottles, with linerless HDPE or polypropylene caps – 60 mL

6.3.12 pH Paper, range 0-14 - (Whatman® Panpeha™ or equivalent), 0.5-unit readability

6.3.13 Analog or digital vortex mixer, single or multi-tube (Fisher Scientific 02-215-452, or equivalent)

6.3.14 Volumetric flasks, Class A

6.3.15 Disposable polypropylene collection tubes (13 x 100 mm, 8 mL)

6.3.16 Variable speed mixing table (Fisherbrand™ Nutating mixer or equivalent)

6.4 Filtration

- 6.4.1 Silanized glass wool (Sigma-Aldrich, Cat # 20411 or equivalent) – store in a clean glass jar and rinsed with methanol (2 times) prior to use.
- 6.4.2 Disposable syringe filter, 25-mm, 0.2-µm Nylon membrane, PALL/Acrodisc or equivalent
- 6.4.3 Glass fiber filter, 47 mm, 1 µm, PALL A/E or equivalent

6.5 Centrifuge apparatus

- 6.5.1 Centrifuge (Thermo Scientific Legend RT+, 16 cm rotor, or equivalent), capable of reaching at least 3000 rpm
- 6.5.2 Centrifuge tubes – Disposable polypropylene centrifuge tubes (50 mL)

6.6 Pipettes

- 6.6.1 Norm-Ject® syringe (or equivalent), polypropylene/HDPE, 5 mL
- 6.6.2 Variable volume pipettes with disposable HDPE or polypropylene tips (10 µL to 5 mL) – used for preparation of calibration standards and spiked samples.
- 6.6.3 **Disposable** glass pipets
- 6.6.4 **Calibrated** mechanical pipettes or Hamilton graduated syringes

6.7 Solid-Phase Extraction

- 6.7.1 Solid-phase extraction (SPE) cartridges (Waters Oasis WAX 150 mg, Cat # 186002493 or equivalent). The SPE sorbent must have a pKa above 8 so that it remains positively charged during the extraction.

Note: SPE cartridges with different bed volume (e.g., 500 mg) may be used; however, the laboratory must demonstrate that the bed volume does not negatively affect analyte absorption and elution, by performing the initial demonstration of capability analyses described in Section 9.2.

- 6.7.2 Vacuum manifold for SPE Cartridges (Waters™ extraction manifold #WAT200607 or equivalent)

6.8 Evaporation

- 6.8.1 Automatic or manual solvent evaporation system (TurboVap® LV or equivalent)
- 6.8.2 Evaporation/concentrator tubes: 60 mL clear glass vial, 30 x 125 mm, without caps (Wheaton Cat # W226060 or equivalent). Cover with foil if required.

6.9 Vials

- 6.9.1 Snap cap/crimp top vials, 300 µL, polypropylene (12 x 32 mm) – used in sample pre-screening (DWK Life Sciences Cat # 225180 or equivalent)
- 6.9.2 Polypropylene crimp/snap vials, 1 mL (Agilent Cat # 5182-0567 or equivalent)
- 6.9.3 Clear snap cap, PVDC film/white silicone, 11 mm (American Chromatography Supplies Cat # C299-11 or equivalent)
- 6.9.4 Single step filter vials (Restek Thomson SINGLE StEP® Standard Filter Vials, 0.2 µm Nylon membrane, with Black Preslit caps Cat # 25891 or equivalent) – used in sample pre-screening.

6.10 Instrument

- 6.10.1 Ultra high-performance liquid chromatograph (UPLC also called UHPLC) or high-performance liquid chromatograph (HPLC) equipped with tandem quadrupole mass spectrometer (Waters Xevo TQ-S Micro or equivalent).
 - 6.10.2 C18 column, 1.7 µm, 50 x 2.1 mm (Waters Acquity UPLC® BEH or equivalent)
 - 6.10.3 Guard column (Phenomenex Kinetex® Evo C18 or equivalent)
 - 6.10.4 Trap/delay column (Purospher Star RP-18 endcapped [3 µm] Hibar® RT 50-4 or equivalent)
- 6.11 Bottles, HDPE or glass, with linerless HDPE or polypropylene caps. Various sizes. To store prepared reagents.

7.0 Reagents and standards

7.1 Reagents

Reagents prepared by the laboratory may be stored in either glass or HDPE containers. Proper cleaning procedures (Section 4.2) must be followed prior to using the containers.

- 7.1.1 Acetic acid - ACS grade or equivalent, store at room temperature
- 7.1.2 Acetic acid (0.1%) - dissolve acetic acid (1 mL) in reagent water (1 L), store at room temperature, replace after 3 months. *This reagent is used only for sample extract dilution.*
- 7.1.3 Acetonitrile – UPLC grade or equivalent, verified before use, store at room temperature
- 7.1.4 Ammonium acetate - (Caledon Ultra LC/MS grade, or equivalent), store at 2-8° C, replace 2 years after opening date
- 7.1.5 Ammonium hydroxide - certified ACS+ grade or equivalent, 30% in water, store at room temperature

- 7.1.6** Aqueous ammonium hydroxide (3%) - add ammonium hydroxide (10 mL, 30%) to reagent water (90 mL), store at room temperature, replace after 3 months
- 7.1.7** Methanolic ammonium hydroxide
- 7.1.7.1** Methanolic ammonium hydroxide (0.3%) - add ammonium hydroxide (1 mL, 30%) to methanol (99 mL), store at room temperature, replace after 1 month
 - 7.1.7.2** Methanolic ammonium hydroxide (1%) - add ammonium hydroxide (3.3 mL, 30%) to methanol (97 mL), store at room temperature, replace after 1 month
 - 7.1.7.3** Methanolic ammonium hydroxide (2%) - add ammonium hydroxide (6.6 mL, 30%) to methanol (93.4 mL), store at room temperature, replace after 1 month
- 7.1.8** Methanolic potassium hydroxide (0.05 M) – add 3.3 g of potassium hydroxide to 1 L of methanol, store at room temperature, replace after 3 months
- 7.1.9** Methanol with 4% water, 1% ammonium hydroxide and 0.625% acetic acid - add ammonium hydroxide (3.3 mL, 30%), reagent water (1.7 mL) and acetic acid (0.625 mL) to methanol (92 mL), store at room temperature, replace after 1 month. This solution is used to prepare the instrument blank (Section 7.3.6).
- 7.1.10** Eluent A – Acetonitrile, Caledon Ultra LCMS grade or equivalent
- 7.1.11** Eluent B - 2 mM ammonium acetate in 95:5 water/acetonitrile. Dissolve 0.154 g of ammonium acetate (Section 7.1.4) in 950 mL of water and 50 mL of acetonitrile (Caledon Ultra LCMS grade, or equivalent). Store at room temperature, shelf life 2 months.
- 7.1.12** Formic acid - (greater than 96% purity or equivalent), verified by lot number before use, store at room temperature
- 7.1.13** Formic acid
- 7.1.13.1** Formic acid (aqueous, 0.1 M) - dissolve formic acid (4.6 g) in reagent water (1 L), store at room temperature, replace after 2 years
 - 7.1.13.2** Formic acid (aqueous, 0.3 M) - dissolve formic acid (13.8 g) in reagent water (1 L), store at room temperature, replace after 2 years
 - 7.1.13.3** Formic acid (aqueous, 5% v/v) - mix 5 mL formic acid with 95 mL reagent water, store at room temperature, replace after 2 years
 - 7.1.13.4** Formic acid (aqueous, 50% v/v) - mix 50 mL formic acid with 50 mL reagent water, store at room temperature, replace after 2 years
 - 7.1.13.5** Formic acid (methanolic 1:1, 0.1 M formic acid/methanol) - mix equal volumes of methanol and 0.1 M formic acid, store at room temperature, replace after 2 years
- 7.1.14** Methanol - (HPLC grade or better, 99.9% purity), verified by lot number before use, store at room temperature

- 7.1.15** Potassium hydroxide – certified ACS or equivalent, store at room temperature, replace after 2 years
- 7.1.16** Reagent water – Laboratory reagent water, test by lot/batch number for residual PFAS content
- 7.1.17** Carbon – EnviCarb® 1-M-USP or equivalent, verified by lot number before use, store at room temperature. Loose carbon allows for better adsorption of interferent organics.

Note: The single-laboratory validation laboratory achieved better performance with loose carbon than carbon cartridges. Loose carbon will be used for the multi-laboratory validation to set statistically based method criteria. Once the method is multi-laboratory validated, laboratories will have the flexibility to use carbon cartridges as long as all method QC criteria are met.

- 7.1.18** Toluene – HPLC grade, verified by lot number before use. Store at room temperature.
- 7.1.19** Acetone - Pesticide grade, verified by lot number before use in rinsing tissue dissection and processing equipment.
- 7.1.20** Dichloromethane (methylene chloride), pesticide grade, verified by lot number before use in rinsing tissue dissection and processing equipment.
- 7.2** Reference matrices – Matrices in which PFAS and interfering compounds are not detected by this method. These matrices are to be used to prepare the batch QC samples (e.g., method blank, and ongoing precision and recovery sample).
- 7.2.1** Reagent water – purified water, Type I
- 7.2.2** Solids reference matrix – Ottawa or reagent-grade sand
- 7.2.3** Tissue reference matrix – chicken breast or similar animal tissue
- 7.3** Standard solutions – Prepare from materials of known purity and composition or purchase as solutions or mixtures with certification to their purity, concentration, and authenticity. Observe the safety precautions in Section 5.

Purchase of commercial standard solutions or mixtures is highly recommended for this method; however, when these are not available, preparation of stock solutions from neat materials may be necessary. If the chemical purity is 98% or greater, the weight may be used without correction to calculate the concentration of the standard. Dissolve an appropriate amount of assayed reference material in the required solvent. For example, weigh 10 to 20 mg of an individual compound to three significant figures in a 10-mL ground-glass-stoppered volumetric flask and fill to the mark with the required solvent. Once the compound is completely dissolved, transfer the solution to a clean vial and cap.

When not being used, store standard solutions in the dark at less than 4 °C unless the vendor recommends otherwise in screw-capped vials with foiled-lined caps. Place a mark on the vial at the level of the solution so that solvent loss by evaporation can be detected. Replace the solution if solvent loss has occurred.

Note: Native PFAS standards are available from several suppliers. Isotopically labeled compounds are available from Cambridge Isotope Laboratories and Wellington Laboratories, but may also be available from other suppliers. Listing of these suppliers does not constitute a recommendation or endorsement for use. All diluted solutions must be stored in glass or HDPE containers that have been thoroughly rinsed with methanol.

¹⁸O-mass labeled perfluoroalkyl sulfonates may undergo isotopic exchange with water under certain conditions, which lowers the isotopic purity of the standards over time.

The laboratory must maintain records of the certificates for all standards for traceability purposes. Copies of the certificates must be provided as part of the data packages in order to check that proper calculations were performed.

- 7.3.1** Extracted Internal Standard (EIS) – (a.k.a. isotopically labeled compound) Prepare the EIS solution containing the isotopically labeled compounds listed in Table 3 as extracted internal standards in methanol from prime stocks. An aliquot of EIS solution, typically 50 µL, is added to each sample prior to extraction. Table 3 presents the nominal amounts of EIS compounds added to each sample. The list of isotopically labeled compounds in Table 3 represents the compounds that were available at the time this method was validated. Other isotopically labeled compounds may be used as they become available.
- 7.3.2** Non-Extracted Internal Standard (NIS) - The NIS solution containing the isotopically labeled compounds listed in Table 3 as non-extracted internal standards is prepared in methanol from prime stock. An aliquot of NIS solution, typically 50 µL, is added to each sample prior to instrumental analysis. Table 3 presents the nominal amounts of NIS compounds added to each sample.
- 7.3.3** Native Standards Solution - Prepare a spiking solution, containing the method analytes listed in Table 4, in methanol from prime stocks. The solution is used to prepare the calibration standards and to spike the known reference QC samples that are analyzed with every batch. Quantitative standards containing a mixture of branched and linear isomers must be used for method analytes if they are commercially available. Currently, these include PFOS, PFHxS, NMeFOSAA, and NeFOSAA.
- 7.3.4** Calibration standard solutions – A series of calibration solutions containing the target analytes and the ¹³C-, ¹⁸O-, and deuterium-labeled extracted internal standards (EIS) and non-extracted internal standards (NIS) is used to establish the initial calibration of the analytical instrument. The concentration of the method analytes in the solutions varies to encompass the working range of the instrument, while the concentrations of the EIS and NIS remain constant. The calibration solutions are prepared using methanol, methanolic ammonium hydroxide (2%), water, acetic acid and the method analyte and isotopically labeled compound standard solutions. After dilution, the final solution will match the solvent mix of sample extracts, which contain methanol with 4% water, 1% ammonium hydroxide and 0.625% acetic acid (Section 7.1.9). Calibration standard solutions do not undergo solid phase extraction/cleanup.

Concentrations for seven calibration solutions are presented in Table 4. A minimum of six contiguous calibration standards are required for a valid analysis when using a linear calibration model, with at least five of the six calibration standards being within the quantitation range (e.g., from the LOQ to the highest calibration standard). If a second-order calibration model is used, then a minimum of seven calibration standards are

required, with at least six of the seven calibration standards within the quantitation range. The lowest level calibration standard must meet a signal-to-noise ratio of 3:1 and be at a concentration less than or equal to the Limit of Quantitation (LOQ). All initial calibration requirements listed in Table 7 must be met. An instrument sensitivity check (ISC) standard at the concentration of the lowest calibration standard within the quantitation range is required to be analyzed at the beginning of the analytical run (Section 10.3.3.1 and Section 13.3). A mid-level calibration solution is analyzed at least every ten samples or less, on an ongoing basis for the purpose of calibration verification. A mid-level calibration verification (CV) standard must also be analyzed after all sample analyses in order to bracket the analytical batch.

Note: Additional calibration standards, at levels lower than the lowest calibration standard listed in the method, may be added to accommodate a lower limit of quantitation if the instrument sensitivity allows. Calibration standards at the high end of the calibration may be eliminated if the linearity of the instrument is exceeded or at the low end if those calibration standards do not meet the S/N ratio criterion of 3:1, as long as the required number of calibration points is met. All analytes with commercially available stable isotope analogues must be quantified using isotope dilution.

- 7.3.5** Qualitative Standards - Standards that contain mixtures of the branched and linear isomers of the method analytes and that are used for comparison against suspected branched isomer peaks in field samples. These qualitative standards are **not** required for those analytes where the quantitative standards in Section 7.3.3 already contain the branched and linear isomers. Qualitative standards that are currently commercially available include PFOA, PFNA, PFOSA, NMeFOSA, NEtFOSA, NEtFOSE, and NMeFOSE.
- 7.3.6** Instrument Blank – During the analysis of a batch of samples, a solvent blank is analyzed after samples containing high level of target compounds (e.g., calibration, CV) to monitor carryover from the previous injection. The injection blank consists of the solution in Section 7.1.9 fortified with the EIS and NIS for quantitation purposes.
- 7.3.7** Stability of solutions – Standard solutions used for quantitative purposes (Sections 7.3.1 through 7.3.5) should be assayed periodically (e.g., every 6 months) against certified standard reference materials (SRMs) from the National Institute of Science and Technology (NIST), if available, or certified reference materials from a source that will attest to the authenticity and concentration, to assure that the composition and concentrations have not changed.
- 7.4** Sodium iodide/cesium iodide mass calibration solution – 2 mg/mL NaI and 50 µg/mL CsI in (1:1) isopropyl alcohol:water (Waters 700000889, or equivalent) or other solution, based on manufacturer’s specifications.
- 7.5** Taurodeoxycholic Acid (TDCA) or Sodium taurodeoxycholate hydrate – (Sigma Aldrich 580221-5GM, or equivalent). This compound is used to evaluate the chromatographic program relative to the risk of an interference from bile salts in tissue samples. Prepare solution at a concentration of 100 mg/L in the same solvent as the calibration standards.

8.0 Sample collection, preservation, storage, and holding times

- 8.1** Collect samples in HDPE containers following conventional sampling practices (Reference 5). All sample containers must have linerless HDPE or polypropylene caps. Other sample collection techniques, or sample volumes may be used, if documented.

8.2 Aqueous samples

- 8.2.1** Samples that flow freely are collected as grab samples or in refrigerated bottles using automatic sampling equipment. Collect 500 mL of sample (other than leachates) in an HDPE bottle. Do not fill the bottle past the shoulder, to allow room for expansion during frozen storage.

Note: Collect at least two aliquots of all aqueous samples to allow sufficient volume for the determination of percent solids and for pre-screening analysis. That second aliquot may be collected in a smaller sample container (e.g., 250-mL or 125-mL).

Because the target analytes are known to bind to the interior surface of the sample container, the entire aqueous sample that is collected must be prepared and analyzed and subsampling avoided whenever possible. Therefore, if a sample volume smaller than 500 mL is to be used for analysis, collect the sample in an appropriately sized HDPE container.

- 8.2.2** Leachate samples from landfills can present significant challenges and therefore only 100 mL of sample is collected for the analysis. Collect two 100-mL leachate sample aliquots in a similar manner as described in Section 8.2.1, using appropriately sized containers.
- 8.2.3** Maintain all aqueous samples protected from light at 0 - 6 °C from the time of collection until shipped to the laboratory. Samples must be shipped as soon as practical with sufficient ice to maintain the sample temperature below 6 °C during transport and be received by the laboratory within 48 hours of collection. The laboratory must confirm that the sample temperature is 0 - 6 °C upon receipt. Once received by the laboratory, the samples must be stored at ≤ -20 °C until sample preparation.

8.3 Solid (soil, sediment, biosolid), excluding tissue

- 8.3.1** Collect samples as grab samples using wide-mouth jars and fill no more than ¾ full (see Section 6.1.1.2 for container size and type).
- 8.3.2** Maintain solid samples protected from light (in HDPE containers) at 0 - 6 °C from the time of collection until receipt at the laboratory. The laboratory must confirm that the sample temperature is 0 - 6 °C upon receipt. Once received by the laboratory, the samples must be stored at ≤ -20 °C until sample preparation.

8.4 Fish and other tissue samples

The nature of the tissues of interest may vary by project. Field sampling plans and protocols should explicitly state the samples to be collected and if any processing will be conducted in the field (e.g., filleting of whole fish or removal of organs). All field procedures must involve materials and equipment that have been shown to be free of PFAS.

- 8.4.1** Fish may be cleaned, filleted, or processed in other ways in the field, such that the laboratory may expect to receive whole fish, fish fillets, or other tissues for analysis.

- 8.4.2** If whole fish are collected, wrap the fish in aluminum foil or food-grade polyethylene tubing, and maintain at 0 - 6 °C from the time of collection until receipt at the laboratory, to a maximum time of 24 hours. If a longer transport time is necessary, freeze the sample before shipping. Ideally, fish should be frozen upon collection and shipped to the laboratory on dry ice.
- 8.4.3** Once received by the laboratory, the samples must be maintained protected from light at ≤ -20 °C until prepared. Store unused samples in HDPE containers or wrapped in aluminum foil at ≤ -20 °C.

8.5 Holding times

- 8.5.1** Aqueous samples (including leachates) should be analyzed as soon as possible; however, samples may be held in the laboratory for up to 90 days from collection, when stored at ≤ -20 °C and protected from the light. When stored at 0 - 6 °C and protected from the light, aqueous samples may be held for up to 28 days, with the caveat that issues were observed with certain perfluorooctane sulfonamide ethanols and perfluorooctane sulfonamidoacetic acids after 7 days. These issues are more likely to elevate the observed concentrations of other PFAS compounds via the transformation of these precursors if they are present in the sample.
- 8.5.2** Solid samples (soils and sediments) and tissue samples may be held for up to 90 days, if stored by the laboratory in the dark at either 0 - 6 °C or ≤ -20 °C, with the caveat that samples may need to be extracted as soon as possible if NFDHA is an important analyte.
- 8.5.3** Biosolids samples may be held for up to 90 days, if stored by the laboratory in the dark at 0 - 6 °C or at -20 °C. Because microbiological activity in biosolids samples at 0 - 6 °C may lead to production of gases which may cause the sample to be expelled from the container when it is opened, as well as producing noxious odors, EPA recommends that samples be frozen if they need to be stored for more than a few days before extraction.
- 8.5.4** Store sample extracts in the dark at less than 0 - 4 °C until analyzed. If stored in the dark at less than 0 - 4 °C, sample extracts may be stored for up to 90 days, with the caveat that issues were observed for some ether sulfonates after 28 days. These issues may elevate the observed concentrations of the ether sulfonates in the extract over time. Samples may need to be extracted as soon as possible if NFDHA is an important analyte.

9.0 Quality Control

- 9.1** Each laboratory that uses this method is required to operate a formal quality assurance program (Reference 6). The minimum requirements of this program consist of an initial demonstration of laboratory capability, analysis of samples spiked with isotopically labeled compounds to evaluate and document data quality, and analysis of standards and blanks as tests of continued performance. Laboratory performance is compared to established performance criteria to determine if the results of analyses meet the performance characteristics of the method.

If the method is to be applied to a sample matrix other than water (e.g., soils, biosolids, tissue), the appropriate alternative reference matrix (Sections 7.2.2 - 7.2.3) is substituted for the reagent water matrix (Section 7.2.1) in all performance tests.

- 9.1.1** The laboratory must make an initial demonstration of the ability to generate acceptable precision and recovery with this method. This demonstration is given in Section 9.2.
- 9.1.2** In recognition of advances that are occurring in analytical technology, and to overcome matrix interferences, the laboratory is permitted certain options to improve separations or lower the costs of measurements. These options include alternative extraction, concentration, and cleanup procedures, and changes in sample volumes, columns, and detectors. Alternative determinative techniques and changes that degrade method performance, are *not* allowed without prior review and approval.

Note: For additional flexibility to make modifications without prior EPA review, see 40 CFR Part 136.6.

- 9.1.2.1** Each time a modification is made to this method, the laboratory is required to repeat the procedure in Section 9.2. If calibration will be affected by the change, the instrument must be recalibrated per Section 10. Once the modification is demonstrated to produce results equivalent or superior to results produced by this method as written, that modification may be used routinely thereafter, so long as the other requirements in this method are met (e.g., isotopically labeled compound recovery).
- 9.1.2.2** The laboratory is required to maintain records of any modifications made to this method. These records include the following, at a minimum:
- a) The names, titles, business addresses, and telephone numbers of the analyst(s) that performed the analyses and modification, and of the quality control officer that witnessed and will verify the analyses and modifications.
 - b) A listing of pollutant(s) measured, by name and CAS Registry number.
 - c) A narrative stating reason(s) for the modifications (see Section 1.6).
 - d) Results from all quality control (QC) tests comparing the modified method to this method, including:
 - i. Calibration (Section 10)
 - ii. Calibration verification (Section 14.3)
 - iii. Initial precision and recovery (Section 9.2.1)
 - iv. Isotopically labeled compound recovery (Section 9.3)
 - v. Analysis of blanks (Section 9.5)
 - vi. Accuracy assessment (Section 9.4)
 - e) Data that will allow an independent reviewer to validate each determination by tracing the instrument output (peak height, area, or other signal) to the final result. These data are to include:
 - i. Sample numbers and other identifiers
 - ii. Extraction dates
 - iii. Analysis dates and times
 - iv. Analysis sequence/run chronology
 - v. Sample weight or volume (Section 11)
 - vi. Extract volume prior to each cleanup step (Section 12)

- vii. Extract volume after each cleanup step (Section 12)
- viii. Final extract volume prior to injection (Section 12)
- ix. Injection volume (Section 13.3)
- x. Dilution data, differentiating between dilution of a sample or extract (Section 15.3)
- xi. Instrument
- xii. Column (dimensions, liquid phase, solid support, film thickness, etc.)
- xiii. Operating conditions (temperatures, temperature program, flow rates)
- xiv. Detector (type, operating conditions, etc.)
- xv. Chromatograms, printer tapes, and other recordings of raw data
- xvi. Quantitation reports, data system outputs, and other data to link the raw data to the results reported

9.1.2.3 Alternative columns and column systems – If a column or column system other than those specified in this method is used, that column or column system must meet all the requirements of this method.

Note: *The use of alternative columns or programs will likely result in a different elution order.*

9.1.3 Analyses of method blanks are required on an on-going basis to demonstrate the extent of background contamination in any reagents or equipment used to prepare and analyze field samples (Section 4.3). The procedures and criteria for analysis of a method blank are described in Section 9.5.

9.1.4 The laboratory must spike all samples with isotopically labeled compounds to monitor method performance. This test is described in Section 9.3. When results of these spikes indicate atypical method performance for samples, the samples are diluted to evaluate whether the performance issue is caused by the sample matrix. Procedures for dilution are given in Section 15.3.

9.1.5 The laboratory must, on an ongoing basis, demonstrate that the analytical system is in control through calibration verification and the analysis of ongoing precision and recovery standards (OPR), spiked at low (LLOPR) and mid-level, and blanks. These procedures are given in Sections 14.1 through 14.7.

9.1.6 The laboratory must maintain records to define the quality of data generated. Development of accuracy statements is described in Section 9.4.

9.2 Initial Demonstration of Capability

9.2.1 Initial precision and recovery (IPR) – To establish the ability to generate acceptable precision and recovery, the laboratory must perform the following operations for each sample matrix type to which the method will be applied by that laboratory.

9.2.1.1 Extract, concentrate, and analyze four aliquots of the matrix type to be tested (Section 7.2.1 through 7.2.3), spiked with 200 µL of the native standard solution (Section 7.3.3), 50 µL of the EIS solution (Section 7.3.1), and 50 µL of NIS solution (Section 7.3.2). At least one method blank, matching the matrix being analyzed, must be prepared with the IPR batch. In the event that more than one MB was prepared and analyzed with the IPR batch, all blank results must be reported. All sample processing steps that are to be used for processing samples,

including preparation and extraction (Sections 11.2 – 11.4), cleanup (Section 12.0) and concentration (Section 12.0), must be included in this test.

9.2.1.2 Using results of the set of four analyses, compute the average percent recovery (R) of the extracts and the relative standard deviation (RSD) of the concentration for each target and EIS compound.

9.2.1.3 For each native and isotopically labeled compound, compare RSD and % recovery with the corresponding limits for initial precision and recovery in Table 5. If RSD and R for all compounds meet the acceptance criteria, system performance is acceptable, and analysis of blanks and samples may begin. If, however, any individual RSD exceeds the precision limit or any individual R falls outside the range for recovery, system performance is unacceptable for that compound. Correct the problem and repeat the test (Section 9.2).

9.2.2 Method detection limit (MDL) - Each laboratory must also establish MDLs for all the analytes using the MDL procedure at 40 CFR Part 136, Appendix B. An MDL determination must be performed for all compounds. The minimum level of quantification (ML) is then calculated by multiplying the MDL by 3.18 and rounding the result to the nearest 1, 2 or 5×10^n , where n is zero or an integer. Example matrix-specific detection limits are listed in Table 6.

9.3 To assess method performance on the sample matrix, the laboratory must spike all samples with the isotopically labeled compound standard solution (Section 7.3.1) and all sample extracts with the NIS spiking solution (Section 7.3.2).

9.3.1 Analyze each sample according to the procedures in Sections 11.0 through 16.0.

9.3.2 Compute the percent recovery of the isotopically labeled compound using the non-extracted internal standard method (Section 15.2) and the equation in Section 14.5.2.

9.3.3 The recovery of each isotopically labeled compound must be within the limits in Tables 9 and 10 (*once the tables are finalized*). If the recovery of any compound falls outside of these limits, method performance is unacceptable for that compound in that sample. Additional cleanup procedures must then be employed to attempt to bring the recovery within the normal range. If the recovery cannot be brought within the normal range after all cleanup procedures have been employed, water samples are diluted, and smaller amounts of soils, biosolids, sediments, and other matrices are prepared and analyzed, per Section 15.3.

9.4 Recovery of isotopically labeled compounds from samples must also be assessed and records maintained.

9.4.1 After the analysis of 30 samples of a given matrix type (water, soil, biosolids, tissues, etc.) for which the isotopically labeled compounds pass the tests in Section 9.3, compute the R and the standard deviation of the percent recovery (S_R) for the isotopically labeled compounds only. Express the assessment as a percent recovery interval from $R - 2S_R$ to $R + 2S_R$ for each matrix. For example, if $R = 90\%$ and $S_R = 10\%$ for five analyses of soil, the recovery interval is expressed as 70 to 110%.

9.4.2 Update the accuracy assessment for each isotopically labeled compound in each matrix on a regular basis (e.g., after each five to ten new measurements).

9.5 Method blanks – A method blank is analyzed with each sample batch (Section 4.3) to demonstrate freedom from contamination. The matrix for the method blank must be similar to the sample matrix for the batch (e.g., reagent water blank [Section 7.2.1], solids matrix blank [Section 7.2.2], or tissue blank [Section 7.2.3]).

9.5.1 Analyze the cleaned extract (Section 12.0) of the method blank aliquot before the analysis of the OPRs (Section 14.5).

9.5.2 If any PFAS is found in the blank at 1) at a concentration greater than the ML for the analyte, 2) at a concentration greater than one-third the regulatory compliance limit, or 3) at a concentration greater than one-tenth the concentration in a sample in the extraction batch, whichever is greatest, analysis of samples must be halted, and the problem corrected. Other project-specific requirements may apply; therefore, the laboratory may adopt more stringent acceptance limits for the method blank at their discretion. If the contamination is traceable to the extraction batch, samples affected by the blank must be re-extracted and analyzed, provided enough sample volume is available and the sample are still within holding time.

If, continued re-testing results in repeated blank contamination, the laboratory must document and report the failures (e.g., as qualifiers on results), unless the failures are not required to be reported as determined by the regulatory/control authority. Results associated with blank contamination for an analyte regulated in a discharge cannot be used to demonstrate regulatory compliance. QC failures do not relieve a discharger or permittee of reporting timely results.

9.6 The specifications contained in this method can be met if the apparatus used is calibrated properly and then maintained in a calibrated state. The standards used for initial calibration (Section 10.3), calibration verification (Sections 14.2 and 14.3), and for initial (Section 9.2.1) and ongoing (Section 14.5) precision and recovery may be prepared from the same source; however, the use of a secondary source for calibration verification is highly recommended whenever available. If standards from a different vendor are not available, a different lot number from the same vendor can be considered a secondary source. A LC-MS/MS instrument will provide the most reproducible results if dedicated to the settings and conditions required for determination of PFAS by this method.

9.7 Depending on specific program requirements, field replicates may be collected to determine the precision of the sampling technique, and spiked samples may be required to determine the accuracy of the analysis when the extracted internal standard method is used.

9.8 Matrix spikes generally are not required for isotope dilution methods because any deleterious effects of the matrix should be evident in the recoveries of the isotopically labeled compounds spiked into every sample. However, because some of the compounds are quantified by a non-analogous isotopically labeled compounds (e.g., PFPeS is quantified by $^{13}\text{C}_3\text{-PFHxS}$), the analysis of matrix spike samples may help diagnose matrix interferences for specific compounds.

10.0 Calibration and Standardization

10.1 Mass Calibration

The mass spectrometer must undergo mass calibration to ensure accurate assignments of m/z's by the instrument. This mass calibration must be performed at least annually to maintain instrument

sensitivity and stability. Mass calibration must be repeated on an as-needed basis (e.g., QC failures, ion masses fall outside of the instrument required mass window, major instrument maintenance, or if the instrument is moved). Mass calibration must be performed using the calibration compounds and procedures prescribed by the manufacturer. The procedures used for mass calibration and mass calibration verification must evaluate an ion range that encompasses the ion range (Q1 and Q2 m/z) of the analytes of interest of this method (Table 2).

Multiple Reaction Monitoring (MRM) analysis is required to achieve better sensitivity than full-scan analysis. The ions to be monitored (Q1 and Q2 m/z) for each native compound, isotopically labeled compound, and NIS are given in Table 2.

- 10.1.1** During the development of this method, instrumental parameters were optimized for the precursor and product ions listed on Table 2. Product ions other than those listed may be selected; however, the use of ions with lower mass or common ions that may not provide sufficient discrimination between analytes of interest and co-eluting interferences must be avoided.
- 10.1.2** Optimize the response of the precursor ion $[M-H]^-$ or $[M-CO_2]$ for each method analyte following the manufacturer's guidance. MS parameters (e.g., source voltages, source and desolvation temperatures, gas flow, etc.) must be methodically changed until optimal analyte responses are determined. Typically, carboxylic acids have similar MS/MS conditions and sulfonic acids have similar MS/MS conditions. However, since analytes may have different optimal parameters, some compromise on the final operating conditions may be required.
- 10.1.3** Establish suitable operating conditions using the manufacturer's instructions and use the table below for the MS conditions used during the development of this method as guidance.

Operating Conditions for Waters Acquity UPLC, TQ-S Xevo MS/MS

Injection volume	2.0 μ L (This is the default volume, and may be changed to improve performance)
	Source Temp ($^{\circ}$ C) 140
	Desolvation Temp ($^{\circ}$ C) 500
MS/MS Conditions	Capillary Voltage (kV) 0.70
	Cone Gas (L/h) $\sim$ 70
	Desolvation gas (L/h) $\sim$ 800

- 10.1.4** In the absence of manufacturer-specific instructions and acceptance criteria, the following procedure may be used for mass calibration.
 - 10.1.4.1** Introduce the NaCsI calibration solution (Section 7.4) to the MS at the flow rate necessary to produce a stable aerosol spray (e.g., 10 μ L/min).
 - 10.1.4.2** Scan the MS/MS over the mass range from 20 to 3000 atomic mass units (amu) (or Daltons [Da]). Adjust the source parameters to optimize peak intensity and shape across the mass range. The exact m/z's for NaCsI calibration are:

Calibration Masses (Daltons)

22.9898	922.3552	1971.6149
132.9054	1072.2494	2121.5091
172.8840	1222.1437	2271.4033
322.7782	1372.0379	2421.2976
472.6725	1521.9321	2571.1918
622.5667	1671.8264	2721.0861
772.4610	1821.7206	2870.9803

- 10.1.4.3** Mass calibration is judged on the basis of the presence or absence of the exact calibration masses (e.g., a limit of the number of masses that are “missed”). Absent vendor-specific instructions, all masses from 22.9898 to 1971.6149 must be present. If peaks in this range are missing or not correctly identified, adjust the MS/MS, and repeat the test. Only after the MS/MS is properly calibrated may standards, blanks, and samples be analyzed.
- 10.1.4.4** Mass spectrometer optimization – Prior to measurements of a given analyte the mass spectrometer must be separately optimized for that analyte.
- 10.1.4.5** Using the post-column pump, separately infuse a solution containing 2 - 5 µg/mL of each compound in methanol into the MS.
- 10.1.4.6** Optimize sensitivity to the product ion m/z for each compound. Precursor-product ion m/z's other than those listed may be used provided requirements in this method are met.
- 10.1.4.7** After MS calibration and optimization and LC-MS/MS calibration, the same LC-MS/MS conditions must be used for analysis of all standards, blanks, IPR and OPR standards, and samples.

10.1.5 Mass Calibration Verification

A mass calibration verification must be performed following mass calibration, prior to standards and samples analysis. Mass verification checks must also be performed after any subsequent mass calibrations. Each laboratory must follow the instructions for their individual instrument software to confirm the mass calibration, mass resolution and peak relative response. Mass calibration verification must be performed using standards whose mass range brackets the masses of interest (quantitative and qualitative ions).

- 10.1.5.1** Check the instrument mass resolution to ensure that it is at least unit resolution. Inject a mid-level CAL standard under LC-MS/MS conditions to obtain the retention times of each method analyte. Divide the chromatogram into retention time windows each of which contains one or more chromatographic peaks. During MS/MS analysis, fragment a small number of selected precursor ions ([M-H]⁺) for the analytes in each window and choose the most abundant product ion. The product ions (also the quantitation ions) chosen during method development are in Table 2, although these will be instrument dependent. Unit resolution is demonstrated when the value of the peak width at half-height is within 0.5 ± 0.1 amu or Da.

- 10.1.5.2** Check the mass calibration by measuring the amount of peak drift from the expected masses. If the peak apex has shifted more than approximately 0.1 Da, then the instrument will need to be recalibrated following the manufacturer's instructions.

10.2 Chromatographic conditions

- 10.2.1** The chromatographic conditions should be optimized for compound separation and sensitivity. The same optimized operating conditions must be used for the analysis of all standards, blanks, IPR and OPR standards, and samples. The following table gives the suggested chromatographic conditions for this method using the specified instrument and column. Different instruments may require slightly different operating conditions.
Modification of the solvent composition of the standard or extract by increasing the aqueous content to prevent poor peak shape is not permitted. The peak shape of early eluting compounds may be improved by increasing the volume of the injection loop or increasing the aqueous content of the initial mobile phase composition.

General LC Conditions			
Column Temp (°C)	40		
Max Pressure (bar)	1100.0		
LC Gradient Program			
Time (min)	Flow mixture ^{1,2}	Flow Rate Program	Gradient Curve
0.0	2% eluent A, 98% eluent B	0.35 mL/min	Initial
0.2	2% eluent A, 98% eluent B	0.35 mL/min	2
4.0	30% eluent A, 70% eluent B	0.40 mL/min	7
7	55% eluent A, 45% eluent B	0.40 mL/min	8
9	75% eluent A, 25% eluent B	0.40 mL/min	8
10	95% eluent A, 5% eluent B	0.40 mL/min	6
10.4	2% eluent A, 98% eluent B	0.40 mL/min	10
11.8	2% eluent A, 98% eluent B	0.40 mL/min	7
12.0	2% eluent A, 98% eluent B	0.35 mL/min	1

¹ Eluent A = Acetonitrile

² Eluent B = 2 mM ammonium acetate in 95:5 water/acetonitrile

Note: LC system components, as well as the mobile phase constituents, may contain many of the analytes in this method. Thus, these PFAS will build up on the head of the LC column during mobile phase equilibration. To minimize the background PFAS peaks and to keep baseline levels constant, the time the LC column sits at initial conditions must be kept constant and as short as possible (while ensuring reproducible retention times). In addition, priming the mobile phase and flushing the column with at least 90% methanol before initiating a sequence may reduce background contamination.

10.2.2 Retention time calibration

- 10.2.2.1** Inject compound solution(s) to determine its retention time. The laboratory may want to inject compounds separately the first time they perform the calibration. All native compounds for which there is an isotopically labeled analog will elute slightly before or with the labeled analog. Store the retention time (RT) for each compound in the data system.

- 10.2.2.2** Once RT windows have been confirmed for each analyte, once per ICAL and at the beginning of the analytical sequence, the position of each method analyte, EIS analyte, and NIS analyte peaks shall be set using the midpoint standard of the ICAL curve when ICAL is performed. When ICAL is not performed, the initial CV retention times or the midpoint standard of the ICAL curve can be used to establish the RT window position.
- 10.2.2.3** Method analyte, EIS analyte, and NIS analyte RTs must fall within 0.4 minutes of the predicted retention times from the midpoint standard of the ICAL or initial daily CV, whichever was used to establish the RT window position for the analytical batch. All branched isomer peaks identified in either the calibration standard or the qualitative (technical grade) standard must fall within in the retention time window for that analyte.
- 10.2.2.4** For all method analytes with exact corresponding isotopically labeled analogs, method analytes must elute within 0.1 minutes of the associated EIS.
- 10.2.2.5** When establishing the chromatographic conditions, it is important to consider the potential interference of bile salts during analyses of tissue samples. Inject a standard containing TDCA (Section 7.5) during the retention time calibration process and adjust the conditions to ensure that TDCA does not coelute with any of the target analytes, EIS, or NIS standards. Analytical conditions must be set to allow a separation of at least 1 minute between the bile salts and PFOS.

10.3 Initial calibration

Initial calibration is performed using a series of at least six solutions, with at least five of the six calibration standards being within the quantification. (If a second-order calibration model is used, then one additional concentration is required.) The initial calibration solutions contain the entire suite of isotopically labeled compounds, NISs, and target compounds. Calibration is verified with a calibration verification (CV) standard at least once every ten field samples or less, by analysis of a mid-level calibration solution. Calibration verification uses the mean RRs or RFs determined from the initial calibration to calculate the analyte concentrations in the verification standard.

Note: Six calibration standards is the minimum number that must be used in the initial calibration; however, the laboratory may use more standards, as long as the criteria in Section 10.3.3.3 can be met.

Prior to the analysis of samples, and after the mass calibration check has met all criteria in Section 10.1.4, each LC-MS/MS system must be calibrated at a minimum of 6 standard concentrations (Section 7.3.4 and Table 4). This method procedure calibrates and quantifies 40 PFAS target analytes, using the isotopically labeled compounds added to the sample prior to extraction, by one of two approaches:

- True isotope dilution quantification (ID), whereby the response of the target compound is compared to the response of its isotopically labeled analog. Twenty-four target compounds are quantified in this way.
- Extracted internal standard quantification (EIS), whereby the response of the target compound is compared to the response of the isotopically labeled analog of another compound with chemical and retention time similarities. Sixteen target compounds are quantified in this way.

10.3.1 Initial calibration frequency

Each LC-MS/MS system must be calibrated whenever the laboratory takes corrective action that might change or affect the initial calibration criteria, or if either the CV or Instrument Sensitivity Check (ISC) acceptance criteria have not been met.

10.3.2 Initial calibration procedure

Prepare calibration standards containing the native compounds, EISs, and NISs, at the concentrations described in Table 4. Analyze each calibration standard by injecting 2.0 µL (this volume may be changed to improve performance).

Note: The same injection volume must be used for all standards, samples, blanks, and QC samples.

10.3.3 Initial calibration calculations

10.3.3.1 Instrument sensitivity

Sufficient instrument sensitivity is established if a signal-to-noise ratio $\geq 3:1$ can be achieved when analyzing the lowest concentration standard within the quantitation range that the laboratory includes in its assessment of calibration linearity (Table 4).

10.3.3.2 Response Ratios (RR) and Response Factors (RF)

The response ratio (RR) for each compound calibrated by isotope dilution is calculated according to the equation below, separately for each of the calibration standards, using the areas of the quantitation ions (Q1) with the m/z shown in Table 2. RR is used for the 24 compounds quantified by true isotope dilution.

$$RR = \frac{Area_n M_l}{Area_l M_n}$$

where:

- Area_n = The measured area of the Q1 m/z for the native (unlabeled) PFAS
- Area_l = The measured area at the Q1 m/z for the corresponding isotopically labeled PFAS added to the sample before extraction
- M_l = The mass of the isotopically labeled compound in the calibration standard
- M_n = The mass of the native compound in the calibration standard

Similarly, the response factor (RF) for each unlabeled compound calibrated by extracted internal standard is calculated according to the equation below. RF is used for the 16 compounds quantified by extracted internal standard.

$$RF = \frac{Area_s M_{EIS}}{Area_{EIS} M_s}$$

where:

- Area_s = The measured area of the Q1 m/z for the target (unlabeled) PFAS
- Area_{EIS} = The measured area at the Q1 m/z for the isotopically labeled PFAS used as the extracted internal standard (EIS)

- M_{EIS} = The mass of the isotopically labeled PFAS used as the extracted internal standard (EIS) in the calibration standard
- M_s = The mass of the target (unlabeled) PFAS in the calibration standard

A response factor (RF_s) is calculated for each isotopically labeled compound in the calibration standard using the equation below. RF_s is used for the 24 isotopically labeled compounds quantified by non-extracted internal standard.

$$RF_s = \frac{Area_l M_{NIS}}{Area_{NIS} M_l}$$

where:

- $Area_l$ = The measured area of the Q1 m/z for the isotopically labeled PFAS standard added to the sample before extraction
- $Area_{NIS}$ = The measured area at the Q1 m/z for the isotopically labeled PFAS used as the non-extracted internal standard (NIS)
- M_{NIS} = The mass of the isotopically labeled compound used as the non-extracted internal standard (NIS) in the calibration standard
- M_l = The mass of the isotopically labeled PFAS standard added to the sample before extraction

Note: Other calculation approaches may be used, such as linear regression or non-linear regression based on the capability of the data system used by the laboratory.

10.3.3.3 Instrument Linearity

One of the following two approaches must be used to evaluate the linearity of the instrument calibration:

Option 1: Calculate the relative standard deviation (RSD) of the RR or RF values of the six initial calibration standards for each native compound and isotopically labeled compound. The RSD must be $\leq 20\%$ to establish instrument linearity.

Option 2: Calculate the relative standard error (RSE) of the six initial calibration standards for each native compound and isotopically labeled compound. The RSE for all method analytes must be $\leq 20\%$ to establish instrument linearity.

10.3.4 Initial calibration corrective actions

If the instrument sensitivity or the instrument linearity criteria for initial calibration are not met, inspect the system for problems and take corrective actions to achieve the criteria. This may require the preparation and analysis of fresh calibration standards. All initial calibration criteria must be met before any samples or required blanks are analyzed.

10.3.5 Bile salts interference check

The laboratory must analyze a TDCA standard after the initial calibration, prior to the analysis of tissue samples, to check for interferences caused by bile salts. If an interference is present, the chromatographic conditions must be modified to eliminate the interference from TDCA (e.g., changing the retention time of TDCA such that it falls outside the

retention window for PFOS by at least one minute), and the initial calibration repeated. If tissue sample analyses are not being conducted, this check may be skipped.

11.0 Sample preparation and extraction

For aqueous samples that contain particles and solid samples, percent solids are determined using the procedures in Section 11.1. This section describes the sample preparation procedures for aqueous samples with < 50 mg solids (Section 11.2), solid (soil, sediment or biosolid) samples (Section 11.3) and tissue samples (Section 11.4).

Note: It is highly recommended that the laboratory pre-screens all samples prior to performing the analysis (see Appendix A). For aqueous samples, use the secondary container provided for percent solids to perform the pre-screening. If high levels of PFAS are present in the sample, a lower volume is required for analysis.

The laboratory may subsample the aqueous samples as described in Appendix B; however, subsampling must meet project-specific requirements. The laboratory must notify the client before proceeding with subsampling. Once the laboratory becomes familiar with the levels of PFAS in the samples for their clients, the samples should be collected in the appropriate sample container size to avoid subsampling. The sample data report must state when subsampling has been employed.

Do not use any fluoropolymer articles or task wipes in these extraction procedures. Use only HDPE or polypropylene wash bottles and centrifuge tubes. Reagents and solvents for cleaning syringes may be kept in glass containers.

11.1 Determination of Percent Solids

11.1.1 Determination of percent suspended solids – Aqueous liquids and multi-phase samples consisting of mainly an aqueous phase

11.1.1.1 Desiccate and weigh a glass fiber filter (Section 6.4.3) to three significant figures.

11.1.1.2 Filter 10.0 ± 0.02 mL of well-mixed sample through the filter.

11.1.1.3 Dry the filter a minimum of 12 hours at 110 ± 5 °C and cool in a desiccator.

11.1.1.4 Calculate percent solids as follows:

$$\% \text{ solids} = \frac{\text{weight of sample aliquot after drying (g)} - \text{weight of filter (g)}}{10 \text{ g}} \times 100$$

11.1.2 Solids (excluding tissues)

11.1.2.1 Weigh 5 to 10 g of sample to three significant figures in a tared beaker.

11.1.2.2 Dry a minimum of 12 hours at 110 ± 5 °C, and cool in a desiccator.

11.1.2.3 Calculate percent solids as follows:

$$\% \text{ solids} = \frac{\text{weight of sample aliquot after drying (g)}}{\text{weight of sample aliquot before drying (g)}} \times 100$$

11.2 Aqueous Sample Processing

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. Smaller sample volumes may be analyzed for samples containing solids greater than specified for this method, or when unavoidable due to high level of PFAS; however, subsampling should be avoided whenever possible. Typical sample size is 500 mL; however, sample size may be up to 1000 mL. The sample is to be analyzed in its entirety and should not be filtered. Leachate samples are analyzed using a 100-mL sample volume. Therefore, they must not be included in the same sample preparation batch as aqueous samples analyzed which are analyzed using 500-mL sample volumes.

- 11.2.1** Homogenize the sample by inverting the sample 3 – 4 times and allowing the sample to settle. Do not filter the sample. The standard procedure is to analyze the entire sample, plus a basic methanol rinse of the container.
- 11.2.2** The volume of the aqueous sample analyzed is determined by weighing the full sample bottle and then the empty sample bottle (see Section 12.2). Weigh each sample bottle (with the lid) to 0.1 g.
- 11.2.3** Prepare a method blank and two OPRs using PFAS-free water in HDPE bottles. Select a volume of water that is typical of the samples in the batch. Spike one OPR sample with native standard solution (Section 7.3.3) at 2x the LOQ (LLOPR). This aliquot will serve to verify the LOQ. Spike the other OPR sample at the concentration of the mid-level calibration point. This aliquot will serve as the traditional OPR.

Note: If matrix spikes are required for a specific project, spike the field sample bottles designated for use as MS/MSD samples with native standard solution (Section 7.3.3) at a concentration 3 to 5 times the background concentration determined during screening of the unspiked sample. If screening was not performed, then spike those samples at the concentration of the mid-level calibration point.

- 11.2.4** Spike an aliquot of EIS solution (Section 7.3.1) directly into the sample in the original bottle (or subsampled bottle) as well as to the bottles prepared for the QC samples. Mix by swirling the sample container.
- 11.2.5** Check that the pH is 6.5 ± 0.5 . If necessary, adjust pH with 50% formic acid (Section 7.1.13.4) or ammonium hydroxide (or with 5% formic acid [Section 7.1.13.3] and 3% aqueous ammonium hydroxide [Section 7.1.6.2]). The extract is now ready for solid-phase extraction (SPE) and cleanup (Section 12.0).

11.3 Solid Sample (excluding tissues) Processing

Use a stainless spoon to mix the sample in its original jar. If it is impractical to mix the sample within its container transfer the sample to a larger container. Remove rocks, invertebrates, and foreign objects. Vegetation can either be removed from the sample before homogenization or cut into small pieces and included in the sample, based on project requirements. Mix the sample thoroughly, stirring from the bottom to the top and in a circular motion along the sides of the jar, breaking particles to less than 1 mm by pressing against the side of the container. The homogenized sample should be even in colour and have no separate layers. Store the homogenized material in its

original container or in multiple smaller containers. Determine the percent solids as per Section 11.1.2.

Note: The maximum sample weight for sediment or soil is 5 g dry weight. The maximum sample weight for biosolids is 0.5 g dry weight.

Small amounts of reagent free water used for method blanks (10% of sample weight or less) can be added to unusually dry samples. This is an option, not a requirement.

- 11.3.1** Weigh out an aliquot of solid sample, not dried (aliquot should provide 5 g dry weight for soil and sediment or 0.5 g dry-weight for biosolids) into a 50-mL polypropylene centrifuge tube. Because biosolids samples are analyzed with a 0.5-g sample, they must not be included in the same sample preparation batch as solid samples analyzed with 5-g sample masses.
- 11.3.2** Prepare batch QC samples using 5 g of reference solid (Section 7.2.2) wetted with 2.5 g of reagent water for the method blank and two OPRs (use 0.5 g of reference solid with 0.25 g of reagent water for biosolid sample batches). The addition of reagent water to the sand provides a matrix closer in composition to real-world samples. Spike one OPR sample with native standard solution (Section 7.3.3) at 2x the LOQ (LLOPR). This aliquot will serve to verify the LOQ. Spike the other OPR sample at the concentration of the mid-level calibration point. This aliquot will serve as the traditional OPR.
-

Note: If matrix spikes are required for a specific project, spike the field sample aliquots designated for MS/MSD samples with native standard solution (Section 7.3.3) at the concentration 3 to 5 times the background concentration determined during screening of the unspiked sample. If screening was not performed, then spike those samples at the concentration of the mid-level calibration point.

- 11.3.3** Spike an aliquot of EIS solution (Section 7.3.1) directly into each centrifuge tube containing the aliquoted field and QC samples. Vortex the sample to disperse the standard and allow to equilibrate for at least 30 minutes.
- 11.3.4** Add 10 mL of 0.3% methanolic ammonium hydroxide (Section 7.1.7.1) to each centrifuge tube. Vortex to disperse, then shake for 30 minutes on a variable speed mixing table. Centrifuge at 2800 rpm for 10 minutes and transfer the supernatant to a clean 50-mL polypropylene centrifuge tube.
- 11.3.5** Add 15 mL of 0.3% methanolic ammonium hydroxide (Section 7.1.7.1) to the remaining solid sample in each centrifuge tube. Vortex to disperse, then shake for 30 minutes on a variable speed mixing table. Centrifuge at 2800 rpm for 10 minutes and decant the supernatant from the second extraction into the centrifuge tube with the supernatant from the first extraction.
- 11.3.6** Add another 5 mL of 0.3% methanolic ammonium hydroxide (Section 7.1.7.1) to the remaining sample in each centrifuge tube. Shake by hand to disperse, centrifuge at 2800 rpm for 10 minutes and decant the supernatant from the third extraction into the centrifuge tube with supernatant from the first and second extractions.
- 11.3.7** Using a 10-mg scoop, add 10 mg of carbon (Section 7.1.17) to the combined extract, mix by occasional hand shaking for no more than five minutes and then centrifuge at 2800 rpm

for 10 minutes. Immediately decant the extract into a 60-mL glass evaporation or concentrator tube.

- 11.3.8** Dilute to approximately 35 mL with reagent water. A separate concentrator tube marked at the 35-mL level may be kept for a visual reference to get the approximate volume. Samples containing more than 50% water may yield extracts that are greater than 35 mL in volume; therefore, do not add water to these. Determine the water content in the sample as follows (percent moisture is determined from the % solids):

$$\text{Water Content in Sample} = \frac{\text{Sample Weight (g)} \times \text{Moisture (\%)}}{100}$$

- 11.3.9** Concentrate each extract at approximately 55 °C with a N₂ flow of approximately 1.2 L/min to a final volume that is based on the water content of the sample (*see table below*). Allow extracts to concentrate for 25 minutes, then mix (by vortex if the volume is < 20 mL or using a glass pipette if the volume is > 20 mL). Continue concentrating and mixing every 10 minutes until the extract has been reduced to the required volume as specified in the table below. If the extract volume appears to stop dropping, the concentration must be stopped and the volume at which it was stopped recorded.

Water Content in Sample	Concentrated Final Volume
< 5 g	7 mL
5 – 8 g	8 mL
8 – 9 g	9 mL
9 – 10 g	10 mL

Note: Slowly concentrating extracts, in 1-mL increments, is necessary to prevent excessive concentration and the loss of neutral compounds (methyl and ethyl FOSEs and FOSAs) and other highly volatile compounds. The extract must be concentrated to remove the methanol as excess methanol during SPE clean-up results in poor recovery of C13 and C14 carboxylic acids and C10 and C12 sulfonates.

- 11.3.10** Add 40 - 50 mL of reagent water to the extract and vortex. Check that the pH is 6.5 ±0.5 and adjust as necessary with 50% formic acid (Section 7.1.13.4) or 30% ammonium hydroxide (or with 5% formic acid [Section 7.1.13.3] and 3% aqueous ammonium hydroxide [Section 7.1.6.2]). The extracts are ready for SPE and cleanup (Section 12.0).

11.4. Tissue Sample Processing

Prior to processing tissue samples, the laboratory must determine the exact tissue to be analyzed. Common requests for analysis of fish tissue include whole fish with the skin on, whole fish with the skin removed, edible fish fillets (filleted in the field or by the laboratory), specific organs, and other portions. Once the appropriate tissue has been determined, the samples must be prepared and homogenized.

If the laboratory must dissect the whole fish to obtain the appropriate tissue for analysis, cover the benchtop with clean aluminum foil and use clean processing equipment (knives, scalpels, tweezers) to dissect each sample to prevent cross-contamination. Samples should be handled in a semi-thawed state for compositing and/or homogenization. All tissue comprising a sample is collected in a stainless-steel bowl during grinding, then mixed using a stainless-steel spoon. Homogenized samples must be stored in clean HDPE containers and stored frozen for subsequent use.

If using a grinder, after the entire sample has been processed, mix the ground tissue with a spoon, transfer back to the grinder, and repeat the grinding at least two more times until the homogenize tissue has a consistent texture and color.

- 11.4.1** For each sample, weigh a 2-g aliquot of homogenized tissue into a 15-mL polypropylene centrifuge tube. Reseal the container with the remaining homogenized portion of the sample and return it to frozen storage in the event that it needs to be used for reanalysis.

Note: The default sample weight for tissue is 2 g wet weight; however, a 1-g sample may be used. Higher sample weights are not recommended for this method.

- 11.4.2** Prepare the batch QC samples using 2 g of reference tissue matrix (Section 7.2.3) for the method blank and two OPRs. Spike one OPR sample with native standard solution (Section 7.3.3) at 2x the LOQ (LLOPR). This aliquot will serve to verify the LOQ. Spike the other OPR sample at the concentration of the mid-level calibration point. This aliquot will serve as the traditional OPR.

Note: If matrix spikes are required for a specific project, spike the field sample aliquots designated as MS/MSD samples with native standard solution (Section 7.3.3) at the concentration 3 to 5 times the background concentration determined during screening of the unspiked sample. If screening was not performed, then spike those samples at the concentration of the mid-level calibration point.

- 11.4.3** Spike an aliquot of EIS solution (Section 7.3.1) directly into each field and QC sample. Vortex and allow to equilibrate for at least 30 minutes.
- 11.4.4** Add 10 mL of 0.05M KOH in methanol (Section 7.1.8) to each sample. Vortex to disperse the tissue then place tubes on a variable speed mixing table to extract for at least 16 hours. Centrifuge at 2800 rpm for 10 minutes and collect the supernatant in a 50-mL polypropylene centrifuge tube.
- 11.4.5** Add 10 mL of acetonitrile to remaining tissue in the 15-mL centrifuge tube, vortex to mix and disperse the tissue. Sonicate for 30 minutes. Centrifuge at 2800 rpm for 10 minutes and collect the supernatant, adding it to the 50-mL centrifuge tube containing the initial extract.
- 11.4.6** Add 5 mL of 0.05M KOH in methanol (Section 7.1.8) to the remaining sample in each centrifuge tube. Vortex to disperse the tissue and hand mix briefly. Centrifuge at 2800 rpm for 10 minutes and collect the supernatant, adding it to the 50-mL centrifuge tube containing the first two extracts.
- 11.4.7** Using a 10-mg scoop, add 10 mg of carbon (Section 7.1.17) to the combined extract, mix by occasional hand shaking over a period of no more than five minutes and then centrifuge at 2800 rpm for 10 minutes. Immediately decant the extract into a 60-mL glass evaporation or concentrator tube.
- 11.4.8** Add 1 mL of reagent water to each evaporation/concentrator tube, set the evaporator/concentrator to 55 °C with a N₂ flow of 1.2 L/min and concentrate the extract to 2.5 mL (only ~1 mL of the methanol should remain).
- 11.4.9** Add reagent water to each evaporation/concentrator tube to dilute the extracts to 50 mL. Check that the pH = 6.5 ± 0.5 and adjust as needed with 50% formic acid (Section 7.1.13.4)

or ammonium hydroxide (or with 5% formic acid [Section 7.1.13.3] and 3% aqueous ammonium hydroxide [7.1.6.2]). The extracts are ready for SPE and cleanup (Section 12.0).

12.0 Extraction, Cleanup, and Concentration

All matrices (including batch QC) must undergo SPE and carbon cleanup to remove interferences (Section 12.1). Sample elution as well as any further extract treatment is matrix specific and may be found in Sections 12.2 through 12.4.

Note: Carbon cleanup is required. Carbon cleanup may remove analytes if the sample has a very low organic carbon content (this is unusual for non-drinking water environmental samples). This will be apparent if the isotope dilution standard recoveries are significantly higher on the reanalysis. If the laboratory can demonstrate that the carbon cleanup is detrimental to the sample analysis (by comparing results when skipping the carbon cleanup during reanalysis), then the carbon cleanup may be skipped for that specific sample.

12.1 All sample matrices

- 12.1.1 Pack clean silanized glass wool to half the height of the WAX SPE cartridge barrel (Section 6.7.1).
- 12.1.2 Set up the vacuum manifold with one WAX SPE cartridge plus a reservoir and reservoir adaptor for each cartridge for each sample and QC aliquot.
- 12.1.3 Pre-condition the cartridges by washing them with 15 mL of 1% methanolic ammonium hydroxide (Section 7.1.7.2) followed by 5 mL of 0.3M formic acid (Section 7.1.13.2) (do not use the vacuum for this step). Do not allow the WAX SPE to go dry. Discard the wash solvents.
- 12.1.4 Pour the sample into the reservoir (do not use a pipette), taking care to avoid splashing while loading. Adjust the vacuum and pass the sample through the cartridge at 5 mL/min. Retain the empty sample bottle and allow it to air dry for later rinsing (Section 12.2.2). Discard eluate.

Note: For aqueous samples, in the event the SPE cartridge clogs during sample loading, place a second pre-conditioned cartridge and continue loading the remaining sample aliquot using the same reservoir. Proceed to Section 12.1.5.

- 12.1.5 Rinse the walls of the reservoir with 5 mL reagent water (twice) followed by 5 mL of 1:1 0.1M formic acid/methanol (Section 7.1.13.5) and pass those rinses through the cartridge using vacuum. Dry the cartridge by pulling air through for 15 seconds. Discard the rinse solution. Continue to the elution and concentration steps based on the matrix (Section 12.2 – Aqueous, Section 12.3 – Solids and Section 12.4 – Tissue).

12.2 Elution and Extract Concentration of Aqueous Samples

Note: If two cartridges were used, perform Sections 12.2.1 through 12.2.3 with each cartridge. Filter the eluates through a 25-mm, 0.2- μ m syringe filter. Combine both sets of filtered eluates into a clean tube, add the NIS solution, and vortex to mix. Transfer 350 μ L of the filtered extract into a 1-mL polypropylene microvial and mark the level. Add another

350- μ L portion and using a gentle stream of nitrogen (water bath at 40 °C), concentrate to the 350- μ L mark and submit for LC-MS/MS analysis. This concentration step is only applicable to situations where two SPE cartridges were eluted, each with 5 mL of elution solvent.

- 12.2.1** Place clean collection tubes (13 x 100 mm polypropylene) inside the manifold, ensuring that the extract delivery needles do not touch the walls of the tubes. DO NOT add NIS to these collection tubes.
 - 12.2.2** Rinse the inside of the sample bottle with 5 mL of 1% methanolic ammonium hydroxide (Section 7.1.7.2), then, using a glass pipette, transfer the rinse to the SPE reservoir, washing the walls of the reservoir. Use vacuum to pull the elution solvent through the cartridge and into the collection tubes.
-

Note: Air dry the empty sample bottle after the rinse is transferred. Weigh the empty bottle with the cap on and subtract from the weight with the sample determined in Section 11.2.2.

- 12.2.3** Add 25 μ L of concentrated acetic acid to each sample eluted in the collection tubes and vortex to mix. Add 10 mg of carbon (Section 7.1.17) to each sample and batch QC extract, using a 10-mg scoop. Hand-shake occasionally for no more than 5 minutes. It is important to minimize the time the sample extract is in contact with the carbon. Immediately vortex (30 seconds) and centrifuge at 2800 rpm for 10 minutes.
- 12.2.4** Add NIS solution (Section 7.3.2) to a clean collection tube. Place a syringe filter (25-mm filter, 0.2- μ m nylon membrane) on a 5-mL polypropylene syringe. Take the plunger out and carefully decant the sample supernatant into the syringe barrel. Replace the plunger and filter the entire extract into the new collection tube containing the NIS. Vortex to mix and transfer a portion of the extract into a 1-mL polypropylene microvial for LC-MS/MS analysis. Cap the collection tube containing the remaining extract and store at 0 - 4 °C.

12.3 Elution and Extract Concentration of Solid Samples

- 12.3.1** Add NIS solution (Section 7.3.2) to a clean collection tube (13 x 100 mm polypropylene) for each sample and QC aliquot and place them into the manifold rack, ensuring the extract delivery needles are not touching the walls of the tubes.
- 12.3.2** Rinse the inside of the evaporation/concentrator tube using 5 mL of 1% methanolic ammonium hydroxide (Section 7.1.7.2), then, using a glass pipette, transfer the rinse to the reservoir, washing the walls of the reservoir. Use the vacuum to pull the elution solvent through the cartridge and into the collection tubes.
- 12.3.3** Add 25 μ L of concentrated acetic acid to each sample extract in its collection tube and swirl to mix. Place a syringe filter (25-mm filter, 0.2- μ m nylon membrane) on a 5 mL polypropylene syringe. Take the plunger out and carefully decant ~1 mL of sample extract into the syringe barrel. Replace the plunger and filter into a 1-mL polypropylene microvial for LC-MS/MS analysis. Cap the collection tube containing the remaining extract and store at 0 - 4 °C.

12.4 Elution and Extract Concentration of Tissue Samples

- 12.4.1 Add NIS solution (Section 7.3.2) to clean collection tubes (13 x 100 mm, polypropylene) for each sample and QC aliquot. Place the tubes into the manifold rack and ensure the extract delivery needles are not touching the walls of the tubes.
- 12.4.2 Rinse the inside of the evaporation/concentrator tube using 5 mL of 1% methanolic ammonium hydroxide (Section 7.1.7.2), then, using a glass pipette, transfer the rinse to the reservoir, washing the walls of the reservoir. Use the vacuum to pull the elution solvent through the cartridge and into the collection tubes.
- 12.4.3 Add 25 µL of concentrated acetic acid to each sample extract. Place a syringe filter (25-mm filter, 0.2-µm nylon membrane) on a 5 mL polypropylene syringe. Take the plunger out and carefully decant an aliquot (~1 mL) of the sample extract into the syringe barrel. Replace the plunger and filter into a 1-mL polypropylene microvial for LC-MS/MS analysis. Cap the collection tube containing the remaining extract and store at 0 - 4 °C.

13.0 Instrumental Analysis

Analysis of sample extracts for PFAS by LC-MS/MS is performed on an ultrahigh performance liquid chromatograph coupled to a triple quadrupole mass spectrometer, running manufacturer's software. The mass spectrometer is run with unit mass resolution in the multiple reaction monitoring (MRM) mode.

- 13.1 Perform mass calibration (Section 10.1), establish the operating conditions (Section 10.2), and perform an initial calibration (Section 10.3) prior to analyzing samples. If tissue samples are to be analyzed during the analytical shift, repeat the TDCA interference check in Section 10.3.5 before analyzing any field samples.
- 13.2 Only after all performance criteria are met may blanks, MDLs, IPRs/OPRs, and samples be analyzed.
- 13.3 After a successful initial calibration has been completed, the analytical sequence for a batch of samples analyzed during the same time period is as follows. The volume injected for samples and QCs must be identical to the volume used for calibration (Section 10.3). Standards and sample extracts must be brought to room temperature and vortexed prior to aliquoting into an instrument vial in order to ensure homogeneity of the extract.
 1. Instrument Blank
 2. Instrument Sensitivity Check (see Section 10.3.3.1)
 3. Calibration Verification Standard
 4. Qualitative Identification Standards
 5. Instrument Blank
 6. Method Blank
 7. Low-level OPR (LLOPR)
 8. OPR
 9. TDCA standard (only if tissue samples are being analyzed)
 10. Samples (10 or fewer)
 11. Calibration Verification Standard
 12. Instrument Blank
 13. Samples (10 or fewer)
 14. Calibration Verification Standard
 15. Instrument Blank

If the results are acceptable, the closing calibration verification solution (#14 above) may be used as the opening solution for the next analytical sequence.

- 13.4** If the response exceeds the calibration range for any sample, extracts are diluted as per Section 15.3 to bring all target responses within the calibration range.

Note: If the analytes that exceed the calibration range in the original analysis are known to not be of concern for the specific project (e.g., are not listed in a discharge permit), then the laboratory may consult with the client regarding the possibility of reporting that sample from the undiluted analysis.

14.0 Performance Tests during Routine Operations

The following performance tests must be successfully completed as part of each routine instrumental analysis shift described in Section 13.3 above.

- 14.1** MS resolution – A mass calibration must be performed prior to analysis of the calibration curve. LC-MS/MS system performance is checked by performing an MS resolution verification after the mass calibration. MS resolution must be verified prior to any samples or QC as per Section 10.1. If the requirements in Section 10.1 cannot be met, the problem must be corrected before analyses can proceed. If any of the samples in the previous shift may be affected by poor mass resolution, the extracts of those samples must be re-analyzed.

14.2 Instrument sensitivity check

The signal-to-noise ratio of the ISC standard (Section 7.3.4) must be greater than or equal to 3:1. If the requirements cannot be met, the problem must be corrected before analyses can proceed.

Note: An interim limit of 70-130% for 90% of the native and isotopically labeled compounds should be used, with the other recoveries achieving 50-150%.

14.3 Calibration verification (CV)

After a passing MS resolution (Section 14.1) and a successful initial calibration (Section 10.3.3.3) is achieved, prior to the analysis of any samples, analyze a mid-level calibration standard (Section 7.3.4).

- 14.3.1** The calibration is verified by analyzing a CV standard at the beginning of each analytical sequence, every ten samples or less, and at the end of the analytical sequence.
- 14.3.2** Calculate concentration for each native and isotopically labeled compound in the CV using the equation in Section 15.2.
- 14.3.3** The recovery of native and isotopically labeled compounds for the CVs must be within 70 - 130%.
- 14.3.4** If the CV criterion in Section 14.3.3 is not met, recalibrate the LC-MS/MS instrument according to Section 10.3.

14.4 Retention times and resolution

- 14.4.1** For all method analytes with exact corresponding isotopically labeled analogs, method analytes must elute within ± 0.1 minutes of the associated EIS.
- 14.4.2** The retention times of each native and isotopically labeled compound must be within ± 0.4 minutes of the ICAL or CV used to establish the RT windows for the samples and batch QC.

14.5 Ongoing precision and recovery (OPR)

- 14.5.1** After verification, analyze the extract of the OPR (Sections 12.2.4, 12.3.3, and 12.4.3) prior to analysis of samples from the same batch to ensure the analytical process is under control.
- 14.5.2** Compute the percent recovery of the native compounds by the appropriate quantification method depending on the compound (Section 10.3). Compute the percent recovery of each isotopically labeled compound by the non-extracted internal standard method (Sections 1.2 and 10.3).

$$\text{Recovery (\%)} = \frac{\text{Concentration found (ng/mL)}}{\text{Concentration spiked (ng/mL)}} \times 100$$

- 14.5.3** For the native compounds and isotopically labeled compounds, compare the recovery to the OPR limits given in Table 5. If all compounds meet the acceptance criteria, system performance is acceptable, and analysis of blanks and samples may proceed. If, however, any individual concentration falls outside of the given range, the extraction/concentration processes are not being performed properly for that compound. In this event, correct the problem, re-prepare, extract, and clean up the sample batch and repeat the ongoing precision and recovery test.
- 14.5.4** If desired, add results that pass the specifications in Section 14.5.3 to initial and previous ongoing data for each compound in each matrix. Update QC charts to form a graphic representation of continued laboratory performance. Develop a statement of laboratory accuracy for each compound in each matrix type by calculating the average percent recovery (R) and the standard deviation of percent recovery (SR). Express the accuracy as a recovery interval from $R - 2SR$ to $R + 2SR$. For example, if $R = 95\%$ and $SR = 5\%$, the accuracy is 85 to 105%.
- 14.6** Instrument blank – At the beginning of the analytical sequence and after the analysis of high concentration samples (e.g., highest calibration standard, CV), analyze an instrument blank to ensure no instrument contamination has occurred.
- 14.7** Method blank – After the analysis of the solvent blank and prior to the analysis of samples, analyze a method blank (Section 9.5).
- 14.8** A qualitative identification standard (Section 7.3.5) containing all available isomers (branched and linear) is analyzed once daily at the beginning of the analytical sequence, to confirm the retention time of each linear and known branched isomer or isomer group.

14.9 Instrument sensitivity (optional)

This step is recommended as a follow-up step if the ISC does not meet criteria.

Compare the NIS peak areas from the QC and field samples to the average area of the corresponding NIS on the calibration standards to check for possible bad injections of NIS solution or loss of instrument sensitivity. The QC and field sample NIS areas should be within 50 – 200% of that in the standards. If the areas are low for all the samples and QC in the batch, it suggests a loss of instrument sensitivity, while low areas on only some QC or field samples suggests a possible bad injection.

15.0 Data Analysis and Calculations

15.1 Qualitative determination and peak identification

A native or isotopically labeled compound is identified in a standard, blank, sample, or QC sample when all of the criteria in Sections 15.1.1 through 15.1.5 are met.

15.1.1 Peak responses must be at least three times the background noise level (S/N 3:1). If the S/N ratio is not met due to high background noise, the laboratory must correct the issue (e.g., perform instrument troubleshooting to check and if needed, replace, the transfer line, column, detector, liner, filament, etc.). If the S/N ratio is not met but the background is low, then the analyte is to be considered a non-detect.

15.1.2 Target analyte, EIS analyte, and NIS analyte RTs must fall within ± 0.4 minutes of the predicted retention times from the midpoint standard of the ICAL or initial daily CV, whichever was used to establish the RT window position for the analytical batch. The retention time window used must be of sufficient width to detect earlier-eluting branched isomers. For all method analytes with exact corresponding isotopically labeled analogs, method analytes must elute within ± 0.1 minutes of the associated EIS.

15.1.3 The laboratory must follow the identification requirements specified by the client for the project. In the event there are no project-specific requirements, the following general requirements apply. For concentrations at or above the method LOQ, the total (branched and linear isomer) quantification ion response to the total (branched and linear isomer) confirmation ion response ratio must fall within $\pm 50\%$ of the ratio observed in the midpoint initial calibration standard. If project-specific requirements involve reporting sample concentrations below the LOQ or ML, the response ratio must also fall within $\pm 50\%$ of the ratio observed in the initial daily CV.

The response of all isomers in the quantitative standards should be used to define ratio. In samples, the total response should include only the branched isomer peaks that have been identified in either the quantitative or qualitative standard (see Section 7.3 regarding records of traceability of all standards). If standards (either quantitative or qualitative) are not available for purchase, only the linear isomer can be identified and quantitated in samples. The ratio requirement does not apply for PFBA, PFPeA, NMeFOSE, NEtFOSE, PFMPA, and PFMBa because suitable (not detectable or inadequate S/N) secondary transitions are unavailable.

15.1.4 If the field sample result does not all meet the criteria stated in Sections 15.1.2 through 15.1.3, and all sample preparation avenues (e.g., extract cleanup, sample dilution, etc.) have been exhausted, the result may only be reported with a data qualifier alerting the data user

that the result could not be confirmed because it did not meet the method-required criteria and therefore should be considered an estimated value. If the criteria listed above are not met for the standards, the laboratory must stop analysis of samples and correct the issue.

15.2 Quantitative determination

Concentrations of the target analytes are determined with respect to the extracted internal standard (EIS) which is added to the sample prior to extraction. The EIS is quantitated with respect to a non-extracted internal standard (NIS), as shown in Table 2, using the response ratios or response factors from the most recent multi-level initial calibration (Section 10.3). Other equations may be used if the laboratory demonstrates that those equations produce the same numerical result as produced by the equations below.

For the native analytes:

$$\text{Concentration (ng/L or ng/g)} = \frac{\text{Area}_n M_l}{\text{Area}_l (\overline{RR} \text{ or } \overline{RF})} \times \frac{1}{W_s}$$

where:

Area_n = The measured area of the Q1 m/z for the native (unlabeled) PFAS

Area_l = The measured area at the Q1 m/z for the isotopically labeled PFAS (EIS). *See note below.*

M_l = The mass of the isotopically labeled compound added (ng)

$\overline{RR}$ = Average response ratio used to quantify target compounds by the isotope dilution method

$\overline{RF}$ = Average response factor used to quantify target compounds by the extracted internal standard method

W_s = Sample volume (L) or weight (g)

Note: For better accuracy, PFTrDA is quantitated using the average of the areas of labeled compounds $^{13}\text{C}_2\text{-PFTrDA}$ and $^{13}\text{C}_2\text{-PFDoA}$.

And for the EIS analytes:

$$\text{Concentration (ng/L or ng/g)} = \frac{\text{Area}_l M_{nis}}{\text{Area}_{nis} \overline{RF}_s} \times \frac{1}{W_s}$$

where:

Area_l = The measured area at the Q1 m/z for the isotopically labeled PFAS (EIS)

Area_{nis} = The measured area of the Q1 m/z for the non-extracted internal standard (NIS)

M_{nis} = The mass of the added non-extracted internal standard (NIS) compound (ng)

W_s = Sample volume (L) or weight (g)

$\overline{RF}_s$ = Average response factor used to quantify the isotopically labeled compound by the non-extracted internal standard method

Results for native compounds are recovery corrected by the method of quantification. Extracted internal standard (EIS) recoveries are determined similarly against the non-extracted internal standard (NIS) and are used as general indicators of overall analytical quality.

The instrument measures the target analytes as either their anions or neutral forms. **The default approach for Clean Water Act uses of the method is to report the analytes in their acid or neutral forms,** using the following equation to convert the concentrations:

$$C_{Acid} = C_{Anion} \times \frac{MW_{Acid}}{MW_{Anion}}$$

where:

C_{Anion} = The analyte concentration in anion form

MW_{Acid} = The molecular weight of the acid form

MW_{Anion} = The molecular weight of the anion form

15.3 Sample dilutions

15.3.1 If the Q1 area for any compound exceeds the calibration range of the system, dilute a subsample of the sample extract with 0.1% acetic acid (Section 7.1.2) by a factor no greater than 10x adjust the amount of the NIS in the diluted extract, then analyze the diluted extract using the percent recovery of the EIS from the original analysis. If the compound cannot be measured reliably by isotope dilution, dilute and analyze aqueous sample, or analyze a smaller aliquot of soil, biosolid, sediment, or tissue sample. Adjust the compound concentrations, detection limits, and minimum levels to account for the dilution.

15.3.2 If the recovery of any isotopically labeled compound is outside of the acceptance limits (Table 5), a diluted aqueous sample or smaller aliquot (for solids and tissue) must be analyzed (Section 15.3.1). If the recovery of any isotopically labeled compound in the diluted sample is outside of the normal range, the method does not apply to the sample being analyzed and the result may not be reported or used for permitting or regulatory compliance purposes. In this case, an alternative column could be employed to resolve the interference. If all cleanup procedures in this method and an alternative column have been employed and isotopically labeled compound recovery remains outside of the normal range, extraction and/or cleanup procedures that are beyond this scope of this method will be required to analyze the sample.

15.4 Reporting of analytical results (acid/neutral forms)

The data reporting practices described here are focused on NPDES monitoring needs and may not be relevant to other uses of the method. For analytes reported in their acid form, use the equations in Section 15.2 and the analyte names Table 1. For analytes reported in their anion form, see Table 8 for the appropriate names and CAS Registry Numbers.

15.4.1 Report results for aqueous samples in ng/L. Report results for solid samples in ng/g, on a dry-weight basis, and report the percent solids for each sample separately. Report results for tissue samples in ng/g, on a wet-weight basis. Other units may be used if required in a permit or for a project. Report all QC data with the sample results.

15.4.2 Reporting level

Unless specified otherwise by a regulatory authority or in a discharge permit, results for analytes that meet the identification criteria are reported down to the concentration of the ML established by the laboratory through calibration of the instrument (see the glossary for the derivation of the ML). EPA considers the terms “reporting limit,” “quantitation limit,” “limit of quantitation,” and “minimum level” to be synonymous.

15.4.2.1 Report a result for each analyte in each field sample or QC standard at or above the ML to 3 significant figures. Report a result for each analyte found in each field sample or QC standard below the ML as “<ML,” where ML is the

concentration of the analyte at the ML, or as required by the regulatory/control authority or permit.

- 15.4.2.2** Report a result for each analyte in a blank at or above the MDL to 2 significant figures. Report a result for each analyte found in a blank below the MDL as “<MDL,” where MDL is the concentration of the analyte at the MDL, or as required by the regulatory/control authority or permit.
 - 15.4.2.3** Report a result for an analyte found in a sample or extract that has been diluted at the least dilute level at which the area at the quantitation m/z is within the calibration range (e.g., above the ML for the analyte and below the highest calibration standard) and with isotopically labeled compound recoveries within their respective QC acceptance criteria. This may require reporting results for some analytes from different analyses.
 - 15.4.2.4** Report recoveries of all associated EIS compounds for all field samples and QC standards.
- 15.4.3** Results from tests performed with an analytical system that is not in control (i.e., that does not meet acceptance criteria for any QC tests in this method) must be documented and reported (e.g., as a qualifier on results), unless the failure is not required to be reported as determined by the regulatory/control authority. Results associated with a QC failure cannot be used to demonstrate regulatory compliance. QC failures do not relieve a discharger or permittee of reporting timely results. If the holding time would be exceeded for a reanalysis of the sample, the regulatory/control authority should be consulted for disposition.

16.0 Method Performance

Routine method performance is validated through analysis of matrix-specific reference samples, including spikes and certified reference materials. Ongoing method performance is monitored through QC samples analyzed alongside samples. The parameters monitored include percent recovery of isotopically labeled compounds, blank concentrations, and native compound recoveries.

This method is being validated, and performance specifications will be developed using data from DOD’s interlaboratory validation study (Reference 10). A summary of the single-laboratory performance is presented in Tables 5, 9, and 10.

17.0 Pollution Prevention

- 17.1** Pollution prevention encompasses any technique that reduces or eliminates the quantity or toxicity of waste at the point of generation. Many opportunities for pollution prevention exist in laboratory operation. EPA has established a preferred hierarchy of environmental management techniques that places pollution prevention as the management option of first choice. Whenever feasible, laboratory personnel should use pollution prevention techniques to address waste generation. When wastes cannot be reduced feasibly at the source, EPA recommends recycling as the next best option.
- 17.2** The compounds in this method are used in extremely small amounts and pose little threat to the environment when managed properly. Standards should be prepared in volumes consistent with laboratory use to minimize the disposal of excess volumes of expired standards.

- 17.3** For information about pollution prevention that may be applied to laboratories and research institutions, consult *Less is Better: Laboratory Chemical Management for Waste Reduction* (Reference 7).

18.0 Waste Management

- 18.1** The laboratory is responsible for complying with all Federal, State, and local regulations governing waste management, particularly the hazardous waste identification rules and land disposal restrictions, and to protect the air, water, and land by minimizing and controlling all releases from fume hoods and bench operations. Compliance is also required with any sewage discharge permits and regulations. An overview of requirements can be found in *Environmental Management Guide for Small Laboratories* (Reference 8).
- 18.2** Samples at $\text{pH} < 2$ or $\text{pH} > 12$, are hazardous and must be handled and disposed of as hazardous waste or neutralized and disposed of in accordance with all federal, state, and local regulations. It is the laboratory's responsibility to comply with all federal, state, and local regulations governing waste management, particularly the hazardous waste identification rules and land disposal restrictions.
- 18.3** For further information on waste management, consult *The Waste Management Manual for Laboratory Personnel* and *Less is Better-Laboratory Chemical Management for Waste Reduction*, (Reference 9).

19.0 References

1. "Working with Carcinogens," Department of Health, Education, & Welfare, Public Health Service, Centers for Disease Control, NIOSH, Publication 77-206, August 1977, NTIS PB-277256.
2. "OSHA Safety and Health Standards, General Industry," OSHA 2206, 29 CFR 1910.
3. "Safety in Academic Chemistry Laboratories," ACS Committee on Chemical Safety, 1979.
4. "Standard Methods for the Examination of Water and Wastewater," 18th edition and later revisions, American Public Health Association, 1015 15th St, NW, Washington, DC 20005, 1-35: Section 1090 (Safety), 1992.
5. "Standard Practice for Sampling Water," ASTM Annual Book of Standards, ASTM, 1916 Race Street, Philadelphia, PA 19103-1187, 1980.
6. "Handbook of Analytical Quality Control in Water and Wastewater Laboratories," USEPA EMSL, Cincinnati, OH 45268, EPA 600/4-79-019, April 1979.
7. "Less is Better: Laboratory Chemical Management for Waste Reduction," American Chemical Society, 1993. Available from the American Chemical Society's Department of Government Relations and Science Policy, 1155 16th Street NW, Washington, DC 20036.
8. "Environmental Management Guide for Small Laboratories," USEPA, Small Business Division, Washington DC, EPA 233-B-00-001, May 2000.

9. “The Waste Management Manual for Laboratory Personnel,” American Chemical Society, 1990. Available from the American Chemical Society's Department of Government Relations and Science Policy, 1155 16th Street NW, Washington, DC 20036.
10. *DOD single- laboratory study reference will be added here.*
11. *DOD interlaboratory study reference will be added here.*
12. DoD QSM (US Department of Defense Quality Systems Manual for Environmental Laboratories, version 5.3, 2019)
13. Woudneh, Million B., Bharat Chandramouli, Coreen Hamilton, Richard Grace, 2019, “Effects of Sample Storage on the Quantitative Determination of 29 PFAS: Observation of Analyte Interconversions during Storage”, Environmental Science and Technology 53(21): 12576-12585.

20.0 Tables, Diagrams, Flowcharts, and Validation Data

Table 1. Names, Abbreviations, and CAS Registry Numbers for Target PFAS, Extracted Internal Standards and Non-extracted Internal Standards¹

Target Analyte Name	Abbreviation	CAS Number
Perfluoroalkyl carboxylic acids		
Perfluorobutanoic acid	PFBA	375-22-4
Perfluoropentanoic acid	PFPeA	2706-90-3
Perfluorohexanoic acid	PFHxA	307-24-4
Perfluoroheptanoic acid	PFHpA	375-85-9
Perfluorooctanoic acid	PFOA	335-67-1
Perfluorononanoic acid	PFNA	375-95-1
Perfluorodecanoic acid	PFDA	335-76-2
Perfluoroundecanoic acid	PFUnA	2058-94-8
Perfluorododecanoic acid	PFDoA	307-55-1
Perfluorotridecanoic acid	PFTTrDA	72629-94-8
Perfluorotetradecanoic acid	PFTeDA	376-06-7
Perfluoroalkyl sulfonic acids		
Acid Form		
Perfluorobutanesulfonic acid	PFBS	375-73-5
Perfluoropentanesulfonic acid	PFPeS	2706-91-4
Perfluorohexanesulfonic acid	PFHxS	355-46-4
Perfluoroheptanesulfonic acid	PFHpS	375-92-8
Perfluorooctanesulfonic acid	PFOS	1763-23-1
Perfluorononanesulfonic acid	PFNS	68259-12-1
Perfluorodecanesulfonic acid	PFDS	335-77-3
Perfluorododecanesulfonic acid	PFDoS	79780-39-5
Fluorotelomer sulfonic acids		
1H,1H, 2H, 2H-Perfluorohexane sulfonic acid	4:2FTS	757124-72-4
1H,1H, 2H, 2H-Perfluorooctane sulfonic acid	6:2FTS	27619-97-2
1H,1H, 2H, 2H-Perfluorodecane sulfonic acid	8:2FTS	39108-34-4
Perfluorooctane sulfonamides		
Perfluorooctanesulfonamide	PFOSA	754-91-6
N-methyl perfluorooctanesulfonamide	NMeFOSA	31506-32-8
N-ethyl perfluorooctanesulfonamide	NEtFOSA	4151-50-2
Perfluorooctane sulfonamidoacetic acids		
N-methyl perfluorooctanesulfonamidoacetic acid	NMeFOSAA	2355-31-9
N-ethyl perfluorooctanesulfonamidoacetic acid	NEtFOSAA	2991-50-6
Perfluorooctane sulfonamide ethanols		
N-methyl perfluorooctanesulfonamidoethanol	NMeFOSE	24448-09-7
N-ethyl perfluorooctanesulfonamidoethanol	NEtFOSE	1691-99-2
Per- and Polyfluoroether carboxylic acids		
Hexafluoropropylene oxide dimer acid	HFPO-DA	13252-13-6
4,8-Dioxa-3H-perfluorononanoic acid	ADONA	919005-14-4
Perfluoro-3-methoxypropanoic acid	PFMPA	377-73-1
Perfluoro-4-methoxybutanoic acid	PFMBA	863090-89-5
Nonafluoro-3,6-dioxaheptanoic acid	NFDHA	151772-58-6

Table 1. Names, Abbreviations, and CAS Registry Numbers for Target PFAS, Extracted Internal Standards and Non-extracted Internal Standards¹

Target Analyte Name	Abbreviation	CAS Number
Ether sulfonic acids		
9-Chlorohexadecafluoro-3-oxanonane-1-sulfonic acid	9Cl-PF3ONS	756426-58-1
11-Chloroeicosafluoro-3-oxaundecane-1-sulfonic acid	11Cl-PF3OUdS	763051-92-9
Perfluoro(2-ethoxyethane)sulfonic acid	PFEESA	113507-82-7
Fluorotelomer carboxylic acids		
3-Perfluoropropyl propanoic acid	3:3FTCA	356-02-5
2H,2H,3H,3H-Perfluorooctanoic acid	5:3FTCA	914637-49-3
3-Perfluoroheptyl propanoic acid	7:3FTCA	812-70-4
EIS Compounds		
Perfluoro-n-[¹³ C ₄]butanoic acid	¹³ C ₄ -PFBA	NA
Perfluoro-n-[¹³ C ₅]pentanoic acid	¹³ C ₅ -PFPeA	
Perfluoro-n-[1,2,3,4,6- ¹³ C ₅]hexanoic acid	¹³ C ₅ -PFHxA	
Perfluoro-n-[1,2,3,4- ¹³ C ₄]heptanoic acid	¹³ C ₄ -PFHpA	
Perfluoro-n-[¹³ C ₈]octanoic acid	¹³ C ₈ -PFOA	
Perfluoro-n-[¹³ C ₉]nonanoic acid	¹³ C ₉ -PFNA	
Perfluoro-n-[1,2,3,4,5,6- ¹³ C ₆]decanoic acid	¹³ C ₆ -PFDA	
Perfluoro-n-[1,2,3,4,5,6,7- ¹³ C ₇]undecanoic acid	¹³ C ₇ -PFUnA	
Perfluoro-n-[1,2- ¹³ C ₂]dodecanoic acid	¹³ C ₂ -PFD ₂ OA	
Perfluoro-n-[1,2- ¹³ C ₂]tetradecanoic acid	¹³ C ₂ -PFTeDA	
Perfluoro-1-[2,3,4- ¹³ C ₃]butanesulfonic acid	¹³ C ₃ -PFBS	
Perfluoro-1-[1,2,3- ¹³ C ₃]hexanesulfonic acid	¹³ C ₃ -PFHxS	
Perfluoro-1-[¹³ C ₈]octanesulfonic acid	¹³ C ₈ -PFOS	
Perfluoro-1-[¹³ C ₈]octanesulfonamide	¹³ C ₈ -PFOSA	
N-methyl-d ₃ -perfluoro-1-octanesulfonamidoacetic acid	D ₃ -NMeFOSAA	
N-ethyl-d ₅ -perfluoro-1-octanesulfonamidoacetic acid	D ₅ -NEtFOSAA	
1H,1H,2H,2H-Perfluoro-1-[1,2- ¹³ C ₂]hexan sulfonic acid	¹³ C ₂ -4:2FTS	
1H,1H,2H,2H-Perfluoro-1-[1,2- ¹³ C ₂]octanesulfonic acid	¹³ C ₂ -6:2FTS	
1H,1H,2H,2H-Perfluoro-1-[1,2- ¹³ C ₂]decanesulfonic acid	¹³ C ₂ -8:2FTS	
Tetrafluoro-2-heptafluoropropoxy- ¹³ C ₃ -propanoic acid	¹³ C ₃ -HFPO-DA	
N-methyl-d ₇ -perfluorooctanesulfonamidoethanol	D ₇ -NMeFOSE	
N-ethyl-d ₉ -perfluorooctanesulfonamidoethanol	D ₉ -NEtFOSE	
N-ethyl-d ₅ -perfluoro-1-octanesulfonamide	D ₅ -NEtFOSA	
N-methyl-d ₃ -perfluoro-1-octanesulfonamide	D ₃ -NMeFOSA	
NIS Compounds		
Perfluoro-n-[2,3,4- ¹³ C ₃]butanoic acid	¹³ C ₃ -PFBA	NA
Perfluoro-n-[1,2,3,4- ¹³ C ₄]octanoic acid	¹³ C ₄ -PFOA	
Perfluoro-n-[1,2- ¹³ C ₂]decanoic acid	¹³ C ₂ -PFDA	
Perfluoro-n-[1,2,3,4- ¹³ C ₄]octanesulfonic acid	¹³ C ₄ -PFOS	
Perfluoro-n-[1,2,3,4,5- ¹³ C ₅] nonanoic acid	¹³ C ₅ -PFNA	
Perfluoro-n-[1,2- ¹³ C ₂]hexanoic acid	¹³ C ₂ -PFHxA	
Perfluoro-1-hexane[¹⁸ O ₂]sulfonic acid	¹⁸ O ₂ -PFHxS	

¹ The target analyte names are for the acid and neutral forms of the analytes. See Table 8 for the names and CASRN of the corresponding anion forms, where applicable.

NA Not assigned a CASRN

Table 2. Analyte Ions Monitored, Extracted Internal Standard, and Non-extracted Internal Standard Used for Quantification

Abbreviation	Example Retention Time ¹	Parent Ion Mass	Quantification Ion Mass	Confirmation Ion Mass	Typical Ion Ratio	Quantification Reference Compound
Target Analytes						
PFBA	1.96	212.8	168.9	NA	NA	¹³ C ₄ -PFBA
PFPeA	4.18	263.0	219.0	68.9	NA	¹³ C ₅ -PFPeA
PFHxA	4.81	313.0	269.0	118.9	13	¹³ C ₅ -PFHxA
PFHpA	5.32	363.1	319.0	169.0	3.5	¹³ C ₄ -PFHpA
PFOA	6.16	413.0	369.0	169.0	3.0	¹³ C ₈ -PFOA
PFNA	6.99	463.0	419.0	219.0	4.9	¹³ C ₉ -PFNA
PFDA	7.47	512.9	469.0	219.0	5.5	¹³ C ₆ -PFDA
PFUnA	7.81	563.1	519.0	269.1	6.9	¹³ C ₇ -PFUnA
PFDoA	8.13	613.1	569.0	319.0	10	¹³ C ₂ -PFDoA
PFTTrDA ²	8.53	663.0	619.0	168.9	6.7	avg. ¹³ C ₂ -PFTeDA and ¹³ C ₂ -PFDoA
PFTeDA	8.96	713.1	669.0	168.9	6.0	¹³ C ₂ -PFTeDA
PFBS	4.79	298.7	79.9	98.8	2.1	¹³ C ₃ -PFBS
PFPeS	5.38	349.1	79.9	98.9	1.8	¹³ C ₃ -PFHxS
PFHxS	6.31	398.7	98.9	79.9	1.9	¹³ C ₃ -PFHxS
PFHpS	7.11	449.0	79.9	98.8	1.7	¹³ C ₈ -PFOS
PFOS	7.59	498.9	79.9	98.8	2.3	¹³ C ₈ -PFOS
PFNS	7.92	548.8	79.9	98.8	1.9	¹³ C ₈ -PFOS
PFDS	8.28	599.0	79.9	98.8	1.9	¹³ C ₈ -PFOS
PFDoS	9.14	699.1	79.9	98.8	1.9	¹³ C ₈ -PFOS
4:2FTS	4.67	327.1	307.0	80.9	1.7	¹³ C ₂ -4:2FTS
6:2FTS	5.81	427.1	407.0	80.9	1.9	¹³ C ₂ -6:2FTS
8:2FTS	7.28	527.1	507.0	80.8	3.0	¹³ C ₂ -8:2FTS
PFOSA	8.41	498.1	77.9	478.0	47	¹³ C ₈ -PFOSA
NMeFOSA	9.70	511.9	219.0	169.0	0.66	D ₃ -NMeFOSA
NEtFOSA	9.94	526.0	219.0	169.0	0.63	D ₅ -NEtFOSA
NMeFOSAA	7.51	570.1	419.0	483.0	2.0	D ₃ -NMeFOSAA
NEtFOSAA	7.65	584.2	419.1	526.0	1.2	D ₅ -N-EtFOSAA
NMeFOSE	9.57	616.1	58.9	NA	NA	D ₇ -NMeFOSE
NEtFOSE	9.85	630.0	58.9	NA	NA	D ₉ -NEtFOSE
HFPO-DA	4.97	284.9	168.9	184.9	1.95	¹³ C ₃ -HFPO-DA
ADONA	5.79	376.9	250.9	84.8	2.8	¹³ C ₃ -HFPO-DA
9Cl-PF3ONS	7.82	530.8	351.0	532.8→353.0	3.2	¹³ C ₃ -HFPO-DA
11Cl-PF3OUdS	8.62	630.9	450.9	632.9→452.9	3.0	¹³ C ₃ -HFPO-DA
3:3FTCA	3.89	241.0	177.0	117.0	1.70	¹³ C ₅ -PFPeA
5:3FTCA	5.14	341.0	237.1	217.0	1.16	¹³ C ₅ -PFHxA
7:3FTCA	6.76	441.0	316.9	336.9	0.69	¹³ C ₅ -PFHxA
PFEESA	5.08	314.8	134.9	82.9	9.22	¹³ C ₅ -PFHxA
PFMPA	3.21	229.0	84.9	NA	NA	¹³ C ₅ -PFPeA
PFMBA	4.53	279.0	85.1	NA	NA	¹³ C ₅ -PFPeA
NFDHA	4.84	295.0	201.0	84.9	1.46	¹³ C ₅ -PFHxA
Extracted Internal Standards						
¹³ C ₄ -PFBA	1.95	216.8	171.9	NA		¹³ C ₃ -PFBA
¹³ C ₅ -PFPeA	4.18	268.3	223.0	NA		¹³ C ₂ -PFHxA
¹³ C ₅ -PFHxA	4.80	318.0	273.0	120.3		¹³ C ₂ -PFHxA
¹³ C ₄ -PFHpA	5.32	367.1	322.0	NA		¹³ C ₂ -PFHxA
¹³ C ₈ -PFOA	6.16	421.1	376.0	NA		¹³ C ₄ -PFOA

Table 2. Analyte Ions Monitored, Extracted Internal Standard, and Non-extracted Internal Standard Used for Quantification

Abbreviation	Example Retention Time ¹	Parent Ion Mass	Quantification Ion Mass	Confirmation Ion Mass	Typical Ion Ratio	Quantification Reference Compound
¹³ C ₉ -PFNA	6.99	472.1	427.0	NA		¹³ C ₅ -PFNA
¹³ C ₆ -PFDA	7.47	519.1	474.1	NA		¹³ C ₂ -PFDA
¹³ C ₇ -PFUnA	7.81	570.0	525.1	NA		¹³ C ₂ -PFDA
¹³ C ₂ -PFDoA	8.13	615.1	570.0	NA		¹³ C ₂ -PFDA
¹³ C ₂ -PFTeDA	8.96	715.2	670.0	NA		¹³ C ₂ -PFDA
¹³ C ₃ -PFBS	4.78	302.1	79.9	98.9		¹⁸ O ₂ -PFHxS
¹³ C ₃ -PFHxS	6.30	402.1	79.9	98.8		¹⁸ O ₂ -PFHxS
¹³ C ₈ -PFOS	7.59	507.1	98.9	79.9		¹³ C ₄ -PFOS
¹³ C ₂ -4:2FTS	4.67	329.1	80.9	309.0		¹⁸ O ₂ -PFHxS
¹³ C ₂ -6:2FTS	5.82	429.1	80.9	409.0		¹⁸ O ₂ -PFHxS
¹³ C ₂ -8:2FTS	7.28	529.1	80.9	509.0		¹⁸ O ₂ -PFHxS
¹³ C ₈ -PFOSA	8.41	506.1	77.8	NA		¹³ C ₄ -PFOS
D ₃ -NMeFOSA	9.70	515.0	219.0	NA		¹³ C ₄ -PFOS
D ₅ -NEtFOSA	9.94	531.1	219.0	NA		¹³ C ₄ -PFOS
D ₃ -NMeFOSAA	7.51	573.2	419.0	NA		¹³ C ₄ -PFOS
D ₅ -NEtFOSAA	7.65	589.2	419.0	NA		¹³ C ₄ -PFOS
D ₇ -NMeFOSE	9.56	623.2	58.9	NA		¹³ C ₄ -PFOS
D ₉ -NEtFOSE	9.83	639.2	58.9	NA		¹³ C ₄ -PFOS
¹³ C ₃ -HFPO-DA	4.97	284.9	168.9	184.9		¹³ C ₂ -PFHxA
Non-Extracted Internal Standards						
¹³ C ₃ -PFBA	1.95	216.0	172.0	NA		
¹³ C ₂ -PFHxA	4.80	315.1	270.0	119.4		
¹³ C ₄ -PFOA	6.16	417.1	172.0	NA		
¹³ C ₅ -PFNA	6.99	468.0	423.0	NA		
¹³ C ₂ -PFDA	7.47	515.1	470.1	NA		
¹⁸ O ₂ -PFHxS	6.30	403.0	83.9	NA		
¹³ C ₄ -PFOS	7.59	502.8	79.9	98.9		

¹ Times shown are in decimal minute units. Example retention times are based on the instrument operating conditions and column specified in Section 10.2.

² For improved accuracy, PFTrDA is quantitated using the average areas of the labeled compounds ¹³C₂-PFTeDA and ¹³C₂-PFDoA.

Table 3. Nominal Masses of Spike Added to Samples or Extracts

Analyte	Amount Added (ng)
Extracted Internal Standards	
¹³ C ₄ -PFBA	40
¹³ C ₅ -PFPeA	20
¹³ C ₅ -PFHxA	10
¹³ C ₄ -PFHpA	10
¹³ C ₈ -PFOA	10
¹³ C ₉ -PFNA	5
¹³ C ₆ -PFDA	5
¹³ C ₇ -PFUnA	5
¹³ C ₂ -PFD ₀ A	5
¹³ C ₂ -PFTeDA	5
¹³ C ₃ -PFBS	10
¹³ C ₃ -PFHxS	10
¹³ C ₈ -PFOS	10
¹³ C ₂ -4:2FTS	20
¹³ C ₂ -6:2FTS	20
¹³ C ₂ -8:2FTS	20
¹³ C ₈ -PFOSA	10
D ₃ -NMeFOSA	10
D ₅ -NEtFOSA	10
D ₃ -NMeFOSAA	20
D ₅ -NEtFOSAA	20
D ₇ -NMeFOSE	100
D ₉ -NEtFOSE	100
¹³ C ₃ -HFPO-DA	40
Non-extracted Internal Standards	
¹³ C ₃ -PFBA	20
¹³ C ₂ -PFHxA	10
¹³ C ₄ -PFOA	10
¹³ C ₅ -PFNA	5
¹³ C ₂ -PFDA	5
¹⁸ O ₂ -PFHxS	10
¹³ C ₄ -PFOS	10

Table 4. Calibration Solutions (ng/mL)

Compound	CS1 (LOQ)	CS2	CS3	CS4 (CV ¹)	CS5	CS6	CS7 ²
Perfluoroalkyl carboxylic acids							
PFBA	0.8	2	5	10	20	50	250
PFPeA	0.4	1	2.5	5	10	25	125
PFHxA	0.2	0.5	1.25	2.5	5	12.5	62.5
PFHpA	0.2	0.5	1.25	2.5	5	12.5	62.5
PFOA	0.2	0.5	1.25	2.5	5	12.5	62.5
PFNA	0.2	0.5	1.25	2.5	5	12.5	62.5
PFDA	0.2	0.5	1.25	2.5	5	12.5	62.5
PFUnA	0.2	0.5	1.25	2.5	5	12.5	62.5
PFDoA	0.2	0.5	1.25	2.5	5	12.5	62.5
PFTTrDA	0.2	0.5	1.25	2.5	5	12.5	62.5
PFTeDA	0.2	0.5	1.25	2.5	5	12.5	62.5
Perfluoroalkyl sulfonic acids							
PFBS	0.2	0.5	1.25	2.5	5	12.5	62.5
PFPeS	0.2	0.5	1.25	2.5	5	12.5	62.5
PFHxS	0.2	0.5	1.25	2.5	5	12.5	62.5
PFHpS	0.2	0.5	1.25	2.5	5	12.5	62.5
PFOS	0.2	0.5	1.25	2.5	5	12.5	62.5
PFNS	0.2	0.5	1.25	2.5	5	12.5	62.5
PFDS	0.2	0.5	1.25	2.5	5	12.5	62.5
PFDoS	0.2	0.5	1.25	2.5	5	12.5	62.5
Fluorotelomer sulfonic acids							
4:2FTS	0.8	2	5	10	20	50	NA
6:2FTS	0.8	2	5	10	20	50	NA
8:2FTS	0.8	2	5	10	20	50	NA
Perfluorooctane sulfonamides							
PFOSA	0.2	0.5	1.25	2.5	5	12.5	62.5
NMeFOSA	0.2	0.5	1.25	2.5	5	12.5	62.5
NEtFOSA	0.2	0.5	1.25	2.5	5	12.5	62.5
Perfluorooctane sulfonamidoacetic acids							
NMeFOSAA	0.2	0.5	1.25	2.5	5	12.5	62.5
NEtFOSAA	0.2	0.5	1.25	2.5	5	12.5	62.5
Perfluorooctane sulfonamide ethanols							
NMeFOSE	2	5	12.5	25	50	125	625
NEtFOSE	2	5	12.5	25	50	125	625
Per- and polyfluoroether carboxylic acids							
HFPO-DA	0.8	2	5	10	20	50	250
ADONA	0.8	2	5	10	20	50	250
PFMPA	0.4	1	2.5	5	10	25	125
PFMBA	0.4	1	2.5	5	10	25	125
NFDHA	0.4	1	2.5	5	10	25	125
Ether sulfonic acids							
9Cl-PF3ONS	0.8	2	5	10	20	50	250
11Cl-PF3OUdS	0.8	2	5	10	20	50	250
PFEESA	0.4	1	2.5	5	10	25	125

Table 4. Calibration Solutions (ng/mL)

Compound	CS1 (LOQ)	CS2	CS3	CS4 (CV ¹)	CS5	CS6	CS7 ²
Fluorotelomer carboxylic acids							
3:3FTCA	1.0	2.5	6.26	12.5	25	62.4	312
5:3FTCA	5.0	12.5	31.3	62.5	125	312	1560
7:3FTCA	5.0	12.5	31.3	62.5	125	312	1560
Extracted Internal Standard (EIS) Analytes							
¹³ C ₄ -PFBA	10	10	10	10	10	10	10
¹³ C ₅ -PFPeA	5	5	5	5	5	5	5
¹³ C ₅ -PFHxA	2.5	2.5	2.5	2.5	2.5	2.5	2.5
¹³ C ₄ -PFHpA	2.5	2.5	2.5	2.5	2.5	2.5	2.5
¹³ C ₈ -PFOA	2.5	2.5	2.5	2.5	2.5	2.5	2.5
¹³ C ₉ -PFNA	1.25	1.25	1.25	1.25	1.25	1.25	1.25
¹³ C ₆ -PFDA	1.25	1.25	1.25	1.25	1.25	1.25	1.25
¹³ C ₇ -PFUnA	1.25	1.25	1.25	1.25	1.25	1.25	1.25
¹³ C ₂ -PFDaA	1.25	1.25	1.25	1.25	1.25	1.25	1.25
¹³ C ₂ -PFTeDA	1.25	1.25	1.25	1.25	1.25	1.25	1.25
¹³ C ₃ -PFBS	2.5	2.5	2.5	2.5	2.5	2.5	2.5
¹³ C ₃ -PFHxS	2.5	2.5	2.5	2.5	2.5	2.5	2.5
¹³ C ₈ -PFOS	2.5	2.5	2.5	2.5	2.5	2.5	2.5
¹³ C ₂ -4:2 FTS	5	5	5	5	5	5	5
¹³ C ₂ -6:2 FTS	5	5	5	5	5	5	5
¹³ C ₂ -8:2 FTS	5	5	5	5	5	5	5
¹³ C ₈ -PFOSA	2.5	2.5	2.5	2.5	2.5	2.5	2.5
D ₃ -NMeFOSA	2.5	2.5	2.5	2.5	2.5	2.5	2.5
D ₅ -NEtFOSA	2.5	2.5	2.5	2.5	2.5	2.5	2.5
D ₃ -NMeFOSAA	5	5	5	5	5	5	5
D ₅ -NEtFOSAA	5	5	5	5	5	5	5
D ₇ -NMeFOSE	25	25	25	25	25	25	25
D ₉ -NEtFOSE	25	25	25	25	25	25	25
¹³ C ₃ -HFPO-DA	10	10	10	10	10	10	10
Non-extracted Internal Standard (NIS) Analytes							
¹³ C ₃ -PFBA	5	5	5	5	5	5	5
¹³ C ₂ -PFHxA	2.5	2.5	2.5	2.5	2.5	2.5	2.5
¹³ C ₄ -PFOA	2.5	2.5	2.5	2.5	2.5	2.5	2.5
¹³ C ₅ -PFNA	1.25	1.25	1.25	1.25	1.25	1.25	1.25
¹³ C ₂ -PFDA	1.25	1.25	1.25	1.25	1.25	1.25	1.25
¹⁸ O ₂ -PFHxS	2.5	2.5	2.5	2.5	2.5	2.5	2.5
¹³ C ₄ -PFOS	2.5	2.5	2.5	2.5	2.5	2.5	2.5

¹ This calibration point is used as the calibration verification (CV)

² A minimum of six contiguous calibrations standards are required for linear models and a minimum of seven calibration standards are required for second-order models.

Table 5. Single-Laboratory Validation Performance Summary for Target Compounds and Extracted Internal Standards

Compounds	Blank (ng/mL)	Aqueous Matrices ¹			Solid Matrices ¹			Tissue Matrices ¹		
		IPR Rec (%)	RSD (%)	OPR Rec (%)	IPR Rec (%)	RSD (%)	OPR Rec (%)	IPR Rec (%)	RSD (%)	OPR Rec (%)
Target Compounds										
PFBA	< 0.4	89 - 107	4.8	89 – 113	95 – 99	1.0	92 – 108	89 – 104	3.9	90 – 110
PFPeA	< 0.2	85 - 106	5.5	89 – 121	92 – 105	3.4	94 – 115	80 – 98	5.0	96 – 114
PFHxA	< 0.1	75 - 109	9.1	89 – 111	93 – 101	2.2	89 – 107	72 – 110	10.2	90 – 111
PFHpA	< 0.1	87 - 102	4.1	90 – 110	94 – 102	2.2	89 – 107	87 – 102	4.0	87 – 118
PFOA	< 0.1	88 - 98	2.8	87 – 112	92 – 100	2.0	90 – 106	78 – 85	2.4	82 – 114
PFNA	< 0.1	88 - 104	4.1	90 – 111	91 – 102	2.7	88 – 112	85 – 110	6.3	87 – 119
PFDA	< 0.1	82 - 115	8.3	92 – 115	97 – 103	1.5	89 – 118	76 – 115	10.2	84 – 112
PFUnA	< 0.1	83 - 98	4.2	89 – 112	91 – 107	4.0	92 – 111	83 – 102	5.1	91 – 117
PFDoA	< 0.1	58 - 111	15.7	84 – 123	73 – 120	12.1	88 – 119	83 – 105	5.7	77 – 141
PFTrDA	< 0.1	80 - 111	8.1	92 – 119	91 – 112	5.2	89 – 125	92 – 114	5.3	106 – 133
PFTeDA	< 0.1	88 - 103	4.1	89 – 116	94 – 104	2.5	92 – 110	76 – 103	7.4	91 – 111
PFBS	< 0.1	85 - 111	6.6	87 – 116	91 – 103	3.2	91 – 111	69 – 105	10.3	89 – 117
PFPeS	< 0.1	87 - 115	6.9	87 – 115	87 – 103	4.3	89 – 112	77 – 96	5.4	89 – 112
PFHxS	< 0.1	90 - 107	4.4	97 – 119	98 – 106	2.0	96 – 113	81 – 101	5.3	91 – 123
PFHpS	< 0.1	84 - 126	10.2	86 – 114	87 – 104	4.4	88 – 104	77 – 108	8.4	86 – 108
PFOS	< 0.1	93 - 122	6.7	91 – 120	95 – 108	3.4	94 – 115	98 – 112	3.2	97 – 124
PFNS	< 0.1	64 - 141	18.8	86 – 123	98 – 111	3.0	76 – 117	65 – 88	7.5	85 – 114
PFDS	< 0.1	75 - 121	11.7	84 – 107	83 – 102	5.2	84 – 107	82 – 94	3.6	78 – 110
PFDoS	< 0.1	74 - 114	10.6	78 – 102	76 – 99	6.5	77 – 100	73 – 96	6.9	29 – 108
4:2FTS	< 0.4	76 - 123	12.0	91 – 119	98 – 100	0.5	87 – 113	66 – 126	15.6	90 – 103
6:2FTS	< 0.4	71 - 148	17.5	81 – 129	94 – 123	6.5	60 – 166	77 – 105	7.8	92 – 119
8:2FTS	< 0.4	85 - 109	6.1	99 – 124	109 – 128	3.8	104 – 127	66 – 148	19.3	102 – 136
PFOSA	< 0.1	90 - 107	4.4	91 – 122	92 – 106	3.4	94 – 114	92 – 116	5.7	96 – 121
NMeFOSA	< 0.1	78 - 90	3.6	84 – 112	87 – 104	4.4	91 – 117	81 – 100	5.5	86 – 117
NEtFOSA	< 0.1	79 - 97	5.0	83 – 108	98 – 102	1.0	96 – 115	74 – 114	10.7	90 – 127
NMeFOSAA	< 0.1	82 - 115	8.2	81 – 120	91 – 107	4.0	90 – 113	89 – 136	10.4	93 – 117
NEtFOSAA	< 0.1	79 - 120	10.3	85 – 124	102 – 108	1.6	87 – 117	53 – 115	18.3	90 – 117
NMeFOSE	< 1	87 - 102	3.9	92 – 115	98 – 103	1.3	94 – 112	71 – 292	30.3	118 – 344
NEtFOSE	< 1	87 - 104	4.7	91 – 118	97 – 104	1.9	96 – 115	97 – 133	8.0	61 – 159
HFPO-DA	< 0.4	88 - 114	6.5	84 – 118	83 – 105	5.9	80 – 120	73 – 100	7.8	86 – 114
ADONA	< 0.4	77 - 106	7.9	77 – 117	85 – 96	3.2	76 – 124	82 – 95	3.8	86 – 132
PFMPA	< 0.2	86 - 106	6.6	83 – 120	91 – 98	1.8	85 – 117	78 – 93	4.2	86 – 109
PFMBA	< 0.2	62 - 122	5.2	81 – 115	88 – 97	2.6	85 – 120	74 – 104	8.4	84 – 117
NFDHA	< 0.2	44 - 149	16.3	56 – 138	53 – 103	16.2	58 – 136	49 – 86	13.8	56 – 115
9Cl-PF3ONS	< 0.4	84 - 101	27.4	80 – 120	84 – 100	4.4	79 – 131	69 – 98	8.7	95 – 126
11Cl-PF3OUdS	< 0.4	80 - 95	4.5	76 – 116	84 – 96	3.3	77 – 127	85 – 100	4.3	94 – 138
PFEESA	< 0.2	80 - 104	4.4	85 – 115	80 – 93	3.8	89 – 109	68 – 99	9.3	88 – 107
3:3FTCA	< 0.5	84 - 103	5.0	66 – 127	86 – 98	3.3	76 – 116	66 – 94	9.0	41 – 126
5:3FTCA	< 2.5	84 - 101	4.6	84 – 113	83 – 94	3.1	80 – 101	95 – 131	7.9	78 – 199
7:3FTCA	< 2.5	78 - 103	7.0	82 – 116	90 – 106	4.1	75 – 104	84 – 111	6.7	99 – 139

Table 5. Single-Laboratory Validation Performance Summary for Target Compounds and Extracted Internal Standards

Compounds	Blank (ng/mL)	Aqueous Matrices ¹			Solid Matrices ¹			Tissue Matrices ¹		
		IPR Rec (%)	RSD (%)	OPR Rec (%)	IPR Rec (%)	RSD (%)	OPR Rec (%)	IPR Rec (%)	RSD (%)	OPR Rec (%)
Extracted Internal Standard (EIS)										
¹³ C ₄ -PFBA	N/A	85 - 91	1.6	88 – 108	92 – 99	1.6	95 – 109	93 – 97	1.0	95 – 105
¹³ C ₅ -PFPeA	N/A	87 - 95	2.4	84 – 111	86 – 106	5.3	80 – 110	85 – 108	6.0	89 – 103
¹³ C ₅ -PFHxA	N/A	85 - 92	1.9	83 – 108	83 – 101	4.8	92 – 106	79 – 111	8.5	88 – 98
¹³ C ₄ -PFHpA	N/A	78 - 100	6.2	83 – 106	87 – 102	4.1	90 – 100	88 – 93	1.3	80 – 102
¹³ C ₈ -PFOA	N/A	77 - 98	6.0	84 – 107	89 – 101	3.2	92 – 104	91 – 98	1.7	86 – 102
¹³ C ₉ -PFNA	N/A	82 - 96	3.8	84 – 107	86 – 101	4.1	90 – 106	91 – 104	3.3	89 – 101
¹³ C ₆ -PFDA	N/A	81 - 98	4.7	84 – 106	79 – 101	6.0	86 – 109	89 – 104	4.0	90 – 104
¹³ C ₇ -PFUnA	N/A	84 - 100	4.4	84 – 109	84 – 104	5.4	91 – 116	84 – 118	8.4	88 – 109
¹³ C ₂ -PFD ₂ O ₂ A	N/A	61 - 103	12.9	73 – 101	70 – 93	7.1	73 – 106	95 – 125	6.8	70 – 108
¹³ C ₂ -PFTeDA	N/A	72 - 89	5.4	74 – 97	83 – 88	1.5	74 – 107	81 – 114	8.5	10 – 110
¹³ C ₃ -PFBS	N/A	87 - 94	2.0	88 – 110	97 – 105	1.8	96 – 109	87 – 114	6.5	95 – 106
¹³ C ₃ -PFHxS	N/A	83 - 89	1.9	85 – 103	92 – 97	1.4	92 – 106	92 – 97	1.4	91 – 103
¹³ C ₈ -PFOS	N/A	78 - 92	3.9	86 – 110	87 – 107	4.9	95 – 109	87 – 93	1.6	95 – 103
¹³ C ₂ -4:2 FTS	N/A	64 - 106	12.1	87 – 137	132 – 135	0.6	123 – 145	106 – 221	17.6	155 – 291
¹³ C ₂ -6:2 FTS	N/A	93 - 102	2.2	67 – 149	118 – 129	2.3	104 – 138	87 – 135	10.8	117 – 149
¹³ C ₂ -8:2 FTS	N/A	99 - 109	2.5	71 – 137	96 – 122	6.1	93 – 123	179 – 299	12.5	79 – 304
¹³ C ₈ -PFOSA	N/A	60 - 107	14.2	57 – 109	69 – 86	5.4	66 – 100	104 – 153	9.4	88 – 120
D ₃ -NMeFOSA	N/A	55 - 85	10.8	39 – 84	47 – 59	5.4	25 – 64	20 – 58	24.5	3 – 34
D ₅ -NEtFOSA	N/A	54 - 91	12.9	43 – 84	43 – 51	4.5	18 – 58	30 – 56	15.2	0 – 56*
D ₃ -NMeFOSAA	N/A	63 - 117	14.9	66 – 117	98 – 107	2.1	86 – 109	102 – 187	14.7	144 – 196
D ₅ -NEtFOSAA	N/A	66 - 115	13.7	63 – 115	98 – 104	1.3	85 – 109	178 – 216	4.9	175 – 223
D ₇ -NMeFOSE	N/A	61 - 106	13.6	42 – 99	50 – 61	5.1	35 – 76	3 – 5	11.6	0 – 8*
D ₉ -NEtFOSE	N/A	63 - 108	13.2	44 – 90	46 – 57	5.5	32 – 72	8 – 33	30.0	0 – 33*
¹³ C ₃ -HFPO-DA	N/A	89 - 106	4.5	88 – 121	98 – 108	2.4	83 – 125	87 – 106	4.9	81 – 106

¹ The recovery limits are applied to all samples, method blanks, IPR, OPR samples for all matrix types.

* Ranges were determined at ± 2 standard deviations from the mean. Because of the low recoveries for these EIS, the calculated lower limits were negative values. Therefore, the lower limits have been set to 0 for these analytes.

Data for this table are derived from the single-laboratory validation study, and are only provided as examples for this draft method. The data will be updated to reflect the interlaboratory study results in a subsequent revision. Therefore, these criteria will change after interlaboratory validation. Several sections of this method state that Table 5 criteria are required, this is standard language that will be applicable when the method is finalized.

Compound	Aqueous (ng/L)		Solid (ng/g)		Tissue (ng/g)	
	MDL_s	ML	MDL_s	ML	MDL_s	ML
PFBA	0.330	6.4	0.401	0.8	0.593	2.0
PFPeA	0.196	3.2	0.021	0.4	0.083	1.0
PFHxA	0.318	1.6	0.020	0.2	0.096	0.5
PFHpA	0.221	1.6	0.029	0.2	0.088	0.5
PFOA	0.302	1.6	0.037	0.2	0.086	0.5
PFNA	0.221	1.6	0.086	0.2	0.160	0.5
PFDA	0.333	1.6	0.031	0.2	0.124	0.5
PFUnA	0.264	1.6	0.033	0.2	0.152	0.5
PFDoA	0.379	1.6	0.059	0.2	0.130	0.5
PFTTrDA	0.238	1.6	0.038	0.2	0.086	0.5
PFTeDA	0.264	1.6	0.032	0.2	0.185	0.5
PFBS	0.245	1.6	0.014	0.2	0.070	0.5
PFPeS	0.204	1.6	0.015	0.2	0.032	0.5
PFHxS ¹	0.217	1.6	0.018	0.2	0.083	0.5
PFHpS	0.137	1.6	0.057	0.2	0.043	0.5
PFOS ¹	0.327	1.6	0.067	0.2	0.294	0.5
PFNS	0.303	1.6	0.046	0.2	0.114	0.5
PFDS	0.334	1.6	0.040	0.2	0.101	0.5
PFDoS	0.179	1.6	0.038	0.2	0.177	0.5
4:2 FTS	2.281	6.4	0.282	0.8	0.740	2.0
6:2 FTS	3.973	6.4	0.116	0.8	1.149	2.0
8:2 FTS	1.566	6.4	0.225	0.8	0.373	2.0
PFOSA	0.227	1.6	0.068	0.2	0.094	0.5
NMeFOSA	0.196	1.6	0.049	0.2	0.161	0.5
NEtFOSA	0.585	1.6	0.038	0.2	0.169	0.5
NMeFOSAA ¹	0.586	1.6	0.030	0.2	0.093	0.5
NEtFOSAA ¹	0.324	1.6	0.044	0.2	0.138	0.5
NMeFOSE	1.191	16	0.203	2.0	9.978	5.0
NEtFOSE	1.022	16	0.247	2.0	1.501	5.0
HFPO-DA	0.406	6.4	0.136	0.8	0.161	2.0
ADONA	0.779	6.4	0.057	0.8	0.082	2.0
PFEESA	0.137	3.2	0.018	0.4	0.045	1.0
PFMPA	0.177	3.2	0.033	0.4	0.070	1.0
PFMBA	0.117	3.2	0.029	0.4	0.069	1.0
NFDHA	1.384	3.2	0.084	0.4	0.294	1.0
9CL-PF3ONS	0.871	6.4	0.038	0.8	0.152	2.0
11CL-PF3OUDS	0.819	6.4	0.071	0.8	0.312	2.0
3:3 FTCA	0.721	8.0	0.060	1.0	0.247	2.5
5:3 FTCA	5.066	40	0.363	5.0	1.537	12.5
7:3 FTCA	5.942	40	0.308	5.0	0.845	12.5

¹ A standard containing a mixture of branched and linear isomer of suitable quality to be used for quantitation is currently available and required to be used for all calibration, calibration verifications, and QC samples. If more become commercially available for other target analytes, they must be utilized in the same manner.

Data for this table are derived from the single-laboratory validation study, and are only provided as examples for this draft method. The data will be updated with the pooled MDLs from the interlaboratory study results in a subsequent revision.

Table 7. Summary of Quality Control

Method Reference	Requirement	Specification and Frequency
Section 10.1	Mass Calibration	Annually and on as-needed basis
Section 10.1.5	Mass Calibration Verification	After mass calibration
Section 10.3	Initial Calibration (ICAL)	Minimum 6 calibration standards for linear model and 7 calibration standards for non-linear models.
Sections 10.2.2, 14.4	Retention Time (RT) window	After ICAL and at the beginning of analytical sequence
Sections 7.3.1, 9.4	Extracted Internal Standard (EIS) Analytes	All CAL standards, batch QC and field samples
Sections 7.3.2	Non-extracted Internal Standards (NIS)	All CAL standards, batch QC and field samples
Sections 7.3.4, 10.3.1, 13.3	Instrument Sensitivity Check (ISC)	Daily, prior to analysis
Section 14.2	Calibration Verification (CV)	At the beginning and every 10 samples
Section 14.6	Instrument Blank	Daily prior to analysis and after high standards
Sections 9.1.3, 9.5, 14.7	Method Blank (MB)	One per preparation batch
Section 14.5	Ongoing Precision Recovery (OPR)	One per preparation batch
Section 11.0	Limit of Quantitation Verification (LLOPR)	Prior to analyzing samples
Section 11.0	Matrix Spike (MS/MSD)	One per preparation batch (if required)

Table 8. Cross-reference of Abbreviations, Analyte Names, CAS Numbers for the Acid and Anion Forms of the Perfluoroalkyl carboxylates and Perfluoroalkyl sulfonates

Perfluoroalkyl carboxylic acids/anions				
Abbreviation	Acid Name	CASRN	Anion Name	CASRN
PFBA	Perfluorobutanoic acid	375-22-4	Perfluorobutanoate	45048-62-2
PFPeA	Perfluoropentanoic acid	2706-90-3	Perfluoropentanoate	45167-47-3
PFHxA	Perfluorohexanoic acid	307-24-4	Perfluorohexanoate	92612-52-7
PFHpA	Perfluoroheptanoic acid	375-85-9	Perfluoroheptanoate	120885-29-2
PFOA	Perfluorooctanoic acid	335-67-1	Perfluorooctanoate	45285-51-6
PFNA	Perfluorononanoic acid	375-95-1	Perfluorononanoate	72007-68-2
PFDA	Perfluorodecanoic acid	335-76-2	Perfluorodecanoate	73829-36-4
PFUnA	Perfluoroundecanoic acid	2058-94-8	Perfluoroundecanoate	196859-54-8
PFDoA	Perfluorododecanoic acid	307-55-1	Perfluorododecanoate	171978-95-3
PFTTrDA	Perfluorotridecanoic acid	72629-94-8	Perfluorotridecanoate	862374-87-6
PFTeDA	Perfluorotetradecanoic acid	376-06-7	Perfluorotetradecanoate	365971-87-5
Perfluoroalkyl sulfonic acids/anions				
PFBS	Perfluorobutanesulfonic acid	375-73-5	Perfluorobutane sulfonate	45187-15-3
PFPeS	Perfluoropentanesulfonic acid	2706-91-4	Perfluoropentane sulfonate	175905-36-9
PFHxS	Perfluorohexanesulfonic acid	355-46-4	Perfluorohexane sulfonate	108427-53-8
PFHpS	Perfluoroheptanesulfonic acid	375-92-8	Perfluoroheptane sulfonate	146689-46-5
PFOS	Perfluorooctanesulfonic acid	1763-23-1	Perfluorooctane sulfonate	45298-90-6
PFNS	Perfluorononanesulfonic acid	68259-12-1	Perfluorononane sulfonate	474511-07-4
PFDS	Perfluorodecanesulfonic acid	335-77-3	Perfluorodecane sulfonate	126105-34-8
PFDoS	Perfluorododecanesulfonic acid	79780-39-5	Perfluorododecane sulfonate	343629-43-6

Table 9. Range of Recoveries for Extracted Internal Standards (EIS) in the Single-laboratory Validation Study, by Matrix

EIS Compounds	Aqueous			Solid			Tissue		
	% Recovery		RSD (%)	% Recovery		RSD (%)	% Recovery		RSD (%)
	Min	Max		Min	Max		Min	Max	
¹³ C ₄ -PFBA	9	97	15.9	3	113	37.4	84	99	8.0
¹³ C ₅ -PFPeA	39	103	13.3	28	112	17.2	86	107	11.1
¹³ C ₅ -PFHxA	73	97	2.7	79	110	5.5	92	95	1.6
¹³ C ₄ -PFHpA	77	95	2.4	73	111	6.0	80	93	8.2
¹³ C ₈ -PFOA	87	95	0.8	86	115	4.4	90	95	2.8
¹³ C ₉ -PFNA	82	95	1.6	87	110	4.2	90	98	4.3
¹³ C ₆ -PFDA	71	93	3.3	87	112	4.9	83	97	7.7
¹³ C ₇ -PFUnA	56	94	6.5	66	124	11.6	71	91	12.9
¹³ C ₂ -PFD ₀ A	34	87	13.7	26	109	24.3	54	96	29.2
¹³ C ₂ -PFTeDA	17	153	26.2	18	110	30.1	31	102	67.8
¹³ C ₃ -PFBS	72	100	4.7	89	120	5.4	89	98	5.1
¹³ C ₃ -PFHxS	79	95	1.6	87	110	4.4	98	99	0.1
¹³ C ₈ -PFOS	67	96	3.6	79	113	5.7	92	103	6.0
¹³ C ₂ -4:2FTS	81	199	14.8	95	248	17.0	192	215	6.2
¹³ C ₂ -6:2FTS	64	183	16.4	76	127	9.4	145	230	27.2
¹³ C ₂ -8:2FTS	65	139	8.4	86	173	15.2	136	220	24.6
¹³ C ₈ -PFOSA	27	93	15.4	61	123	10.0	87	96	4.5
D ₃ -NMeFOSA	14	74	16.4	28	86	22.7	8	38	61.9
D ₅ -NEtFOSA	12	70	16.5	21	70	25.5	8	30	57.8
D ₃ -NMeFOSAA	21	113	7.3	52	142	14.8	106	139	13.1
D ₅ -NEtFOSAA	12	106	8.2	68	151	16.9	79	151	31.8
D ₇ -NMeFOSE	11	77	18.6	13	107	27.9	5	30	81.1
D ₉ -NEtFOSE	8	73	19.6	16	97	30.4	0	29	103.1
¹³ C ₃ -HFPO-DA	92	113	2.0	70	119	10.4	93	102	5.1

Data for this table are derived from the single-laboratory validation study, and are only provided as examples for this draft method. The data will be updated with the interlaboratory study results in a subsequent revision.

Table 10. Range of Recoveries for Non-Extracted Internal Standards in the Single-laboratory Validation Study, by Matrix

NIS Compounds	Aqueous			Solid			Tissue		
	% Recovery		RSD (%)	% Recovery		RSD (%)	% Recovery		RSD (%)
	Min	Max		Min	Max		Min	Max	
¹³ C ₃ -PFBA	60	91	10.3	54	89	6.4	51	82	7.0
¹³ C ₂ -PFHxA	43	94	18.6	52	90	7.4	41	80	19.3
¹³ C ₄ -PFOA	59	87	9.7	54	89	6.4	51	82	9.5
¹³ C ₅ -PFNA	64	87	7.5	59	94	7.1	52	88	11.2
¹³ C ₂ -PFDA	57	86	10.0	55	91	8.6	47	85	19.4
¹⁸ O ₂ -PFHxS	59	87	9.6	53	87	7.1	51	80	8.1
¹³ C ₄ -PFOS	60	82	7.5	58	86	7.0	52	85	10.3

Data for this table are derived from the single-laboratory validation study, and are only provided as examples for this draft method. The data will be updated with the interlaboratory study results in a subsequent revision.

21.0 Glossary

These definitions and purposes are specific to this method, but have been conformed to common usage to the extent possible.

21.1 Units of weight and measure and their abbreviations

21.1.1 Symbols

°C	degrees Celsius
Da	Dalton (equivalent to “amu” below)
µg	microgram
µL	microliter
µm	micrometer
<	less than
≤	less than or equal
>	greater than
≥	greater than or equal
%	percent
±	plus or minus

21.1.2 Alphabetical abbreviations

amu	atomic mass unit (equivalent to Dalton)
cm	centimeter
g	gram
h	hour
L	liter
M	molar
mg	milligram
min	minute
mL	milliliter
mm	millimeter
cm	centimeter
m/z	mass-to-charge ratio
ng	nanogram
Q1	quantitation ion
Q2	confirmation ion
rpm	revolutions per minute
v/v	percent volume per volume

21.2 Definitions and acronyms (in alphabetical order)

Analyte – A PFAS compound included in this method. The analytes are listed in Table 1.

Calibration standard (CS) – A solution prepared from a secondary standard and/or stock solutions and used to calibrate the response of the LC-MS/MS instrument.

Calibration verification standard (CV) – The mid-point calibration standard (CS-4) that is used to verify calibration. See Table 4.

CFR – Code of Federal Regulations

Compound - One of many variants or configurations of a common chemical structure. Individual compounds are identified by the number of carbon atoms and functional group attached at the end of the chain.

Class A glassware – Volumetric glassware that provides the highest accuracy. Class A volumetric glassware complies with the Class A tolerances defined in ASTM E694, must be permanently labeled as Class A, and is supplied with a serialized certificate of precision.

CWA – Clean Water Act

Extracted internal standard (EIS) quantification – The response of the target compound is compared to the response of the labeled analog of another compound in the same LOC.

LC – Liquid chromatograph or liquid chromatography

Internal standard – A labeled compound used as a reference for quantitation of other labeled compounds and for quantitation of native PFAS compounds other than the compound of which it is a labeled analog. See Internal standard quantitation.

Instrument sensitivity check – solution used to check the sensitivity of the instrument. The solution contains the native compounds at the concentration of the LOQ.

Internal standard quantitation – A means of determining the concentration of (1) a naturally occurring (native) compound by reference to a compound other than its labeled analog and (2) a labeled compound by reference to another labeled compound

IPR – Initial precision and recovery; four aliquots of a reference matrix spiked with the analytes of interest and labeled compounds and analyzed to establish the ability of the laboratory to generate acceptable precision and recovery. An IPR is performed prior to the first time this method is used and any time the method or instrumentation is modified.

Isotope dilution (ID) quantitation – A means of determining a naturally occurring (native) compound by reference to the same compound in which one or more atoms has been isotopically enriched. The labeled PFAS are spiked into each sample and allow identification and correction of the concentration of the native compounds in the analytical process.

Isotopically labeled compound – An analog of a target analyte in the method which has been synthesized with one or more atoms in the structure replaced by a stable (non-radioactive) isotope of that atom. Common stable isotopes used are ¹³C (Carbon-13) or Deuterium (D or ²H). These labeled compounds do not occur in nature, so they can be used for isotope dilution quantitation or other method-specific purposes.

Limit of Quantitation (LOQ) – The smallest concentration that produces a quantitative result with known and recorded precision and bias. The LOQ shall be set at or above the concentration of the lowest initial calibration standard (the lowest calibration standard must fall within the linear range).

Method blank – An aliquot of reagent water that is treated exactly as a sample including exposure to all glassware, equipment, solvents, reagents, internal standards, and labeled compounds that are used with samples. The method blank is used to determine if analytes or interferences are present in the laboratory environment, the reagents, or the apparatus.

Method Detection Limit (MDL) – The minimum measured concentration of a substance that can be reported with 99% confidence that the measured analyte concentration is distinguishable from method blank results (40 CFR 136, Appendix B).

MESA – Mining Enforcement and Safety Administration

Minimum level of quantitation (ML) – The lowest level at which the entire analytical system must give a recognizable signal and acceptable calibration point for the analyte. The ML represents the lowest concentration at which an analyte can be measured with a known level of confidence. It may be equivalent to the concentration of the lowest calibration standard, assuming that all method-specified sample weights, volumes, and cleanup procedures have been employed. Alternatively, the ML may be established by multiplying the MDL (pooled or unpooled, as appropriate) by 3.18 and rounding the result to the number nearest to 1, 2, or 5×10^n , where n is zero or an integer (see 68 FR 11770).

MS – Mass spectrometer or mass spectrometry

Matrix Spike/Matrix Spike Duplicate (MS/MSD) – Aliquots of field samples that have been fortified with a known concentration of target compounds, prior to sample preparation and extraction, and analyzed to measure the effect of matrix interferences. The use of MS/MSD samples is generally not required in isotope dilution methods because the labeled compounds added to every sample provide more performance data than spiking a single sample in each preparation batch.

Multiple reaction monitoring (MRM) – Also known as selected reaction monitoring (SRM). A type of mass spectrometry where a parent mass of the compound is fragmented through MS/MS and then specifically monitored for a single fragment ion.

Must – This action, activity, or procedural step is required.

NIOSH – The National Institute of Occupational Safety and Health

Non-extracted internal standard (NIS) – Labeled PFAS compounds spiked into the concentrated extract immediately prior to injection of an aliquot of the extract into the LC-MS/MS.

OPR – Ongoing precision and recovery standard (OPR); a method blank spiked with known quantities of analytes. The OPR is analyzed exactly like a sample. Its purpose is to assure that the results produced by the laboratory remain within the limits specified in this method for precision and recovery.

Precursor Ion – For the purpose of this method, the precursor ion is the deprotonated molecule ($[M-H]^-$) of the method analyte. In MS/MS, the precursor ion is mass selected and fragmented by collisionally activated dissociation to produce distinctive product ions of smaller m/z .

PFAS – Per- and Polyfluoroalkyl substances – A group of man-made fluorinated compounds that are hydrophobic and lipophobic, manufactured and used in a variety of industries globally. These compounds are persistent in the environment as well as in the human body. This method analyzes for the PFAS listed in Table 1.

Reagent water – Water demonstrated to be free from the analytes of interest and potentially interfering substances at the method detection limit for the analyte.

Relative standard deviation (RSD) – The standard deviation multiplied by 100 and divided by the mean. Also termed “coefficient of variation.”

Relative Standard Error (RSE) – The standard error of the mean divided by the mean and multiplied by 100.

RF – Response factor. See Section 10.3.3.2.

RR – Relative response. See Section 10.3.3.2.

RT – Retention time; the time it takes for an analyte or labeled compound to elute off the HPLC/UPLC column

Should – This action, activity, or procedural step is suggested but not required.

Signal-to-noise ratio (S/N) – The height of the signal as measured from the mean (average) of the noise to the peak maximum divided by the width of the noise.

SPE – Solid-phase extraction; a technique in which an analyte is extracted from an aqueous solution or a solid/tissue extract by passage over or through a material capable of reversibly adsorbing the analyte. Also termed liquid-solid extraction.

Stock solution – A solution containing an analyte that is prepared using a reference material traceable to EPA, NIST, or a source that will attest to the purity and authenticity of the reference material.

Appendix A - Sample Pre-screening Instructions

Samples that are known or suspected to contain high levels of analytes may be pre-screened using the following procedure. These are example procedures using smaller sample aliquots spiked with EIS and NIS and no clean up procedures. Other pre-screening procedures may be used.

Aqueous Samples

1. Weight out 10 (± 0.1) g of sample into a 50-mL centrifuge tube.
2. Add 50 μ L of EIS and NIS to the sample and vortex to mix.
3. Filter 1 mL of the sample through 0.2- μ m membrane filter into a microvial. Sample is ready for instrumental analysis.

Solid and Tissue Samples

1. Weigh 1.0 (± 0.1) g sample into 50-mL polypropylene centrifuge tubes.
2. Add 20 mL of 0.3% methanolic ammonium hydroxide (Section 7.1.7.1). Vortex and mix on a shaker table (or equivalent) for 10 min. Allow to settle and/or centrifuge to produce a clear extract.
3. Filter using a Single Step® filter vial:
 - a. Add 20 μ L of EIS to a clean Single Step® filter vial (chamber).
 - b. Add 400 μ L of clear extract from step 2 (e.g., by adding extract until it reaches the fill line), carefully vortex to mix.
 - c. Use filter/plunger part and filter.
4. Transfer 30 μ L of filtrate to a ~300- μ L polypropylene micro-vial and dilute to 300 μ L with 0.3% methanolic ammonium hydroxide (Section 7.1.7.1). Add NIS to the filtrate.
5. The extract is now a 10x dilution.
6. Sample is ready for instrumental analysis.

Calculate results using the equivalent sample weight computed as follows:

$$\text{Equivalent Weight} = \text{Sample weight (g)} \times \frac{0.4 \text{ mL}}{20 \text{ mL}}$$

Note that the EIS concentration in the diluted portion is 0.5x the level in the regular analysis of solid samples.

Appendix B - Aqueous Sample Subsampling Instructions

Warning: Because some target analytes may be stratified within the sample (e.g., AFFF-contaminated media, surfactants), or adhere to the walls of the sample container, subsampling may only be done on a project-specific basis. Subsampling has been shown to increase uncertainty in PFAS analysis, especially on foaming samples.

If a reduced sample size is required, transfer a weighed subsample using the following subsampling procedure to a 60-mL HDPE bottle and dilute to approximately 60 mL using reagent water. This container is now considered the “sample bottle.”

1. Gently invert sample 3-4 times being careful to avoid foam formation and subsample immediately (do not let stand).
2. If foam forms and more than 5 mL is required – pour sample, avoiding any foam.
3. If foaming forms and a volume less than 5 mL is required – pipette from ½ cm below the foam.
4. If no foam forms – pour or pipette based on volume required.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

Management Approval:

Jeffery Dunster Approved on 2/8/2024 7:21:22 AM
Kimberly Hazelwood Approved on 2/19/2024 8:46:17 AM
Rebecca Summerlot Approved on 2/8/2024 8:47:55 AM
Trisha Kelly Approved on 2/20/2024 11:22:42 AM
Thomas Widera Approved on 2/20/2024 11:39:23 AM

©2019-2023, Pace® Analytical Services, LLC

1.0 SCOPE AND APPLICATION

This standard operating procedure (SOP) describes the laboratory procedure for the determination of Per- and Polyfluoroalkyl Substances (PFAS) by isotope dilution anion exchange solid phase extraction and liquid chromatography/tandem mass spectrometry (LCMSMS). This SOP is applicable to drinking waters.

1.1 Target Analyte List and Limits of Quantitation (LOQ)

The target analytes and the normal LOQ that can be achieved with this procedure are provided in Table 1, Appendix A.

LOQ are established in accordance with Pace® policy and SOPs for method validation and for the determination of detection limits (DL) and quantitation limits (LOQ). DL and LOQ are routinely verified and updated when needed. Refer to ENV-SOP-ORB1-0124 for additional information on determination of MDL and LOQ. The current LOQ for each target analyte that can be determined by this SOP as of the effective date of this SOP is provided in Table 1, Appendix A. Current MDLs are located in the laboratory LIMS.

The reporting limit (RL) is the value to which analytes are reported as detected or not detected in the final report. When the RL is less than the lower limit of quantitation (LLOQ), all detects and non-detects at the RL are qualitative. The LLOQ is the lowest point of the calibration curve used for each target analyte.

DL, LOQ, and RL are always adjusted to account for actual amounts used and for dilution.

2.0 SUMMARY OF METHOD

A 250 mL water sample is fortified with isotope dilution standards consisting of isotopically labeled analogues of the target analytes. The sample is passed through a solid phase extraction (SPE) cartridge containing polystyrene divinylbenzene with a positively charged diamino ligand to extract the method analytes and isotopically labeled analogues. The compounds are eluted from the SPE cartridge with methanol containing ammonium hydroxide. The extract is concentrated to dryness with nitrogen in a heated water bath, and then reconstituted to a 1 mL final volume with 20% (vol/vol) water in methanol, which includes isotopically labelled performance standards. The isotopically labelled performance standards function as internal standards for the isotope dilution analogues. 1 µL of the fortified aliquot is injected on an LC equipped with a C18 column that is coupled to an MS/MS detector. The analytes are separated and identified by comparing the acquired mass spectra and retention times to the reference spectra and retention times for calibration standards acquired under identical LC/MS/MS conditions. The concentration of each analyte is determined by using the internal standard isotope dilution technique.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

3.0 INTERFERENCES

PFAS standards, extracts and samples should not come in contact with any glass containers or pipettes as these analytes can potentially adsorb to glass surfaces. PFAS analyte, internal standard (IS) and surrogate standards commercially purchased in glass ampoules are acceptable and subsequent transfers may be performed using glass syringes and pipets. Following extraction, the eluate must be collected in a polypropylene tube prior to concentration to dryness.

Method interferences may be caused by contaminants in solvents, reagents (including reagent water), sample containers and caps, and other sample processing equipment that lead to discrete artifacts and/or elevated baselines in the chromatograms. The method analytes in this method can also be found in many common laboratory supplies and equipment, such as PTFE (polytetrafluoroethylene) products, LC solvent lines, methanol, aluminum foil, SPE sample transfer lines, etc.

Matrix interferences may be caused by contaminants that are co-extracted from the sample. The extent of matrix interferences will vary considerably from source to source, depending upon the nature of the water. Humic and/or fulvic material can be co-extracted during SPE and high levels can cause enhancement and/or suppression in the electrospray ionization source or low recoveries on the SPE sorbent.

Solid phase extraction cartridges may be a source of interferences. The analysis of LRBs provides important information regarding the presence or absence of such interferences. Each brand and lot of SPE devices must be monitored to ensure that contamination does not preclude analyte identification and quantitation. SPE cartridges should be sealed while in storage to prevent ambient contamination of the SPE sorbent.

4.0 DEFINITIONS

- **Isotope Dilution Technique:** an analytical technique for measuring analyte concentration using the ratio of the peak area of the native analyte to that of an isotopically labeled analogue, added to the original sample in a known amount and carried through the entire analytical procedure.
- **Isotope Performance Standard (Internal/Instrument Standard [IPS]):** Quality control compounds that are added to all standard solutions and extracts in a known amount and used to measure the relative response of the isotopically labelled analogues that are components of the same solution. For this method, the isotope performance standards are three isotopically labeled analogues of the method analytes. The isotope performance standards are indicators of instrument performance and are used to calculate the recovery of the isotope dilution analogues through the extraction procedure. In this method, the isotope performance standards are not used in the calculation of the recovery of the native analytes
- **Isotope Dilution Analogues (IDA)/ (SUR):** A pure chemical which chemically resembles method analytes and is extremely unlikely to be found in any sample. This chemical is added to a sample aliquot (field and QC) in known amount(s) before the extraction and analysis processes. The purpose of the IDA/SUR is to monitor method performance from extraction to final chromatographic measurement. For Method 533, the IDA/SUR is used

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

as an isotope dilution standard for measuring the relative response and quantification of other method analytes.

- **Collisionally Activated Dissociation (CAD):** The process of converting the precursor ion's translational energy into internal energy by collisions with neutral gas molecules to bring about dissociation into product ions
- **Precursor Ion:** The gas-phase species corresponding to the method analyte that is produced in the electrospray ionization interface. During tandem mass spectrometry, or MS/MS, the precursor ion is mass selected and fragmented by collisionally activated dissociation to produce distinctive product ions of smaller mass to charge (m/z) ratio. For this method, the precursor ion is usually the deprotonated molecule ($[M - H]^-$) of the method analyte.
- **Product Ion:** A product ion is one of the fragment ions produced in MS/MS by collisionally activated dissociation of the precursor ion.

5.0 HEALTH AND SAFETY

Contact your supervisor or local safety coordinator with questions or concerns regarding safety protocol or safe handling procedures for this procedure.

The following sections provide general health and safety information about chemicals and materials that may be present in the laboratory.

- The toxicity or carcinogenicity of each chemical material used in the laboratory has not been fully established. Each chemical should be regarded as a potential health hazard and exposure to these compounds should be as low as reasonably achievable.
- The laboratory maintains documentation of hazard assessments and OSHA regulations regarding the safe handling of the chemicals specified in each method. Safety data sheets for all hazardous chemicals are available to all personnel. Employees must abide by the health, safety and environmental (EHS) policies and procedures specified in this SOP and in the Pace® Chemical Hygiene / Safety Manual (COR-MAN-0001)
- Personal protective equipment (PPE) such as safety glasses, gloves, and a laboratory coat must be worn in designated areas and while handling samples and chemical materials to protect against physical contact with samples that contain potentially hazardous chemicals and exposure to chemical materials used in the procedure.
- Concentrated corrosives present additional hazards and are damaging to skin and mucus membranes. For procedures that require use of acids, use acids in a fume hood whenever possible with PPE designed for handling these materials. If eye or skin contact occurs, flush with large volumes of water. When working with acids, always add acid to water to prevent violent reactions. For procedures that emit large volumes of solvents (evaporation/concentration processes), these activities must be performed in a fume hood or apparatus that reduces exposure.

6.0 SAMPLE COLLECTION, PRESERVATION, HOLDING TIME, AND STORAGE

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

PAS offers sample collection services for this test method. Refer to SOP ENV-SOP-ORB1-0108, *Bottle and Water Testing* for the procedure used when sample collection is performed by the location referenced in the scope and application of this SOP. If sample collection is performed by a different PAS facility, contact a PAS Project Manager (PM) to obtain the SOP used by that location.

PAS provides containers for the collection of samples upon client request. The routine container type and size provided in provided in the following table along with requirements for sample size, preservation and holding time. Refer to laboratory SOP ENV-SOP-ORB1-0123, *Bottle Preparation*, for procedures related to preparation of bottle kits.

General Requirements

Matrix	Routine Container	Minimum Sample Amount ¹	Preservation	Holding Time
Aqueous	250 mL polypropylene or HDPE bottle with polypropylene or HDPE screw cap	250 mL	Thermal: <10 °C Chemical: 1g/L ammonium acetate	Collection to Prep: 28 days Prep to Analysis: 28 days

¹Minimum amount needed for each discrete analysis.

Field / Matrix QC

Field Reagent Blank (FRB/FB)	MS/MSD	Field Duplicate
One per sampling site/sampling event	MS/MSD per extraction batch	FD per extraction batch unless MSD is prepped

Field Reagent Blanks: The laboratory will provide a bottle of reagent water without preservative and an empty container with preservative. At the sampling site, open the bottle containing the reagent water and pour into the empty Field Blank bottle containing preservative. Seal and label this bottle as the FRB with the data, time, and location of the site.

Samples must be shipped on ice and received with ice. Samples are valid if any ice remains in the cooler when it is received at the laboratory or bottles are received within 2 days of collection and below <10 °C. Thermal preservation is checked and recorded on receipt in the laboratory in accordance with laboratory SOP ENV-SOP-ORB1-0120_ *Sample Management*. Chemical preservation is checked and recorded at time of receipt or prior to sample preparation.

After receipt, samples are stored at ≤6°C until sample preparation. Samples must not be frozen. Prepared samples (extracts, digestates, distillates, other) are stored at room temperature until sample analysis.

7.0 EQUIPMENT AND SUPPLIES

Due to the possibility of adsorption of analytes onto glass, HDPE containers are used for all standard, sample and extraction preparations. Any time a new lot of SPE cartridges/tubes, solvents, cryovials, or autosampler vials are used, it must be demonstrated to be free of contamination through analysis of the method blanks.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

- **Polypropylene bottles:** 4 mL narrow-mouth polypropylene or HDPE bottles (Thermo Cat# 2006-9125 or equivalent).
- **Sample containers:** 250 mL polypropylene or HDPE bottles fitted with polypropylene or HDPE screw caps. Sample bottles must be discarded after use.
- **Centrifuge tubes:** 15 mL conical polypropylene tubes with polypropylene screw caps for storing standard solutions and for collection of the extracts (Sarstedt Cat# 62.554.205 or equivalent)
- **Autosampler vials:** Polypropylene 0.3 mL autosampler vials (Thermo Cat# C4010-11 or equivalent) with polypropylene caps (Thermo Cat#: 5330-10 or equivalent). Polypropylene vials and caps are necessary to prevent contamination of the sample from PTFE coated septa. However, polypropylene caps do not reseal, so evaporation occurs after injection. Thus, multiple injections from the same vial are not advisable.
- **Polypropylene graduated cylinders:** Suggested sizes include 25, 50, 100 and 1000-mL cylinders
- **Microsyringes:** Suggested sizes include 5, 10, 50, 100, 250, 500, and 1000 µL syringes.
- **Micropipettes:** Suggested size up to 1000 µL
- **Plastic pipettes:** Polypropylene or polyethylene disposable pipettes (Fisher Cat# 13-711-7M or equivalent).
- **Top loading balance:** Capable of weighing to the nearest 0.01 g
- **SPE Cartridges:** Strata-XL-AW 100 µm Polymeric Weak Anion (0.5 g, 6mL) (Phenomenex Part # 8B-S051-HCH) or equivalent. Cartridges may not be reconditioned for subsequent analyses.
- **pH meter:** capable of measuring the pH of the aqueous sample prior to anion exchange and verify the pH of the phosphate buffer (Fisher cat. Number 13-636-AB250) or equivalent.
- **Free chlorine test:** DPD Powder (HF Scientific 10504C or equivalent) or Free Chlorine Strips (HF Scientific Catalog # 09940 or equivalent)
- **Vacuum extraction manifold:**
 - Semi-Automated sample preparation system designed for use with SPE cartridges. Care must be taken with automated SPE systems to ensure the PTFE commonly used in these systems does not contribute to unacceptable analyte concentrations in the LRB. (Fluid Management part # EZPFC-12 - 12 Position EZPFC System for water & wastewater analysis, or equivalent)
 - Automated Sample preparation system care must be taken with automated SPE systems to ensure the PTFE commonly used in these systems does not contribute to unacceptable analyte concentrations in the LRB. (Promochrom model SPE-03 Gen 4, or equivalent)

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

- **Vacuum pump:** Sufficient capacity to maintain a vacuum of approximately 15 to 20 inches of mercury for extraction cartridges.
- **Extract concentration system:** Extracts are concentrated by evaporation with nitrogen using a water bath set to 55-60°C (Meyer NEvap, Model 111, Organomation Associates, Inc. or equivalent).
- **Liquid Chromatography (LC)/Tandem Mass Spectrometer (MS/MS):** PFAS can build up in the PTFE solvent transfer lines and PTFE solvent frits. To prevent long delays in purging high levels of PFAS from the LC solvent lines, PEEK tubing and stainless-steel frits are used.
 - LC System – Agilent Model 1290 Infinity II, with Degasser, consisting of (Binary Pump (G7120A), Autosampler (G7167B), Thermostated Column Compartment (G7116B); or equivalent.
 - Tandem Mass Spectrometer (MS/MS) – In negative ion electrospray ionization (ESI) mode. A minimum of 10 scans across the chromatographic peak is required to ensure adequate precision. (Agilent 6495C MS/MS or equivalent)
 - Analytical Column – Agilent Zorbax Eclipse Plus C-18 1.8µm LC column 50 x 2.1mm, (part# 959757 -902); or equivalent
 - Delay Column – Agilent Zorbax Eclipse C-18 Plus 4.6 x 3.5mm (part# 959353-902); or equivalent.
 - In-line Filter – Agilent 0.3 µm (part# 5067-6189); or equivalent

8.0 REAGENTS AND STANDARDS

If not specially detailed below, refer to Pace® Analytical Services SOP ENV-SOP-ORB1-0107_Standard-Reagent Preparation & Traceability, or equivalent replacement, for information regarding standard and reagent preparation, tracking, expiration, and traceability.

8.1 Reagents

Reagent grade or better chemicals should be used. Unless otherwise indicated, it is intended that ACS reagents be used, where such specifications are available. Other grades may be used, provided it is first determined that the reagent is of sufficiently high purity to permit its use without lessening the quality of the analysis.

- **Reagent Water** – Optima LC/MS water (Fisher part# W6-4 or equivalent). The reagent water should not contain any measurable quantities of any method analytes or interfering compounds greater than 1/3 the LOQ for each analyte of interest.
- **Methanol** (MeOH, CH₃OH, CAS#: 67-56-1) – HPLC grade or equivalent, commercially available (Fisher part# A452-4 or equivalent). Expiration per manufacturer.
- **80:20 Methanol in Reagent Water** - Mix 200 mL of reagent water with 800 mL of methanol. Expires 1 year from preparation.
- **Acetic acid (glacial)** – ACS grade or equivalent, commercially available. Expiration per manufacturer.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

- **Ammonium Acetate** ($\text{NH}_4\text{C}_2\text{H}_3\text{O}_2$, CAS#: 631-61-8) – LC/MS grade, commercially available (Fisher part# A114-50 or equivalent). Expiration per manufacturer.
- **1 g/L Ammonium Acetate** – To prepare 1 L, add 1.0 g of ammonium acetate (MW = 77.08 g/mol) to 1 L of reagent water. Expires 1 week from preparation.
- **Sodium Phosphate Monobasic Monohydrate** - ACS grade ($\text{H}_4\text{NaO}_5\text{P}$, CAS # 10049-21-5) commercially available (Fisher part # S369-500 or equivalent). Expiration per manufacturer.
- **Sodium Phosphate Dibasic Anhydrous**, ACS grade ($\text{HNa}_2\text{O}_4\text{P}$, CAS # 7558-79-4) commercially available (Fisher part # S374-500 or equivalent). Expiration per manufacturer.
- **0.1M Phosphate Buffer (pH 7)** – Mix 500 mL of 0.1M sodium phosphate dibasic with approximately 275 mL of 0.1M sodium phosphate monobasic. Or, add 7.15g of sodium phosphate dibasic heptahydrate and 3.83g sodium phosphate monobasic monohydrate to 775mL of reagent water. Verify that the solution pH is approximately 7.0, document in the comments section of the standard log. Expires 1 month from preparation.
- **Ammonium Hydroxide** (NH_4OH , CAS #1336-21-6) – ACS grade, approximately 56.6% in water as ammonium hydroxide (w/w), approximately 28% in water as ammonia, approximately 14.5 N. Commercially available (Fisher part# A669 or equivalent). Expiration per manufacturer.
- **Ammonium Hydroxide in Methanol** (Amm-MeOH, Elution Solvent) – Add 10 mL NH_4OH (Ammonium Hydroxide) to 500 mL of methanol. Invert to mix. Prepare fresh on each day of extraction.
- **5M Ammonium Acetate aq. Solution** ($\text{NH}_4\text{C}_2\text{H}_4\text{O}_2$, CAS#: 631-61-8) – commercially available (Alfa Aesar part # J60688, or equivalent). Expiration per manufacturer.
- **5mM Ammonium Acetate Aqueous Mobile Phase** – To prepare 1 L, add 1.0 mL 5M ammonium acetate to a 1 L volumetric flask and bring to volume with reagent water. This solution is prone to volatility losses. Expires 2 days from preparation.
- **5mM Ammonium Acetate 95%Methanol/5% H₂O Organic Mobile Phase** - To prepare 1 L, add 0.5 mL of 5M ammonium Acetate to 50 mL of reagent water in a 1 L volumetric flask. Bring to volume with methanol. This solution is prone to volatility losses. Expires 2 days from preparation.
- **Nitrogen, Ultra-high Purity Grade** – Nitrogen aids in aerosol generation of the ESI liquid spray and is used as collision gas in some MS/MS instruments. The nitrogen used is also for extract concentration.

8.2 Standards

- **Standard Solutions** – When a compound purity is assayed to be 96% or greater (standards purchased from Wellington are >98%), the weight can be used without correction to calculate the concentration of the stock standard. PFAS analyte, IS, and SUR standards commercially purchased in glass ampoules are acceptable; however, all subsequent transfers or dilutions performed by the analyst must be prepared and stored in HDPE containers. All standards must be stored under basic conditions to prevent esterification. If standards are not received under basic conditions this can be accomplished by the addition of sodium hydroxide

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

(approximately 4 mole equivalents) when diluted in methanol. Target analyte standards contain branched isomers.

- **Correction for analytes obtained in the salt form** – PFHxS, PFOS and other sulfonic acids are not available as the acid form, but rather as their corresponding salts, such as Na⁺ and K⁺. These salts are acceptable for use as stock standards as long as the weight is corrected for the salt content according to the equation below.

$$\text{mass}(\text{acid form}) = \text{mass}(\text{salt form}) \times \frac{MW_{\text{acid}}}{MW_{\text{salt}}}$$

Where:

MW_{acid} = the molecular weight of PFAA

MW_{salt} = the molecular weight of purchased salt

- **Isotope Performance Standards (IPS, 533-IS) Standards** – The standard solution is commercially available (Wellington Labs catalog # EPA-533IS, or equivalent). Expiration per manufacturer. Store at ≤6°C under basic conditions. The internal standards used are comprised of the following compounds:
 - Perfluoro-n-[1,2-¹³C₂] octanoic acid (13C₂-PFOA)
 - Sodium perfluoro-1-[1,2,3,4-¹³C₄]octanesulfonate (13C₄-PFOS)
 - Perfluoro-n-[2,3,4-¹³C₃] butanoic acid (13C₃-PFBA)
- **Isotope Dilution Analogue (IDA, 533-SUR) Standard Solution** – The SUR standard solution is commercially available (Wellington Labs catalog #EPA-533 ES, or equivalent). Expiration per manufacturer. Store at ≤6°C under basic conditions.
- **Analyte Primary Dilution Standards (533-SPK, PDS)** – Analyte standards are commercially available (Wellington Labs catalog #EPA-533PAR, or equivalent). Expiration per manufacturer. Stored at ≤6°C under basic conditions.
- **Initial Calibration Standards (ICAL)** – According to the table below, prepare calibration standards. These standards are prepared using the 80:20 MeOH/H₂O mixture. The ICAL standards expire 2 months from preparation when stored at room temperature, or 12 months from preparation when stored at ≤6°C.

	Analyte PDS (μL)	IDA (SUR) (μL)	IPS (IS) (μL)	Final Volume (mL)	PFAS Conc. (ng/mL)	IDA/SUR Conc. (ng/mL)	IPS/IS Conc. (ng/mL)
533-CAL1	1.0	40	20	2.0	0.25	500/2000	1000/3000
533-CAL2	2.0	40	20	2.0	0.5	500/2000	1000/3000
533-CAL3	4.0	40	20	2.0	1.0	500/2000	1000/3000
533-CAL4	8.0	40	20	2.0	2.0	500/2000	1000/3000
533-CAL5	20	40	20	2.0	5.0	500/2000	1000/3000
533-CAL6	40	40	20	2.0	10.0	500/2000	1000/3000
533-CAL7	80	40	20	2.0	20.0	500/2000	1000/3000
533-CAL8	100	40	20	2.0	25.0	500/2000	1000/3000

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

- **ICV Mix** - Second source standard containing required target analytes at 2.0 ng/mL. (Wellington Labs catalog # PFAC30PAR and PFACMXG, or equivalent). This standard is prepared in 80:20 MeOH/H₂O and expires in 2 months when stored at room temperature, or 12 months when stored at ≤6°C.

Standard Name	Stock Concentration (ng/mL)	Standard Volume (μL)	Final Volume (mL)	Final Conc. (ng/mL)
ICV Mix PFAC30PAR	1000	4.0	2.0	2.0
ICV Mix PFACMXG	2000	2.0		2.0
IDA/SUR	500/2000	40		10/40
IPS/IS	1000/3000	20		10/30

- **Method Blank (MB)** – Spike 250 mL reagent water with 20 μL of the IDA/SUR Mix. Mix well, extract as normal alongside client samples. See Appendix B for acceptance criteria.
- **Laboratory Control Spike (LCS)/Laboratory Fortified Blank (LFB)** - An LFB is required with each Extraction Batch. The concentration of the LFB must be rotated between low, medium, and high concentrations from batch to batch. Fortify the low concentration LFB near the MRL. The high concentration LFB must be near the high end of the calibration range. See Appendix B for acceptance criteria.

9.0 PROCEDURE

9.1 Equipment Preparation

9.1.1 Support Equipment

Refer to Pace® Analytical Services SOP ENV-SOP-ORB1-0105_*Support Equipment* for additional information on calibration requirements for support equipment that may be used in this procedure.

9.1.2 Instrument

9.1.2.1 Routine Instrument Operating Conditions

Refer to Appendix C.

9.2 Initial Calibration (ICAL)

Refer to policy ENV-POL-CORQ-0005 for general laboratory calibration policies and procedures.

9.2.1 Calibration Design

9.2.1.1 Target Analyte Calibration

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

A minimum of five ICAL standards for linear curves or six ICAL standards for quadratic curves are prepared as described above. The lowest concentration ICAL standard must be at or below the LOQ/MRL, which may depend on system sensitivity. It is recommended that at least four of the ICAL standards are at a concentration greater than or equal to the LOQ/MRL.

The LC/MS/MS system is calibrated using the IS technique. Generate a linear or quadratic regression calibration curve for each of the analytes. This curve will be forced through zero. Forcing through zero allows for a better estimate of the background levels of method analytes. Weighting (typically $1/x$ or $1/x^2$) is allowed for linear and nonlinear regressions.

9.2.1.2 Isotope Dilution Analogue (IDA/SUR) Calibration

The isotope dilution analogues are quantified using the internal standard calibration technique. Because the IDA are added at a single concentration level to the calibration standards, calibrate for each using an average response factor.

9.2.1.3 Isotope Performance Standard (IPS/IS) Calibration

Because the IPS are added at a single concentration level to the calibration standards, calibrate for each using an average response factor.

9.2.2 Calibration Sequence

A calibration event will consist of the ICAL standards (lowest to highest), followed by an instrument blank to check for carryover following the highest calibration standard. An ICV Standard is analyzed following the calibration set and carryover blank.

9.2.3 ICAL Evaluation

9.2.3.1 Curve Fit

Removal or replacement of calibration points must be performed in accordance with ENV-POL-CORQ-0005.

9.2.3.2 Calibration Acceptance Criteria

- a) The response of the calibration standards must be evaluated against the calibration curve equation by calculating the concentration of each analyte as an unknown against its regression equation. Analytes fortified at or below the LOQ/MRL must be within 50-150% of the true value. Analytes fortified at all other levels must be within 70-130% of their true value. If these criteria cannot be met, the analyst could have difficulty meeting ongoing QC criteria. In this case, corrective action is recommended such as reanalyzing the calibration standards, restricting the range of calibration, or performing instrument maintenance. If the cause for failure to meet the criteria is due to contamination or standard degradation, prepare fresh calibration standards and repeat the initial calibration.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

9.2.3.3 Initial Calibration Verification (ICV/QCS)

The calculated amount for each analyte must meet the accuracy criteria listed in Appendix B. If measured analyte concentrations are not of acceptable accuracy, correct the problem and rerun the ICV. If the problem persists, repeat the ICAL.

9.2.4 Continuing Calibration Verification (CCV)

9.2.4.1 Frequency - Analyze CCV standards at the beginning of each Analysis Batch, after every tenth field sample, and at the end of the Analysis Batch. The beginning CCV for each Analysis Batch must be at, or below, the MRL for each analyte. This CCV verifies instrument sensitivity prior to the analysis of samples. If standards have been prepared such that all low calibration levels are not in the same solution, it may be necessary to analyze two standards to meet this requirement. Alternatively, the nominal analyte concentrations in the analyte PDS may be customized to meet these criteria. Alternate subsequent CCVs between the mid and high calibration levels.

9.2.4.2 IPS/IS Acceptance Criteria – The absolute area of the quantitation ion for each of the three isotope performance standards must be within 50–150% of the average area measured during the initial calibration. If these limits are exceeded, corrective action is necessary.

9.2.4.3 IDA/SUR Acceptance Criteria - Using the average response factor determined during the initial calibration and the internal standard calibration technique, calculate the percent recovery of each isotope dilution analogue in the CCV. The recovery for each analogue must be within a range of 70–130%. If these limits are exceeded, corrective action is necessary.

9.2.4.4 Target Analyte Acceptance Criteria - Calculate the concentration of each method analyte in the CCV. Each analyte fortified at a level less than or equal to the MRL must be within 50–150% of the true value. The concentration of the analytes in CCVs fortified at all other levels must be within 70–130%. If these limits are exceeded, then all data for the failed analytes must be considered invalid. Any field samples analyzed since the last acceptable CCV that are still within holding time must be reanalyzed after an acceptable calibration has been restored.

9.3 Extract Preparation

This method may be performed manually or by using an automatic sample extraction procedure. Pace® Analytical Services, LLC. – Ormond Beach has the ability to perform the extraction using either the manual or automated procedures outlined below. When the automated procedure is used, the method blank (LRB/MB) will be rotated among the ports to ensure that all the valves and tubing meet the method blank requirements.

9.3.1 Homogenization and Subsampling

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

Some of the PFAS adsorb to surfaces, including polypropylene. During extraction, sample bottles must be rinsed therefore subsampling is not permitted. The entire volume of each sample must be extracted. Duplicate samples cannot be combined to meet volume requirements.

9.3.2 Sample Volume

Determine sample volume. An indirect measurement may be done by weighing the sample and bottle to the nearest 0.01 gram. After extraction, proceed to Section 9.3.10 to complete the volume measurement. Some of the PFAS adsorb to surfaces, thus the sample may not be transferred to a graduated cylinder for volume measurement.

Batch QC is made by adding 250mL of LCMS grade water to pre-preserved sample bottles. Measure the 250mL volume using a PPE graduated cylinder (that has been verified) and pour into the pre-preserved bottles. Shake until preservative is completely dissolved.

9.3.3 Verify Sample pH

Verify that the sample containing 1 g/L ammonium acetate has a pH between 6.0 and 8.0. Acetic acid may be added as needed to reduce the pH.

9.3.4 Fortify QC Samples

Fortify LCS/LFB, SRM/Low-LFB, MS, and MSD, with an appropriate volume of analyte PDS. Rotate the LCS/LFB concentration between mid and high-level spikes with each batch. Cap and invert each sample several times to mix.

9.3.4.1 For UCMR work, only a Low-LFB spiked at the MRL is required. Rotate the concentration of the MS/MSD spikes with each batch between low (at MRL) and mid-level concentrations.

9.3.5 Addition of Isotope Dilution Analogues

Add 20 µL of the 533-SUR to each sample and QC, then cap and invert to mix.

9.3.6 Extraction Procedure for Semiautomatic Extraction System

9.3.6.1 *Cartridge Cleaning and Conditioning:* Do NOT allow cartridge packing material to go dry during any of the conditioning steps. Rinse each cartridge with 5 mL of methanol. Drain down to a layer of solvent above the packaging material. Repeat the methanol rinse again with the same volume (for a total of 10 mLs), do not allow the cartridge packing material to go dry. Next, rinse each cartridge with 5 mL of aqueous 0.1M phosphate buffer, without allowing the buffer to drop below the top edge of the packing. Repeat the rinse again with another 5mL aliquot of aqueous 0.1M phosphate buffer (for a total of 10 mLs), do not allow the cartridge packing material to go dry. If the cartridge goes dry during the conditioning phase, the conditioning must be started over. Add 2-3 mL of aqueous 0.1M phosphate buffer to each cartridge and then fill cartridge using LCMS water. Attach the sample transfer tubes, turn on the vacuum, and begin transferring sample to the cartridge.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

9.3.6.2 *Sample Extraction:* Adjust the vacuum so that the approximate flow rate is 5 mL/min, this flow rate is a slow dropwise addition. Do not allow the cartridge to go dry before all the sample has passed through.

9.3.6.3 *Sample Bottle and Cartridge Rinse:* After the entire sample has passed through the cartridge, rinse the sample bottles with a 10 mL aliquot of 1g/L ammonium acetate in reagent water and draw through the sample transfer tubes and the cartridges. Add 1 mL of methanol to the sample bottle and draw through the sample tube and SPE cartridge. This step is designed to remove most of the water from the cartridge, resulting in the reduction of the salt and water present in the eluate. The methanol rinse may also reduce interferences by removing weakly retained organic material prior to elution.

9.3.6.4 *Cartridge Drying:* Draw air or nitrogen through the cartridge for 5 min at high vacuum (15-20 in. Hg).

9.3.6.5 *Sample Bottle and Cartridge Elution:* Turn off and release the vacuum. Lift the extraction manifold top and insert a rack with collection tubes into the extraction tank to collect the extracts as they are eluted from the cartridges. Rinse the sample bottles with 5 mL of the elution solvent (ammonium hydroxide in methanol) and elute the analytes from the cartridges by pulling the 5 mL of the elution solvent through the sample transfer tubes and the cartridges. Use a low vacuum such that the solvent exits the cartridge in a slow dropwise fashion. Repeat sample bottle rinse and cartridge elution with a second 5 mL aliquot of the elution solvent.

9.3.7 Extraction Procedure for Automated Extraction System

Once all bottles have been prepared for extraction, the inline filters and cleaning bottles are put on the system. Once this is finished the “clean system” step is selected on the automatic extraction unit. The cleaning bottles are then taken off and the sample bottles and QC bottles are placed on the units. Once finished the 533 method is selected on the units (example method Appendix D).

Note: Any troubleshooting for these units are done by referring to the Manual located at the instrument. If this does not solve the issue Promochrom is contacted.

9.3.8 Extract Concentration

Concentrate the extract to dryness under a gentle stream of nitrogen, or equivalent, in a heated water bath (55–60 °C). Reconstitute the extract with 1.0 mL of 20% reagent water in methanol (v/v). Add 10 µL of the isotope performance standards (533-IS) to the extract and vortex.

9.3.9 Extract Transfer and Storage

Transfer the final extract to a polypropylene cryovial. Store extracts at room temperature. A small aliquot can be removed to an autosampler vial for analysis. Recap vials as soon as possible after injection to prevent evaporation losses; the polypropylene caps do not reseal after puncture.

9.3.10 Sample Volume Determination

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

Weigh the empty bottle to the nearest 0.01 gram and subtract this value from the weight recorded prior to extraction. Assume a sample density of 1.0 g/mL. Record the sample volumes for use in the final calculations of analyte concentrations.

9.4 Analysis

9.4.1 Column Flush

Each day of analysis, the column must be thoroughly flushed with 100% MeOH for at least 20 minutes to clear any accumulated impurities and interferents from the sample pathway and equilibrate the system. Ensure that pressure is stable.

9.4.2 Analytical Sequence

Following the daily column flush, analyze a low-level CCV at the MRL. Each analysis batch of no more than 20 field samples must begin with a low-level CCV. This CCV must meet the accuracy criteria in Appendix B for target analytes, IPS, and IDAs. When a passing CCV sample has been evaluated, an instrument blank will be analyzed to demonstrate the absence of system contamination. After system contamination is determined to be acceptable (no target analyte concentrations $>1/3$ MRL), samples may be analyzed. Analyze a mid- or high- level CCV after every 10 field samples and at the end of each analysis batch. The final CCV for the analysis batch must be a mid- or high- level CCV. The acquisition time start time of the final CCV must be within 24 hours of the acquisition start time of the low-level CCV at the beginning of the analysis batch.

		Column Flush
		ICAL (as needed)
		ICV/QCS (as applicable)
	24 Hours	CCV-Low
		Analysis Batch 1 - QC
		Analysis Batch 1 - 10 samples
		CCV-Mid
		Analysis Batch 1 - 10 samples
		CCV-High
	24 Hours	CCV-Low
		Analysis Batch 2 - QC
		Analysis Batch 2 - 10 samples
		CCV-Mid
		Analysis Batch 2 - 10 samples
		CCV-High

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

10.0 DATA ANALYSIS AND CALCULATIONS

10.1 Qualitative Identification

At the conclusion of data acquisition, use the same software settings established during the calibration procedure to identify analyte peaks in the predetermined retention time windows. Confirm the identity of each analyte by comparison of its retention time with that of the corresponding analyte peak in an initial calibration standard or CCV. Proceed with quantitation based on the type of standard available for each method analyte.

If standards containing the branched and linear isomers cannot be purchased, only the linear isomer can be identified and quantitated in field and QC samples because the retention times of the branched isomers cannot be confirmed.

For PFHxS and PFOS, all the chromatographic peaks representing the branched and linear isomers must be integrated and the areas summed. Chromatographic peaks in all field and QC samples must be integrated in the same manner as the calibration standards for analytes containing branched and linear isomers. When available, a technical grade standard must be analyzed at a mid to high calibration level. This must be used to identify and establish the expected retention time. These identification checks must be repeated any time chromatographic changes occur that alter analyte retention times.

For PFOA, identify the branched and linear isomers by analyzing a technical-grade standard which includes both branch and linear isomers at a mid to high level concentration. Ensure all isomers elute within the same acquisition segment. Quantitate field samples and MS/MSD by integrating the total response, accounting for peaks that are identified as linear and branched isomers. Quantitate based on the initial calibration using the quantitative PFOA standard that contains only the linear isomer. These identification checks must be repeated any time chromatographic changes occur that alter analyte retention times.

10.1.1 Manual Integration

Manual change to automated integration is called manual integration. Manual integration is sometimes necessary to correct inaccurate automated integrations but must never be used to meet QC criteria or to substitute for proper instrument maintenance and/or method set-up. To assure that all manual integrations are performed consistently and are ethically justified, all manual integrations must be performed, reviewed, and recorded in accordance with corporate SOP ENV-SOP-CORQ-0006, *Manual Integration*.

10.2 Quantitative Identification

10.2.1 Calculate analyte concentrations using the multipoint calibration and the measured sample volume. Report only those values that fall between the MRL and the highest calibration standard.

10.2.2 The analyst must monitor the peak areas of the isotope performance standards (IPS/IS) in all injections of the Analysis Batch. The isotope performance standard responses (as indicated by peak area) in any chromatographic run must be within 50–150% of the average area measured during the initial calibration.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

10.2.3 Calculate the concentration of each isotope dilution analogue (IDA/SUR) using the multipoint calibration and the measured sample volume. Verify that the percent recovery is within 50–200% of the true value.

10.2.3.1 For UCMR work, the IDA must recover within 70-130% for calibration and CCV standards.

10.2.4 Sample results must not exceed the established calibration range. If an analyte exceeds the upper limit of the calibration curve, a field duplicate of the sample must be extracted at a dilution. Analytical dilutions cannot be performed for this method. Dilute an aliquot of the field duplicate with reagent water to a final volume of 250 mL and preserve with 1 g/L of ammonium acetate. This is the only circumstance when subsampling is allowed and must be noted as a dilution on the final report. Report all analytes that are within the calibration range from the undiluted sample.

10.2.4.1 After analysis of a sample in which the method analyte concentration exceeds the calibration range, one or more LRBs must be analyzed (to detect potential carryover) until the system meets the LRB acceptance criteria.

10.3 Calculations

See the *Laboratory Calculations* SOP ENV-SOP-ORB1-0118 for common calculations.

The linear regression curve is expressed as below: $y = ax + b$

Where a is the slope and b is the y-intercept.

When forced through 0, $b=0$, $y = AS/ASUR$, $x = CS/CSUR$

AS is peak response of target analyte in calibration standards

SUR is peak response of surrogate standard (SUR) in calibration standards

CS is concentration of target analyte in calibration standards

SUR is concentration of surrogate standard (SUR) in calibration standards

11.0 QUALITY CONTROL AND METHOD PERFORMANCE

11.1 Quality Control

Refer to Appendix B for Quality Control frequency, acceptance criteria and corrective action.

11.2 Method Performance

11.2.1 Method Validation

Refer to Pace® ENV corporate SOP ENV-SOP-CORQ-0011 Method Validation and Instrument Verification for these procedures. These steps must be performed initially and for any method modification.

11.2.1.1 Method Detection Limit (MDL) Study

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

Refer to ENV-SOP-ORB1-0124_Determination of LOD & LOQ for additional information on determination of MDL.

11.2.1.2 Minimum Reporting Level (MRL) Confirmation

Establish a target concentration for the MRL based on the intended use of the method. If there is a programmatic MRL requirement, the laboratory MRL must be set at or below this level. In doing so, one should consider that establishing the MRL concentration too low may cause repeated failure of ongoing QC requirements. Perform initial calibration following the procedures in Section 9.2. The lowest calibration standard used to establish the initial calibration (as well as the low-level CCV) must be at, or below, the MRL. Confirm the laboratory's ability to meet the MRL following the procedure outlined below.

- a) **Prepare and Analyze MRL Samples** – Fortify, extract, and analyze seven replicate spiked samples at, or below, the proposed MRL concentration.
- b) **Calculate MRL Statistics** – Calculate the mean and standard deviation for each analyte in these replicates. Determine the Half Range for the Prediction Interval of Results (HRPIR) using the following equation:

$$HR_{PIR} = 3.963 \times S$$

Where, S = the standard deviation and 3.963 is a constant value for seven replicates.

- c) **Calculate the Upper and Lower Limits for the Prediction Interval of results** (PIR = Mean $\pm$ HRPIR).
- d) **MRL Acceptance Criteria** – The laboratory's ability to meet the MRL is confirmed if the Upper PIR Limit is less than, or equal to, 150%; and the Lower PIR Limit is greater than, or equal to, 50%. If these criteria are not met, the MRL has been set too low and must be confirmed again at a higher concentration.

11.2.1.3 Low System Background

Analyze an LRB/MB immediately after the highest calibration standard in the selected calibration range. Confirm that the blank is free from contamination (less than 1/3 MRL). For automated extraction systems, a MB must be performed on each port.

11.2.1.4 Demonstration of Precision

Extract and analyze 7 LFBs spiked near the midpoint of the calibration curve and a MB. The %RSD of the replicate analyses must be <20% for all analytes.

11.2.1.5 Demonstration of Accuracy

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

Extract and analyze 7 LFBs spiked near the midpoint of the calibration curve and a MB. The average recovery for each analyte must be within 70-130% of the true value for each analyte.

11.2.2 Analyst Qualifications and Training

Employees that perform any step of this procedure must have a completed Read and Acknowledgment Statement for this version of the SOP in their training record. In addition, prior to unsupervised (independent) work on any client sample, analysts that prepare or analyze samples must have successful initial demonstration of capability (IDOC) and must successfully demonstrate on-going proficiency on an annual basis. Successful means the initial and on-going DOC met criteria, documentation of the DOC is complete, and the DOC record is in the employee's training file. Refer to laboratory SOP ENV-SOP-ORB1-0113_Employee Orientation and Training, or equivalent replacement, for more information.

12.0 DATA REVIEW AND CORRECTIVE ACTION

12.1 Data Review

The data review process of Pace® Analytical Services includes a series of checks performed at different stages of the process by different people to ensure that SOPs were followed, the analytical record is complete, and properly documented, QC criteria were met, proper corrective actions were taken for QC failure and other nonconformance(s), and test results are reported with proper qualification, when necessary.

The review and checks that are performed by the employee performing the task is called primary review.

All data and test results are also peer reviewed.

This process, known as secondary review is performed to verify SOPs were followed, that calibration, instrument performance, and QC criteria were met and/or proper corrective actions were taken, qualitative ID and quantitative measurement is accurate, all manual integrations are justified and documented, and approved in accordance with the Pace® Analytical Services SOP for manual integration, calculations are correct, the analytical record is complete and traceable, and that results are properly qualified.

Lastly, a third-level review, called a completeness check, is performed by reporting or project management staff to verify the test report is complete.

Refer to laboratory SOP ENV-SOP-ORB1-0119, *Data Review Process* for specific instructions and requirements for each step of the data review process.

12.2 Corrective Action

Corrective action is required when QC or sample results are not within acceptance criteria.

Refer to Appendix B for a complete summary of QC, acceptance criteria, and recommended corrective actions for QC associated with this test method.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

If corrective action is not taken or was not successful, the decision/outcome must be documented in the analytical record. The primary analyst has primary responsibility for taking corrective action when QA/QC criteria are not met. Secondary data reviewers must verify that appropriate action was taken and/or that results reported with QC failure are properly qualified.

Corrective action is also required when carryover is suspected and when results are over range.

Samples analyzed after a high concentration sample must be checked for carryover and reanalyzed if carryover is suspected. Carryover is usually indicated by low concentration detects of the analyte in successive samples analyzed after the high concentration sample.

Sample results at concentrations above the upper limit of quantitation must be diluted and reanalyzed. The result in the diluted samples should be within the upper half of the calibration range. Results less than the mid-range of the calibration indicate the sample was over diluted and analysis should be repeated with a lower level of dilution. If dilution is not performed, any result reported above the upper range is considered a qualitative measurement and must be qualified as an estimated value.

13.0 POLLUTION PREVENTION AND WASTE MANAGEMENT

Pace® proactively seeks ways to minimize waste generated during work processes. Some examples of pollution prevention include but are not limited to reduced solvent extraction, solvent capture, use of reusable cycletainers for solvent management, and real-time purchasing.

The EPA requires that laboratory waste management practices comply with all applicable federal and state laws and regulations. Excess reagents, samples, and method process wastes are characterized and disposed of in an acceptable manner in accordance with the Pace® Chemical Hygiene Plan / Safety Manual. Refer to this manual for these procedures.

14.0 MODIFICATIONS

A modification is a change to a reference test method made by the laboratory. For example, changes in stoichiometry, technology, quantitation ions, reagent or solvent volumes, reducing digestion or extraction times, instrument runtimes, etc. are all examples of modifications. Refer to Pace® ENV corporate SOP ENV-SOP-CORQ-0011 *Method Validation and Instrument Verification* for the conditions under which the procedures in test method SOPs may be modified and for the procedure and document requirements.

15.0 RESPONSIBILITIES

- PAS employees that perform any part this procedure in their work activities must have a signed Read and Acknowledgement Statement (R&A) in their training file for the version(s) of the SOP that were in effect during the time the employee performed the activity.
- PAS supervisors and managers however named, are responsible for training employees on the procedures in this SOP, implementing the SOP in the work area, and monitoring on-going adherence to the SOP in the work area(s) they oversee.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

- PAS employees are responsible for following the procedures in this SOP. Unauthorized deviations or departures from this SOP are not allowed except with documented approval from the local QM and only when those deviations do not violate the Pace® Code of Ethics or Professional Conduct (COR-POL-0004) or associated policy and procedure(s). Hand-edits or manual changes to the SOP are not permitted. If a change is desired or necessary, Pace® employees must follow the procedures for document revision specified in corporate SOPs ENV-SOP-CORQ-0015, Document Management and ENV-SOP-CORQ-0016, SOP for Creation of SOP and SWI.

16.0 ATTACHMENTS

- Appendix A: Target Analyte List and Routine LOQ
- Appendix B: QC Summary
- Appendix C: Routine Instrument Operating Conditions
- Appendix D: Example Automated Extractor Run
- Appendix E: Explanation of each step on the Automated Extractors

17.0 REFERENCES

- Pace® Quality Assurance Manual- most current version.
- The NELAC Institute (TNI) 2016 Standard
- USEPA, "Manual for the Certification of Laboratories Analyzing Drinking Water"; Fifth Edition, January 2005.
- USEPA, "Supplement 1to the Fifth Edition of the Manual for the Certification of Laboratories Analyzing Drinking Water"; June 2008
- USEPA, "Method 533: Determination of Per- and Polyfluoroalkyl Substances in Drinking Water by Isotope Dilution Anion Exchange Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry"; EPA 815-B-19-020; Office of Water: Cincinnati, OH, November 2019.
- USEPA, "UCMR5 Laboratory Approval Manual"; EPA 815-B-20-007; Office of Water: Cincinnati, OH, December 2020.

18.0 REVISION HISTORY

Section	Description of Change
8.1	Updated volume for 80:20.

This document supersedes the following document(s):

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

Document Number	Title	Effective Date
ENV-SOP-ORB1-0083 v06	EPA 533 PFAS	12/26/23

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

Appendix A: Target Analyte List and Routine LOQ/MRL

Table 1: Routine Analyte List and Limits of Quantitation (LOQ)¹

Analyte	CAS #	Drinking Water (ng/L)	UCMR (ng/L)
9-chlorohexadecafluoro-3-oxanone-1-sulfonic acid (9Cl-PF3ONS)	752426-58-1	2.0	2.0
11-chloroeicosafluoro-3-oxaundecane-1-sulfonic acid (11Cl-PF3OUdS)	763051-92-9	2.0	5.0
1H, 1H, 2H, 2H-perfluorodecane sulfonic acid (8:2 FTS)	39108-34-4	2.0	5.0
1H, 1H, 2H, 2H-perfluorooctane sulfonic acid (6:2 FTS)	27619-97-2	2.0	5.0
1H, 1H, 2H, 2H-perfluorohexane sulfonic acid (4:2 FTS)	757124-72-4	2.0	3.0
Hexafluoropropylene oxide dimer acid (GenX/HFPO-DA)	13252-13-6	2.0	5.0
4,8-dioxa-3H-perfluorononanoic acid (ADONA)	919005-14-4	2.0	3.0
Nonafluoro-3,6-dioxaheptanoic acid (NFDHA, 3,6-OPFHpA)	151772-58-6	2.0	20
Perfluoro(2-ethoxyethane)sulfonic acid (PFEEESA)	113507-82-7	2.0	3.0
Perfluorobutanesulfonic acid (PFBS)	375-73-5	2.0	3.0
Perfluoroheptanesulfonic acid (PFHpS)	375-92-8	2.0	3.0
Perfluoropentanesulfonic acid (PFPeS)	2706-91-4	2.0	4.0
Perfluoro-3-methoxypropanoic acid (PFMPA, PF4OPeA)	377-73-1	2.0	4.0
Perfluoro-4-methoxybutanoic acid (PFMBA, PF5OHxA)	863090-89-5	2.0	3.0
Perfluorohexanesulfonic acid (PFHxS)	355-46-4	2.0	3.0
Perfluorobutanoic acid (PFBA)	375-22-4	2.0	5.0
Perfluorodecanoic acid (PFDA)	335-76-2	2.0	3.0
Perfluorododecanoic acid (PFDOA)	307-55-1	2.0	3.0
Perfluoroheptanoic acid (PFHpA)	375-85-9	2.0	3.0
Perfluorohexanoic acid (PFHxA)	307-24-4	2.0	3.0
Perfluorononanoic acid (PFNA)	375-95-1	2.0	4.0
Perfluorooctanoic acid (PFOA)	335-67-1	2.0	4.0
Perfluoropentanoic acid (PFPeA)	2706-90-3	2.0	3.0
Perfluoroundecanoic acid (PFUnA)	2058-94-8	2.0	2.0
Perfluorooctanesulfonic acid (PFOS)	1763-23-1	2.0	4.0

¹ Values in place as of effective date of this SOP. LOQ are subject to change. For the most up to date LOQ, refer to the LIMS or contact the laboratory.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

Appendix B: QC Summary

QC Item	Frequency	Acceptance Criteria	Corrective Action	Qualification(non-UCMR)	Qualification UCMR/DoD
ICAL	Prior to sample analysis at instrument set up, after CCV failure.	Must meet one of curve fit options presented in Section 9.2.	Identify and correct source of problem, repeat	None. Do not proceed with analysis	Do not proceed with analysis
ICV/QCS	After Each ICAL	All analytes must be within 70-130% of the true value. (%R)	Identify source of problem, re-analyze. If repeat failure, repeat ICAL. Analysis may proceed if it can be demonstrated that the ICV exceedance has no impact on analytical measurements. For example, the ICV %R is high, CCV is within criteria, and the analyte is not detected in sample(s).	Qualify analytes with ICV out of criteria if reanalysis is not possible.	UCMR5: Do not report failed QCS to SDWARS. DoD: Qualify analytes with ICV out of criteria if reanalysis is not possible.
CCV	Daily, after every 10 samples, and at end of analytical window. DoD – CCV must be > Low Cal and ≤ midpoint Cal	All analytes must be within 70-130% of the true value. (%R)	Identify source of problem, re-analyze. If repeat failure, repeat ICAL. Analysis may proceed if the CCV %R is high and the analyte is not detected in sample(s). DoD: Reanalysis (2 consecutive passing CCVs) or corrective action must be taken.	Qualify analytes with CCV out of criteria.	UCMR5: Data may be reported only if the CCV %R is high and the analyte is not detected in sample(s). No other qualification is allowed. DoD: qualification is allowed only if samples cannot be reanalyzed.
CRDL/CCV-Low	Daily, before each analysis batch DoD – CCV must be > Low Cal and ≤ midpoint Cal	All analytes must be within 50-150% of the true value. (%R)	Identify source of problem, re-analyze. If repeat failure, repeat ICAL. Analysis may proceed if the CCV %R is high	Qualify analytes with CCV out of criteria.	UCMR5: Data may be reported only if the CCV %R is high and the analyte is not detected in sample(s). No

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

			and the analyte is not detected in sample(s). DoD: Reanalysis (2 consecutive passing CCVs) or corrective action must be taken.		other qualification is allowed. DoD: qualification is allowed only if samples cannot be reanalyzed.
Internal Standards / Isotope Performance Standards	Every field sample, standard and QC sample	Peak area counts must be within 50-150% of the average peak area in the ICAL.	Troubleshoot instrument performance. Reanalyze samples.	Qualify outages and explain in case narrative.	Data associated with failing IPS in samples or QC may not be reported.
Surrogate / Isotope Dilution Analogue	Every field sample, standard and QC sample	50-200% recovery for each analogue. UCMR: 50 to 200% in field and QC samples; 70 to 130% in the initial calibration and CCCs	Rerun sample. If failure confirms and capping CCV is within criteria re-extract if in hold or report with flags. If surrogates exceed maximum criteria BDL samples may be reported.	Quality samples with surrogates out of criteria.	Data associated with failing IDA may not be reported.
Method Blank / LRB	With each extraction batch of no more than 20 samples.	<1/3 MRL DoD: < ½ LOQ, or < 1/10 sample concentration, or < 1/10 regulatory limit) whichever is greater	Reanalyze and/or re-prepare batch of samples with new MB unless they meet exceptions below: Exceptions: 1) If sample ND, report sample without qualification 2) If sample result >10x MB detects and sample cannot be reanalyzed, report sample with appropriate qualifier indicating blank contamination. 3) If sample result <10x MB detects, report sample with appropriate qualifier to indicate an	Qualify samples with detections described in 2) and 3).	UCMR5: Data may not be reported regardless of sample result. DoD: Qualify results only if no more sample left to reprep

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

			estimated value. Client must be alerted and authorize this condition.		
LCS / LFB	With each extraction batch of no more than 20 samples.	Analytes fortified at $\leq 2 \times \text{MRL}$ must be within 50-150% of the true value. Analytes fortified at $> 2 \times \text{MRL}$ must be within 70-130% DoD: All Analytes must be at or $<$ midpoint.	Identify source of problem, re-analyze and/or re-prep batch of samples. Samples may be reported if recovery is high and the analyte is not detected in sample(s). DoD: Corrective action and reanalysis is required if an analyte fails 2 of 3 consecutive LCS.	Qualify samples with LCS and/or LCSD out of criteria.	UCMR5: LCS must be fortified at MRL. Data associated with any LCS failure may not be reported. DoD: Qualify samples with LCS and/or LCSD out of criteria.
MS/MSD	One set with each extraction batch of no more than 20 samples.	Analytes fortified at $\leq 2 \times \text{MRL}$ must be within 50-150% of the true value. Analytes fortified at $> 2 \times \text{MRL}$ must be within 70-130% $\leq 30\%$ RPD ($\leq 50\%$ RPD if analyte concentration is $\leq 2 \times \text{MRL}$)	If MB and LCS meet criteria, then samples may be reported with qualification.	Qualify target analytes on the parent sample.	UCMR5: Recovery and precision criteria do not apply. Do not upload MS/MSD with failing IDA/IPS to SDWARS. DoD: Qualify target analytes on the parent sample
Field Reagent Blank (FRB)	Per project requirements. FRB is required to run for any positive results.	$< 1/3$ MRL	Re-prep, re-analyze to confirm. Notify PM.	Qualify samples	Associated samples must be recollected.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

Appendix C: Routine Instrument Operating Conditions

Step	Total Time (min)	Flow Rate (uL/min)	A: 5mM Ammonium Acetate (%)	B: 5mM NH ₄ Acetate/ 95%Methanol/ 5%H ₂ O (%)
0	0.00	400	90.0	10.0
1	0.50	400	90.0	10.0
2	2.50	400	45.0	55.0
3	9.00	400	10.0	90.0
4	9.5	400	0.0	100.0
5	11.50	400	0.0	100.0
6	15.0	400	90.0	10.0

Parameter	Setting or Value
Sample Loop Size	100 µL
Injection Volume	2.0 µL
Draw Speed	100 µL/min
Eject Speed	400 µL/min
Needle Level	0.0 mm
Column Oven Temperature	50°C
MRM Scan Window	60 sec
Gas Temp	250°C
Sheath Gas Temp	375°C
Nebilizer	25 psi
Gas Flow	11 l/min
Fragmenter Voltage	166
Capillary Voltage	2500
V Charging	0
Ion Funnel Negative High Pressure	90
Ion Funnel Negative Low Pressure	60

Isotope Performance Standard	ISTD?	Precursor Ion	Product Ion
13C2-PFOA	Yes	415	370
13C3-PFBA	Yes	216	172
13C4-PFOS	Yes	503	80

Isotope Dilution Analogue	Isotope Performance Standard Reference	Precursor Ion	Product Ion

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

13C2-4:2FTS	13C4-PFOS	329	309
13C2-6:2FTS	13C4-PFOS	429	409
13C2-8:2FTS	13C4-PFOS	529	509
13C3-HFPO-DA	13C2-PFOA	287	169
13C3-PFBS	13C4-PFOS	302	80
13C3-PFHxS	13C4-PFOS	402	80
13C4-PFHpA	13C2-PFOA	367	322
13C5-PFHxA	13C2-PFOA	318	273
13C5-PFPeA	13C3-PFBA	268	223
13C6-PFDA	13C2-PFOA	519	474
13C7-PFUnA	13C2-PFOA	570	525
13C8-PFOA	13C2-PFOA	421	376
13C8-PFOS	13C4-PFOS	507	80
13C9-PFNA	13C2-PFOA	472	427
13C4-PFBA	13C3-PFBA	217	172
13C2-PFDoA	13C2-PFOA	615	570

Target Analyte	Isotope Dilution Analogue	Precursor Ion	Product Ion
11Cl- PF3OUdS	13C8-PFOS	630.9	450.9
11Cl- PF3OUdS	13C8-PFOS	630.9	83
NFDHA	13C5-PFHxA	295	201
NFDHA	13C5-PFHxA	295	85
NFDHA	13C5-PFHxA	201	85
4-2 FTS	13C2-4:2FTS	327	306.9
4-2 FTS	13C2-4:2FTS	327	80.9
6-2 FTS	13C2-6:2FTS	427	406.8
6-2 FTS	13C2-6:2FTS	427	80.9
8-2 FTS	13C2-8:2FTS	527	506.8
8-2 FTS	13C2-8:2FTS	527	79.9
9Cl-PF3ONS	13C8-PFOS	530.9	350.9
9Cl-PF3ONS	13C8-PFOS	530.9	83
ADONA	13C4-PFHpA	377	250.9
ADONA	13C4-PFHpA	377	85.1
HFPO-DA	13C3-HFPO-DA	285.1	185
HFPO-DA	13C3-HFPO-DA	285.1	169
PFMPA	13C4-PFBA	229	85
PFMBA	13C5-PFPeA	279	85
PFBA	13C4-PFBA	213	169
PFBS	13C3-PFBS	298.9	98.9

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

Target Analyte	Isotope Dilution Analogue	Precursor Ion	Product Ion
11Cl- PF3OUdS	13C8-PFOS	630.9	450.9
11Cl- PF3OUdS	13C8-PFOS	630.9	83
NFDHA	13C5-PFHxA	295	201
NFDHA	13C5-PFHxA	295	85
NFDHA	13C5-PFHxA	201	85
4-2 FTS	13C2-4:2FTS	327	306.9
4-2 FTS	13C2-4:2FTS	327	80.9
6-2 FTS	13C2-6:2FTS	427	406.8
PFBS	13C3-PFBS	298.9	80
PFDA	13C6-PFDA	513	469
PFDA	13C6-PFDA	513	218.7
PFDaA	13C2-PFDaA	613	569
PFDaA	13C2-PFDaA	613	319
PFEESA	13C3-PFBS	314.9	135
PFEESA	13C3-PFBS	314.9	69.1
PFHpA	13C4-PFHpA	362.9	319
PFHpA	13C4-PFHpA	362.9	169
PFHpS	13C8-PFOS	449	80
PFHxA	13C5-PFHxA	313	268.9
PFHxA	13C5-PFHxA	313	119
PFHxS	13C3-PFHxS	398.9	99
PFHxS	13C3-PFHxS	398.9	80
PFNA	13C9-PFNA	463	419
PFNA	13C9-PFNA	463	169
PFOA	13C8-PFOA	413	369
PFOA	13C8-PFOA	413	169
PFOS	13C8-PFOS	498.9	99
PFOS	13C8-PFOS	498.9	80
PFPeA	13C5-PFPeA	263	218.9
PFPeS	13C3-PFHxS	348.9	98.9
PFPeS	13C3-PFHxS	348.9	79.9
PFUnA	13C7-PFUnA	563	519
PFUnA	13C7-PFUnA	563	269

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

Appendix D: Example Automated Extractor Run.

(This is just an example and can be changed to meet updated extraction practices)

- Elute W2, Solvent 1, 10, 10mLs
- Elute W1, Solvent 3, 10, 10mLs
- Elute W1, Solvent 3, 10, 3mLs
- Elute W1, Solvent 2, 10, 3.0mLs
- Add Samp W1, Sample, 5, 300mLs
- Rinse W1, Solvent 4, 5, 10mLs
- Shake, Time based, 30 seconds
- Clean, Solvent 1, 5, 1mLs
- Add Samp W2, Sample, 5, 9mLs
- Blow N2, Time based, 5 min
- Rinse 1, Solvent 5, 2, 5mLs
- Rinse 1, Solvent 5, 2, 5mLs
- Shake, Time based, 10 s
- Collect 1, Sample, 2, 4.5mLs

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0083 v07_EPA 533 PFAS
	Effective Date: 02/20/2024

Appendix E: Explanation of each step on the Automated Extractors

Prime – clean valve and pump with selected solvent(s) Solvent(s) are drawn through valves 1 and 2 into the pumps and then pushed out into the top waste instead of flowing through the SPE columns. This can be used to fill or clean solvent tubing.

Elute – pre-condition or wash cartridge with selected solvent(s) Solvent(s) are pushed through the SPE columns into bottom waste 1 or 2. The difference between Elute and Collect is that Collect will push solvents into Fraction 1 or Fraction 2

Add Sample – load sample onto SPE cartridge Sample(s) are drawn and pushed through the SPE columns and then into bottom waste 1 or 2

Collect – elute with selected solvent(s) and collect eluent in specified fraction tubes Solvent(s) are pushed through the SPE columns to elute analytes into Fraction 1 or Fraction 2. Samples can also be directly collected into Fraction 1 or Fraction 2. This is used when manual rinsing of sample containers is performed and the rinsate needs to be collected

Air-purge – use air to purge residual fluid through columns into fractions/waste or to mix sample Outside air is drawn in and then pushed into sample containers (eg. For mixing internal standards) or delivered through SPE columns into fractions or waste. Air-purge can also be used to mix sample after manually adding internal standards

Blow N2 – Blow nitrogen for a specified amount of time or until user manually triggers next step “Time based” option will turn on nitrogen blowing for the specified amount of time. “Manual resume” option will continue nitrogen blowing until the operator presses “start” to resume. A loud “beep” is triggered when using the “Manual resume” option to get the operator’s attention

Rinse – rinse sample containers and deliver rinsate into fraction tubes or waste

Clean – deliver selected solvent(s) into sample containers followed by optional air purge to clear solvent(s) from liquid lines

Shake – activate resonator for a specified amount of time or until user manually triggers next step

Wait – pause run for a specified amount of time or until user manually triggers next step

Pump-Cln – Rinses pump syringes and plungers with solvent(s) and delivers to fractions or waste

Boost – Moves liquid in the cartridge sorbent back and forth

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

Management Approval: Jeffery Dunster Approved on 12/5/2023 8:12:11 AM

Rebecca Summerlot Approved on 12/5/2023 1:11:12 PM

Trisha Kelly Approved on 12/8/2023 8:28:16 AM

Thomas Widera Approved on 12/8/2023 12:29:30 PM

©2019-2023, Pace® Analytical Services, LLC

1.0 SCOPE AND APPLICATION

This Standard Operating Procedure (SOP) describes the laboratory procedure for the determination of selected Per- and Polyfluorinated Alkyl Substances (PFAS) by Solid Phase Extraction and Liquid Chromatography Tandem Mass Spectrometry (LC/MS/MS). This SOP is applicable to Drinking Water and Non-Potable Water.

1.1 Target Analyte List and Limits of Quantitation (LOQ)

The target analytes and the normal LOQ that can be achieved with this procedure are provided in Table 1, Appendix A.

LOQ are established in accordance with Pace® policy and SOPs for method validation and for the determination of detection limits (DL) and quantitation limits (LOQ). DL and LOQ are routinely verified and updated when needed. Refer to Pace® Analytical Services – Florida SOP ENV-SOP-ORB1-0124, *Determination of Limit of Detection and Limit of Quantitation*, or equivalent replacement, for these procedures. The current LOQ for each target analyte that can be determined by this SOP as of the effective date of this SOP is provided in Table 1, Appendix A. Current MDL are located in the laboratory LIMS.

The reporting limit (RL) is the value to which analytes are reported as detected or not detected in the final report. When the RL is less than the lower limit of quantitation (LLOQ), all detects and non-detects at the RL are qualitative. The LLOQ is the lowest point of the calibration curve used for each target analyte.

DL, LOQ, and RL are always adjusted to account for actual amounts used and for dilution.

2.0 SUMMARY OF METHOD

A 250 mL water sample is fortified with surrogates and passed through a solid phase extraction (SPE) cartridge containing polystyrenedivinylbenzene (SDVB) to extract the method analytes and surrogates. The compounds are eluted from the solid phase with a small amount of methanol. The extract is concentrated to dryness with nitrogen in a heated water bath, and then adjusted to a 1 mL volume with 96:4% (vol/vol) methanol:water after adding the IS(s). A 10 µL injection is made into a Liquid Chromatography (LC) System equipped with a C18 column that is interfaced to a Tandem Mass Spectrometer. The analytes are separated and identified by comparing the acquired mass spectra and retention times to reference spectra and retention times for calibration standards acquired under identical LC/MS/MS conditions. The concentration of each analyte is determined by using the internal standard technique. Surrogate analytes are added to all Field and QC Samples to monitor the extraction efficiency of the method analytes.

3.0 INTERFERENCES

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

All glassware must be meticulously cleaned. Wash glassware with detergent and tap water, rinse with tap water, followed by a reagent water rinse. Non-volumetric glassware can be heated in a muffle furnace at 400 °C for 2 h or solvent rinsed. Volumetric glassware should be solvent rinsed and not be heated in an oven above 120 °C. Store clean glassware inverted or capped. Do not cover with aluminum foil because PFAAs can be potentially transferred from the aluminum foil to the glassware.

PFAA standards, extracts and samples should not come in contact with any glass containers or pipettes as these analytes can potentially adsorb to glass surfaces. PFAA analyte, IS and SUR standards commercially purchased in glass ampoules are acceptable; however, all subsequent transfers or dilutions performed by the analyst must be prepared and stored in polypropylene or HDPE (high density polyethylene) containers.

Method interferences may be caused by contaminants in solvents, reagents (including reagent water), sample bottles and caps, and other sample processing hardware that lead to discrete artifacts and/or elevated baselines in the chromatograms. The method analytes in this method can also be found in many common laboratory supplies and equipment, such as PTFE (polytetrafluoroethylene) products, LC solvent lines, methanol, aluminum foil, SPE sample transfer lines, etc. All items such as these must be routinely demonstrated to be free from interferences (less than 1/3 the MRL for each method analyte) under the conditions of the analysis by analyzing laboratory reagent blanks. Subtracting blank values from sample results is not permitted.

Matrix interferences may be caused by contaminants that are co-extracted from the sample. The extent of matrix interferences will vary considerably from source to source, depending upon the nature of the water. Humic and/or fulvic material can be co-extracted during SPE and high levels can cause enhancement and/or suppression in the electrospray ionization source or low recoveries on the SPE sorbent. Total organic carbon (TOC) is a good indicator of humic content of the sample. Under the LC conditions used during method development, matrix effects due to total organic carbon (TOC) were not observed.

Relatively large quantities of the preservative are added to sample bottles. The potential exists for trace-level organic contaminants in these reagents. Interferences from these sources should be monitored by analysis of laboratory reagent blanks, particularly when new lots of reagents are acquired.

SPE cartridges can be a source of interferences. The analysis of field and laboratory reagent blanks can provide important information regarding the presence or absence of such interferences. Brands and lots of SPE devices should be tested to ensure that contamination does not preclude analyte identification and quantitation.

4.0 DEFINITIONS

Refer to the Laboratory Quality Manual for a glossary of common lab terms and definitions.

- **Lowest Concentration Minimum Reporting Level (LCMRL):** The single laboratory LCMRL is the lowest true concentration for which a future recovery is expected, with 99% confidence, to be between 50 and 150% recovery.
- **Precursor Ion:** For the purpose of this method, the precursor ion is the deprotonated molecule ($[M-H]^-$) of the method analyte. In MS/MS, the precursor ion is mass selected and fragmented by collisionally activated dissociation to produce distinctive product ions of smaller m/z .

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

- **Primary Dilution Standard (PDS) Solution:** A solution containing the analytes prepared in the laboratory from stock standard solutions and diluted as needed to prepare calibration solutions and other needed analyte solutions.
- **Product Ion:** For the purpose of this method, a product ion is one of the fragment ions produced in MS/MS by collisionally activated dissociation of the precursor ion.
- **LLCS:** Low concentration LCS = SRM

5.0 HEALTH AND SAFETY

Contact your supervisor or local safety coordinator with questions or concerns regarding safety protocol or safe handling procedures for this procedure.

The following sections provide general health and safety information about chemicals and materials that may be present in the laboratory.

- The toxicity or carcinogenicity of each chemical material used in the laboratory has not been fully established. Each chemical should be regarded as a potential health hazard and exposure to these compounds should be as low as reasonably achievable.
- The laboratory maintains documentation of hazard assessments and OSHA regulations regarding the safe handling of the chemicals specified in each method. Safety data sheets for all hazardous chemicals are available to all personnel. Employees must abide by the health, safety and environmental (EHS) policies and procedures specified in this SOP and in the Pace® Chemical Hygiene / Safety Manual (COR-MAN-0001)
- Personal protective equipment (PPE) such as safety glasses, gloves, and a laboratory coat must be worn in designated areas and while handling samples and chemical materials to protect against physical contact with samples that contain potentially hazardous chemicals and exposure to chemical materials used in the procedure.
- Concentrated corrosives present additional hazards and are damaging to skin and mucus membranes. For procedures that require use of acids, use acids in a fume hood whenever possible with PPE designed for handling these materials. If eye or skin contact occurs, flush with large volumes of water. When working with acids, always add acid to water to prevent violent reactions. For procedures that emit large volumes of solvents (evaporation/concentration processes), these activities must be performed in a fume hood or apparatus that reduces exposure.

6.0 SAMPLE COLLECTION, PRESERVATION, HOLDING TIME, AND STORAGE

PAS offers sample collection services for this test method. Refer to SOP ENV-SOP-ORB1-0108, *Bottle and Water Testing* for the procedure used when sample collection is performed by the location referenced in the scope and application of this SOP. If sample collection is performed by a different PAS facility, contact a PAS Project Manager (PM) to obtain the SOP used by that location.

PAS provides containers for the collection of samples upon client request. The routine container type and size provided is provided in the following table along with requirements for sample size, preservation and holding time. Refer to laboratory SOP ENV-SOP-ORB1-0123, *Bottle Preparation*, for procedures related to preparation of bottle kits.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

The sample handler must wash their hands before sampling and wear nitrile gloves while filling and sealing the sample bottles. PFAS contamination during sampling can occur from a number of common sources, such as food packaging and certain foods and beverages. Proper hand washing and wearing nitrile gloves will aid in minimizing this type of accidental contamination of the samples.

General Requirements

Matrix	Routine Container	Minimum Sample Amount ¹	Preservation ²	Holding Time
Water	Polypropylene or HDPE bottle and cap	250 mL	Thermal: $\leq 10^{\circ}\text{C}^3$ Chemical: 5.0 g/L Trizma	Collection to Extraction: 14 days Extraction to Analysis: 28 days

¹Minimum amount needed for each discrete analysis.

²Preservative added to the sample bottle as solid crystals.

³Samples must not exceed 10°C during the first 48 hours after collection. After which samples must be kept at $\leq 6^{\circ}\text{C}$

After collecting the sample, cap the bottle and agitate by hand until preservative is dissolved. Keep the sample sealed from time of collection until extraction.

Field / Matrix QC

Field Reagent Blank (FRB)	Matrix Spike/Spike Duplicate (MS/MSD)
See Field Reagent Blank in Appendix B.	See MS/MSD and Duplicate in Appendix B.

A Field Reagent Blank (FRB) is provided along with each sample set. The sample set is composed of samples collected from the same sample site and at the same time. At the laboratory, fill the field blank sample bottle with reagent water, then seal, and ship to the sampling site along with the sample bottles. For each FRB shipped, a second FRB bottle containing only the preservative must also be shipped. At the sampling site, the sampler must open the shipped FRB and pour the preserved reagent water into the empty shipped sample bottle, seal and label this bottle as the FRB. The FRB is shipped back to the laboratory along with the samples and analyzed to ensure that PFAS were not introduced into the sample during sample collection/handling.

The same batch of Trizma preservative is used for the FRBs as is used for the field samples. This is documented during bottle kit creation. The reagent water used for the FRBs is initially analyzed for all method analytes (using the same lot of sample bottles as the field samples) and must meet the MB criteria in Appendix B prior to use. This is to ensure samples are not being discarded due to contaminated reagent water or sample bottles rather than contamination during sampling.

Thermal preservation is checked and recorded on receipt in the laboratory in accordance with Pace® Analytical Services – Florida SOP ENV-SOP-ORB1-0120, *Sample Management*, or equivalent replacement. Chemical preservation is checked and recorded at time of receipt or prior to sample

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

preparation. Trizma is targeted to produce a pH of 7.0 ± 0.5 and to reduce residual free Chlorine to <0.1 mg/L when added to samples as described above.

After receipt, samples are stored at $\leq 6^{\circ}\text{C}$ until sample preparation. Prepared samples (extracts) are stored at room temperature until sample analysis.

7.0 EQUIPMENT AND SUPPLIES

7.1 Equipment

- **Extraction Equipment**

Semi-Automated sample preparation system designed for use with SPE cartridges. Care must be taken with automated SPE systems to ensure the PTFE commonly used in these systems does not contribute to unacceptable analyte concentrations in the LRB. (Fluid Management part # EZPFC-12 - 12 Position EZPFC System for water & wastewater analysis, or equivalent)

Automated Sample preparation system care must be taken with automated SPE systems to ensure the PTFE commonly used in these systems does not contribute to unacceptable analyte concentrations in the LRB. (Promochrom model SPE-03 Gen 4, or equivalent)

- **Extract Concentration System:** Extracts are concentrated by evaporation with nitrogen using a water bath set to no higher than 65°C (Meyer N-Evap, Model 111, Organomation Associates, Inc. or equivalent).
- **Sample Delivery System:** Use of a polypropylene transfer tube system, which transfers the sample directly from the sample container to the SPE cartridge, is recommended but not mandatory. Standard extraction manifolds come equipped with PTFE transfer tube systems. These can be replaced with 1/8 inch O.D. X 1-16 in I.D. polypropylene or polyethylene tubing cut to an appropriate length to ensure no sample contamination from the sample transfer lines. Other types of non PTFE tubing may be used provided it meets the LRB and LFB QC requirements. The PTFE transfer tube may be used but an LRB must be run on each PTFE transfer tube and the QC requirement must be met. In the case of an automated SPE, the removal of PTFE lines may not be feasible; therefore, LRB's will need to be rotated among the ports and must meet QC requirements.
- **Laboratory or Aspirator Vacuum System:** Sufficient capacity to maintain a vacuum of approximately 10 to 15 inches of mercury for extraction cartridges.
- **Instrumentation**
 - **LC System:** Instrument capable of reproducibly injecting up to 10- μL aliquots and performing binary linear gradients at a constant flow rate near the flow rate used for development of this method (0.3 mL/min). The LC must be capable of pumping the water/methanol mobile phase without the use of a degasser which pulls vacuum on the mobile phase bottle (other types of degassers are acceptable). Degassers which pull vacuum on the mobile phase

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

bottle will volatilize the ammonium acetate mobile phase causing the analyte peaks to shift to earlier retention times over the course of the analysis batch. The usage of a column heater is optional.

- **LC/Tandem Mass Spectrometer:** The LC/MS/MS must be capable of negative ion electrospray ionization (ESI) near the suggested LC flow rate of 0.3 mL/min. The system must be capable of performing MS/MS to produce unique product ions for the method analytes within specified retention time segments. A minimum of 10 scans across the chromatographic peak is required to ensure adequate precision.
- **Analytical Column:** A Thermo Accucore (cat#17126-153030 or equivalent) 3 x 150 mm C18 column, packed with 2.6µm C18 solid phase particles is used. Any column that provides adequate resolution, peak shape, capacity, accuracy, and precision may be used (ie Waters XTERRA 2.1 x 150 mm C18 column, 3.5 µm C18 particle size or equivalent).
- **Support Equipment Polypropylene Graduated Cylinders:** Suggested sizes include 25, 50, 100 and 1000 mL cylinders.
- **Micro Syringes:** Suggested sizes include 5, 10, 25, 50, 100, 250, 500 and 1000 µL syringes.
- **Adjustable Autopipettes:** Ranges 10-100 µL, 100-1000 µL, and 1000-5000 µL.
- **Analytical Balance:** Capable of weighing to the nearest 0.01g.

7.2 Supplies

- **Sample Containers:** 250 mL polypropylene or HDPE bottles fitted with polypropylene or HDPE screw caps. Sample bottles must be discarded after use.
- **Polypropylene Bottles:** 4 mL narrow-mouth polypropylene or HDPE bottles (Thermo Cat# 2006-9125 or equivalent).
- **Centrifuge Tubes:** 15 mL conical polypropylene tubes with polypropylene screw caps for storing standard solutions and for collection of the extracts (Sarstedt Cat# 62.554.205 or equivalent).
- **Autosampler Vials:** Polypropylene 0.3 mL autosampler vials (Thermo Cat# C4010-11 or equivalent) with polypropylene caps Thermo Cat#: 5330-10 or equivalent.
- **Plastic Pipette Tips:** Polypropylene or polyethylene disposable pipet tips (Fisher Cat. No.: 13-711-7 or equivalent).
- **SPE Cartridges:** 0.5g, 6 mL SPE cartridges HL DVB extraction cartridge (UCT part number: ECHLD456-P or equivalent).
- **pH Strips:** Narrow range. Fisherbrand 5.5-8.0 (cat# 13-640-517) or equivalent.
- **Free Chlorine DPD Powder:** HF Scientific 10504C or equivalent.
- **Chlorine Strips:** HF Scientific Catalog # 09940 or equivalent. Gradations of 0.5 pH units and detection to 0.1 mg/L.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

- **Microsyringes:** Suggested sizes include 5, 10, 50, 100, 250, 500, and 1000 µL syringes.
- **Micropipettes:** Suggested size up to 1000 µL
- **Top loading balance:** Capable of weighing to the nearest 0.01 g.

8.0 REAGENTS AND STANDARDS

Refer to Pace® Analytical Services – Florida SOP ENV-SOP-ORB1-0107, *Standard and Reagent Prep and Traceability*, or equivalent replacement, for information regarding standard and reagent tracking and traceability.

8.1 Reagents

- **Reagent Water:** (HPLC grade or equivalent) Purified water which does not contain any measurable quantities of any method analytes or interfering compounds greater than 1/3 the MRL for each method analyte of interest. (Fisher Cat# W6-1 or equivalent)
- **Methanol (CH₃OH, CAS#: 67-56-1):** High purity (HPLC grade or equivalent), demonstrated to be free of analytes and interferences (Fisher Cat# A452-4 or equivalent). Expiration per manufacturer.
- **Ammonium Acetate (NH₄C₂H₃O₂, CAS#: 631-61-8):** High purity, demonstrated to be free of analytes and interferences (Sigma-Aldrich HPLC grade or equivalent). Expiration per manufacturer.
- **537.1 Aqueous Mobile Phase (20 mM Ammonium Acetate):** To prepare 1 L, add 1.54g ammonium acetate to 1 L of reagent water. This solution is prone to volatility losses. Expires 7 days from preparation.
- **537.1 Solvent:** Mix 40 mL of Optima grade Water with 960 mL of Optima grade Methanol. Used to prepare all 537.1 standards. Expires 1 year from preparation.
- **Methanol Wash Solution, 10%:** Mix 50 mL of methanol with 450 mL of water. Stored on the instrument in syringe wash bottle position.
- **Trizma Preset Crystals, pH 7.0 (Sigma cat# T-7193 or equivalent):** Reagent grade. Trizma functions as a buffer and removes free chlorine in chlorinated finished waters. Expiration per manufacturer.
- **Nitrogen:** Nitrogen aids in aerosol generation of the ESI liquid spray and is used as collision gas in some MS/MS instruments. The nitrogen used should meet or exceed instrument manufacturer's specifications. Nitrogen is also used to concentrate sample extracts (Ultra High Purity or equivalent).
- **Argon:** Used as collision gas in MS/MS instruments. Argon should meet or exceed instrument manufacturer's specifications. Nitrogen gas may be used as the collision gas provided sufficient sensitivity (product ion formation) is achieved.

8.2 Stock Standards

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

- **Standard Solutions** – When a compound purity is assayed to be 96% or greater (standards purchased from Wellington are >98%), the weight can be used without correction to calculate the concentration of the stock standard. PFAS analyte, IS, and SUR standards commercially purchased in glass ampoules are acceptable; however, all subsequent transfers or dilutions performed by the analyst must be prepared and stored in HDPE containers.
- **Correction for analytes obtained in the salt form** – PFHxS, PFOS and other sulfonic acids are not available as the acid form, but rather as their corresponding salts, such as Na⁺ and K⁺. These salts are acceptable for use as stock standards as long as the weight is corrected for the salt content according to the equation below.

$$mass(acid\ form) = mass(salt\ form) \times \frac{MW_{acid}}{MW_{salt}}$$

Where:

MW_{acid} = the molecular weight of PFAA

MW_{salt} = the molecular weight of purchased salt

- **537.1-SPK:** 537.1 PFAA Spike Standard Full List (Wellington EPA-537PDS-R1 or equivalent). Used for preparation of the calibration curve and spiking solution. Expiration per manufacturer. A second lot may be used for the ICV.

Compound	Concentration (µg/mL)	Compound	Concentration (µg/mL)
PFBS	2.0	PFTeDA	2.0
PFHpA	2.0	PFTTrDA	2.0
PFNA	2.0	PFUnA	2.0
PFOS ¹	2.0	NEtFOSAA ¹	2.0
PFOA	2.0	NMeFOSAA ¹	2.0
PFHxS ¹	2.0	HFPO-DA	2.0
PFDA	2.0	ADONA	2.0
PFDaA	2.0	11Cl-PF3OUdS	2.0
PFHxA	2.0	9Cl-PF3ONS	2.0

¹These analytes are present as linear and branches isomers and are corrected for salt concentration. Standard should be vortexed before use.

- **537.1-SPK:** 537.1 PFAA Spike Standard Mix 2 (Absolute Cat. No. 99896 or equivalent). Used for preparation of the calibration curve and spiking solution. Expiration per manufacturer. A second lot may be used for the ICV.

Compound	Concentration (µg/mL)	Compound	Concentration (µg/mL)

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

PFBS	2.0	PFNA	2.0
PFHpA	2.0	PFOS ¹	2.0
PFOA	2.0	PFTTrDA	2.0
PFHxS ¹	2.0	PFUnA	2.0
PFDA	2.0	NEtFOSAA ¹	2.0
PFDaA	2.0	NMeFOSAA ¹	2.0
PFHxA	2.0	HFPO-DA	2.0
PFTeDA	2.0	ADONA	2.0
9CI-PF3ONS	2.0	11CI-PF3OUdS	2.0

¹These analytes are present as linear and branches isomers and are corrected for salt concentration. Standard should be vortexed before use.

- **537-IS:** Isotopically Labelled Internal Standards (Wellington Labs EPA537IS or equivalent) Add 10 µL to final extract. Store ≤6° C. Expires 180 days from initial use.

Compound	Concentration (µg/mL)	Compound	Concentration (µg/mL)
¹³ C ₂ -PFOA	1.0	NMeFOSAA-d ₃	4.0
¹³ C ₄ -PFOS	3.0	X	X

- **537-SS:** Isotopically Labelled Surrogates (Wellington Labs EPA-537SS-R1 or equivalent). Add 10 µL to 1 mL calibration standards and samples to be extracted. Used for curve prep and sample extraction. Store ≤6° C. Expires 180 days from initial use.

Compound	Concentration (µg/mL)	Compound	Concentration (µg/mL)
¹³ C ₂ -PFHxA	1.0	¹³ C ₂ -PFDA	1.0
¹³ C ₃ -HFPO-DA	1.0	NEtFOSAA-d ₅	4.0

• Working Solutions

- **1,3,6-Polytyrosine:** Instrument tuning compound. Using a standard solution is recommended (C S Bio Co. Cat#CS0272L or equivalent), however solution can be prepared from a neat material.
- **537.1 Spike:** Prepare 2 spiking solutions at concentrations of 0.02 µg/mL and 2.0 µg/mL. The low concentration spike is used for preparation of low and mid-level spiking for LCS and LFSM/LFSMD. The high concentration spike is used only for high level LCS and LFSM/LFSMD preparation. Spiking levels need to be rotated among low-, mid- and high- level concentrations on a per batch basis using the spike rotation logbook. Vortex standard before use.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

- **537.1 ICV Spike:** Prepared using 2nd source standards (different manufacturer or different lot from the same manufacturer). Expiration per manufacturer. Store ≤6° C.

Calibration Level	Amount of ICV Spike (µL)	Amount of 537.1-SS (µL)	Amount of 537.1-IS (µL)	Final Volume (mL)
ICV	5.0	20	20	2.0

- **537.1 Calibration Standards:** Prepared using 537-IS Soln., 537-SS Soln., and 537.1 Spike in 537.1 Solvent. Expires 90 days from prep. Refer to table below.

Note: Method expiration is 14 days from prep. In house stability testing has shown that calibration standards can pass against freshly prepared curves at least three months from the time of preparation.

Calibration Level	Amount of 537.1 Spike (µL)	Amount of 537.1-SS (µL)	Amount of 537.1-IS (µL)	Final Volume (mL)
1	0.5	20	20	2.0
2	1.0	20	20	2.0
3	2.0	20	20	2.0
4	5.0	20	20	2.0
5	10	20	20	2.0
6	20	20	20	2.0
7	40	20	20	2.0
8	100	20	20	2.0

9.0 PROCEDURE

9.1 Equipment Preparation

9.1.1 Support Equipment

Refer to Pace® Analytical Services – Florida SOP ENV-SOP-ORB1-0105, *Support Equipment*, or equivalent replacement, for additional information on calibration requirements for support equipment that may be used in this procedure.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

9.1.2 Instrument

9.1.2.1 Routine Instrument Operating Conditions

Refer to Appendix C.

9.1.2.2 Prior to daily operation, flush the LC column with Methanol for a minimum of 20 minutes to remove residual PFAS compounds from the column.

9.2 Initial Calibration (ICAL)

Refer to policy ENV-POL-CORQ-0005 for general laboratory calibration policies and procedures.

9.2.1 Calibration Design

ANALYTE	CAL1 (µg/L)	CAL2 (µg/L)	CAL3 (µg/L)	CAL4 (µg/L)	CAL5 (µg/L)	CAL6 (µg/L)	CAL7 (µg/L)	CAL8 (µg/L)
PFBS	0.5	1.0	2.0	5.0	10.0	20.0	40.0	100.0
PFDA	0.5	1.0	2.0	5.0	10.0	20.0	40.0	100.0
PFDoA	0.5	1.0	2.0	5.0	10.0	20.0	40.0	100.0
PFHpA	0.5	1.0	2.0	5.0	10.0	20.0	40.0	100.0
PFHxS	0.5	1.0	2.0	5.0	10.0	20.0	40.0	100.0
PFHxA	0.5	1.0	2.0	5.0	10.0	20.0	40.0	100.0
PFNA	0.5	1.0	2.0	5.0	10.0	20.0	40.0	100.0
PFOS	0.5	1.0	2.0	5.0	10.0	20.0	40.0	100.0
PFOA	0.5	1.0	2.0	5.0	10.0	20.0	40.0	100.0
PFTeDA	0.5	1.0	2.0	5.0	10.0	20.0	40.0	100.0
PFTTrDA	0.5	1.0	2.0	5.0	10.0	20.0	40.0	100.0
PFUnA	0.5	1.0	2.0	5.0	10.0	20.0	40.0	100.0
NEtFOSAA	0.5	1.0	2.0	5.0	10.0	20.0	40.0	100.0
NMeFOSAA	0.5	1.0	2.0	5.0	10.0	20.0	40.0	100.0
HFPO-DA	0.5	1.0	2.0	5.0	10.0	20.0	40.0	100.0
ADONA	0.5	1.0	2.0	5.0	10.0	20.0	40.0	100.0
9CIPF3ONS	0.5	1.0	2.0	5.0	10.0	20.0	40.0	100.0
11CIPF3OUdS	0.5	1.0	2.0	5.0	10.0	20.0	40.0	100.0
¹³ C ₂ PFOA (IS)	10.0	10.0	10.0	10.0	10.0	10.0	10.0	10.0
¹³ C ₄ PFOS (IS)	30.0	30.0	30.0	30.0	30.0	30.0	30.0	30.0

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

ANALYTE	CAL1 (µg/L)	CAL2 (µg/L)	CAL3 (µg/L)	CAL4 (µg/L)	CAL5 (µg/L)	CAL6 (µg/L)	CAL7 (µg/L)	CAL8 (µg/L)
NMeFOSAA-d ₃ (IS)	40.0	40.0	40.0	40.0	40.0	40.0	40.0	40.0
¹³ C ₂ -PFHxA (S)	10.0	10.0	10.0	10.0	10.0	10.0	10.0	10.0
¹³ C ₂ -PFDA (S)	10.0	10.0	10.0	10.0	10.0	10.0	10.0	10.0
NEtFOSAA-d ₅ (S)	40.0	40.0	40.0	40.0	40.0	40.0	40.0	40.0
¹³ C ₃ HFPO-DA (S)	10.0	10.0	10.0	10.0	10.0	10.0	10.0	10.0

9.2.2 Calibration Sequence

Prepare a set of at minimum five CAL standards (six if a quadratic curve fit is to be used). Analyze each standard level with the same acquisition method used to analyze samples, changes to retention times or other analytical parameters are saved as part of the local method generated with each analytical sequence, these parameters can be adjusted mid-sequence so long as they are applied to all data. Instrument method adjustments, most commonly adjusting Selected Reaction Monitoring (SRM) windows due to retention time shifts, are done via the Xcalibur Instrument Setup and will require restarting the sequence.

ESI-MS/MS Tuning requirements: As directed by manufacturer's instructions or as needed, most commonly after any instrument maintenance that breaks the MS/MS vacuum.

9.2.2.1 Load a 500 µL syringe filled with 1,3,6-polytyrosine in the syringe pump and connect directly to the probe.

9.2.2.2 Hold syringe pump power button for a few seconds to purge the line.

9.2.2.3 Using the Tune Master software adjust the parameters under the Optimize Compound Dependent Devices tab to a relative signal maximum for peaks 180.066, 506.193, and 995.383 in negative mode and 182.082, 508.208, and 997.398 in positive mode.

9.2.2.4 When done run the Auto Tune & Calib under the System Tune and Calibration Workspace tab to optimize response and peak shape.

9.2.3 ICAL Evaluation

9.2.3.1 Curve Fit

Removal or replacement of calibration points must be performed in accordance with ENV-POL-CORQ-0005.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

The LC/MS/MS system is calibrated using the IS technique. Use the LC/MS/MS data system software to generate a linear regression or quadratic calibration curve for each of the analytes. This curve must always be forced through zero and may be concentration weighted, if necessary. Forcing zero allows for a better estimate of the background levels of method analytes.

Due to the prevalence of PFOA and PFOS detections greater than the upper quantitation limit, quadratic curve fits may cause erroneous non-detection results. Quadratic may be used, but it is recommended that the run is halted, system suitability is assessed, and system is recalibrated before continuing.

Provided a minimum of five calibration points are still being used (six for quadratic curve fits), a point at the top or bottom of the calibration curve may be dropped to achieve recovery requirements across the remaining points. Dropping high concentration points lowers the upper quantitation limit of the calibration and may require that more dilutions are performed. Dropping low calibration points will elevate the MRL for this sequence. Refer to Appendix B for frequency, acceptance criteria, and corrective actions.

9.2.3.2 Initial Calibration Verification (ICV/QCS)

Immediately following the initial calibration, prepare and analyze a mid-level fortified ICV to verify the validity of the calibration standards. The absolute percent recovery of the target analytes should meet the criteria as defined in Appendix B relative to the initial calibration. If the analyte falls outside of this range, first verify the linearity of the calibration curve, then identify possible causes. If the second ICV fails low for the analyte, it may not be reported from the calibration curve. If a second ICV fails high for the analyte, then the analyte may be reported from the calibration curve only if the concentration is below the reporting limit. It is recommended that once an ICV fails twice, the system be maintained and/or recalibrated.

9.2.3.3 Peak Asymmetry Factor

Peak Asymmetry factor must be calculated during the IDOC, with each ICAL, and each time chromatographic changes are made that may affect peak shape using the first two eluting peaks at a mid-level standard. See Section 10.4 for calculations. Refer to Appendix B for frequency, acceptance criteria, and corrective actions. The PFA check is also used for setting the Retention time and updating the retention time for each parameter.

9.2.4 Continuing Calibration Verification (CCV)

9.2.4.1 Frequency - Analyze CCV standards at the beginning of each Analysis Batch, after every tenth field sample, and at the end of the Analysis Batch. The beginning CCV for each Analysis Batch must be at, or below, the MRL for each analyte. This CCV verifies instrument sensitivity prior to the

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

analysis of samples. Alternate subsequent CCVs between the mid and high calibration levels.

9.2.4.2 Target Analyte Acceptance Criteria - Calculate the concentration of each method analyte in the CCV. Each analyte fortified at a level less than or equal to the MRL must be within 50–150% of the true value. The concentration of the analytes in CCVs fortified at all other levels must be within 70–130%.

9.2.4.3 If the CCV fails, determine the nature of the failure (bias high or low). Attempt to determine the root cause of the failure prior reanalyzing the CCV. Potential causes for CCV failures could include, but are not limited to, poor standard introduction into the analytical system, or standard degradation. If the second analysis of the CCV passes criteria, the analysis of the sequence may proceed from the original point of failure. If the second CCV fails, the analytical sequence should be aborted, and the appropriate remedial actions taken. Such actions may include, but are not limited to, recalibrating the analytical system, or preparing new CCV standards. Any field samples analyzed since the last acceptable CCV that are still within holding time must be reanalyzed after an acceptable calibration has been restored.

9.3 Extract Preparation

This method may be performed manually or by using an automatic sample extraction procedure. Pace® Analytical Services, LLC. – Ormond Beach has the ability to perform the extraction using either the manual or automated procedures outlined below. When the automated procedure is used, the method blank (LRB/MB) will be rotated among the ports to ensure that all the valves and tubing meet the method blank requirements

9.3.1 Homogenization and Subsampling

Some of the PFAS adsorb to surfaces, including polypropylene. During extraction, sample bottles must be rinsed therefore subsampling is not permitted. The entire volume of each sample must be extracted. Duplicate samples cannot be combined to meet volume requirements.

9.3.2 Sample Volume

Sample volume is determined gravimetrically. The full sample bottle is weighed prior to extraction and the empty bottle is weighted after extraction. The sample volume is the difference between the full and empty bottle weights. Sample density is assumed at 1 g/mL. Some of the PFAS adsorb to surfaces, thus the sample may not be transferred to a graduated cylinder for volume measurement.

Batch QC is made by adding 250mL of LCMS grade water to pre-preserved sample bottles. Measure the 250mL volume using a PPE graduated cylinder (that has been verified) and pour into the pre-preserved bottles. Shake until preservative is completely dissolved.

9.3.3 Verify Preservation

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

Verify the pH of the samples, MB, and LCS/LCSD and document in the prep log. Notify the applicable Project Manager of any samples that are outside of pH 7.0 ± 0.5 .

Verify the absence of free chlorine in the samples, MB, and LCS/LCSD. Document the information in the prep log. Notify the applicable Project Manager of any samples that have Chlorine detections ≥ 0.01 mg/L.

9.3.4 Fortify QC Samples

Add a 10 μ L aliquot of the 537.1 surrogate solution to all samples and QC. Cap and invert to mix.

LCS and MS QC are extracted at varying concentrations of target analytes. A low level LCS (SRM) is spiked with 25 μ L of 537.1 Spike and is extracted with every batch. Each batch also contains a rotation of a mid or high level LCS and a low or mid-level MS/MSD. If insufficient sample is available, an LCSD at the level of the rotating LCS may be used. The low-level MS/MSD is spiked with 25 μ L of the 537.1 spike and the mid-level LCS/MS/MSD is spiked with 100 μ L of the 537.1 spike and high level LCS/MS/MSD is spiked with 20 μ L of the 537.1 high spike.

9.3.5 Extraction Procedure for Semiautomatic Extraction System

Do NOT allow cartridge packing material to go dry during any of the conditioning steps. Rinse each cartridge with 5 mL of methanol. Drain down to a layer of solvent above the packaging material. Repeat the methanol rinse two more times with the same volume, do not allow the cartridge packing material to go dry. Next, rinse each cartridge with 5 mL of reagent water, without allowing the water to drop below the top edge of the packing. Repeat the water rinse two more times with the same volume, do not allow the cartridge packing material to go dry. If the cartridge goes dry during the conditioning phase, the conditioning must be started over. Add 4-5 mL of reagent water to each cartridge, attach the sample transfer tubes, turn on the vacuum, and begin adding sample to the cartridge.

9.3.5.1 Rotate QC samples (MB and LCS) between extraction locations from one extraction batch to the next and record the MB location in the prep log. This is to ensure that the QC is not performed in dedicated positions on the extraction apparatus.

9.3.6 Sample Extraction

Adjust the vacuum so that the approximate flow rate is 10-15 mL/min. Do not allow the cartridge to go dry before all the sample has passed through.

9.3.7 Sample Bottle and Cartridge Rinse

After the entire sample has passed through the cartridge, rinse the sample bottles with two 7.5 mL aliquots of reagent water and draw each aliquot through the sample transfer tubes and the cartridges. Draw air or nitrogen through the cartridge for 5 min at high vacuum (10-15 in. Hg).

9.3.8 Sample Bottle and Cartridge Elution

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

Turn off and release the vacuum. Lift the extraction manifold top and insert a rack with collection tubes into the extraction tank to collect the extracts as they are eluted from the cartridges. Rinse the sample bottles with 4 mL of methanol and elute the analytes from the cartridges by pulling the 4 mL of methanol through the sample transfer tubes and the cartridges. Use a low vacuum such that the solvent exits the cartridge in a dropwise fashion. Repeat sample bottle rinse and cartridge elution with a second 4-mL aliquot of methanol. Repeat sample bottle rinse and cartridge elution with a final 4-mL aliquot of methanol.

9.3.9 Extraction Procedure for Automated Extraction System

Once all bottles have been prepared for extraction, the inline filters and cleaning bottles are put on the system. Once this is finished the “clean system” step is selected on the automatic extraction unit. The cleaning bottles are then taken off and the sample bottles and QC bottles are placed on the units. Once finished the 537 method is selected on the units (example method Appendix D).

Note: Any troubleshooting for these units are done by referring to the Manual located at the instrument. If this does not solve the issue Promochrom is contacted.

9.3.10 Extract Concentration

The extract is then put into the N-Evap and the settings are set at no higher than 65°C and approximately 13 psi until no solvent remains. Take the samples off the N-Evap and add 990 µL of 537.1 final solvent (96:4 MeOH: reagent water) and 10 µL of 537-IS to the dried extract (for a total final volume of 1 mL), vortex for 20 seconds, and cap. The extract is now ready for analysis.

9.4 Analysis

9.4.1 Begin analyzing Field Samples, including QC samples, at their appropriate frequency by injecting the same size aliquots under the same conditions used to analyze the CAL standards.

9.4.2 At the conclusion of data acquisition, use the same software that was used in the calibration procedure to identify peaks of interest in predetermined retention time windows.

9.4.3 Example Analytical Sequence

PAF Check
PFOA Technical
CAL1
CAL2
CAL3
CAL4
CAL5
CAL6
CAL7
CAL8
CCV-Low/CRDL
ICV/QCS

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

Extraction Batch QC
10 Field samples
CCV-mid
10 Field samples
CCV-high

10.0 DATA ANALYSIS AND CALCULATIONS

10.1 Qualitative Identification

Initial determination of the retention time (RT) window is accomplished by applying a width that is applicable to encompass all method criteria for peak identification and quantitation. The ICAL standards are used to identify the retention times for all analytes with the experience of the analyst determining the appropriate RT window. Refer to Appendix B for RT windows, frequency, acceptance criteria, and corrective actions.

Verification of the RT is accomplished daily by using the PAF Check Standard to confirm the SRM windows in the acquisition method satisfactorily scans the target compound peaks prior to initiation of the calibration curve. See Appendix B for more information.

10.1.1 Use the data system software to examine the ion abundances of the peaks in the chromatogram. Identify an analyte by comparison of its retention time with that of the corresponding method analyte peak in a reference standard. Comparison of the MS/MS mass spectra are not particularly useful given the limited ± 0.5 dalton mass range around a single product ion for each method analyte.

10.1.2 Review the chromatogram for any incorrect peak identification or poor integration.

10.2 Manual Integration

Manual changes to automated integration are called manual integration. Manual integration is sometimes necessary to correct inaccurate automated integrations but must never be used to meet QC criteria or to substitute for proper instrument maintenance and/or method set-up. To assure that all manual integrations are performed consistently and are ethically justified, all manual integrations must be performed, reviewed, and recorded in accordance with Pace® Analytical Services – Corporate SOP ENV-SOP-CORQ-0006, Manual Integration, or equivalent replacement.

10.3 Quantitative Identification

PFHxS, PFOS, NMeFOSAA, and NEtFOSAA have multiple chromatographic peaks using the LC conditions in Appendix C due to the linear and branch isomers of these compounds. The areas of all the linear and branched isomer peaks observed in the CAL standards for each of these analytes must be summed and the concentrations reported as a total for each of these analytes. Peaks in samples that elute prior to the branched isomers peaks present in the calibration standards are not included in the sample area response.

10.3.1 Peaks that are consistent with branched isomers have been observed with other target analytes, in particular PFOA and PFHpA. Per an EPA technical advisory released in September 2016, PFOA is calibrated via the linear isomer but both the linear and branched isomers are to be quantitated against that curve. A standard

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

prepared from technical grade PFOA must be run with every calibration to verify that the software is correctly integrating both the linear and branched isomers.

- 10.3.2** The analyst must not extrapolate beyond the established calibration range. The extract must be diluted with 96:4 (v/v) methanol:water solution. The dilution solvent must contain the internal standards matching the concentration in the undiluted extract. Do not report surrogate recoveries in the diluted extract. Report surrogate recoveries from the undiluted sample. Acceptable SUR performance should be determined from the undiluted sample extract. The resulting data should be documented as a dilution, with an increased MRL.

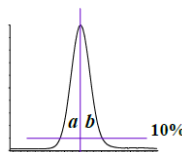
10.4 Calculations

Calculations must utilize all available digits of precision, but final reported concentrations should be rounded to an appropriate number of significant figures (one digit of uncertainty), typically two, and not more than three significant figures.

Refer to the laboratory Quality Assurance Manual and to Pace® Analytical Services – Florida SOP ENV-SOP-ORB1-0118, *Lab Calculations*, or equivalent replacement, for equations used to perform common calculations.

10.4.1 Peak Asymmetry Factor (A_s):

$$A_s = \frac{b}{a}$$



Where: A_s = Peak asymmetry factor

b = Width of the back half of the peak measured (at 10% peak height) from the trailing edge of the peak to a line dropped perpendicularly from the peak apex

a = The width of the front half of the peak measured (at 10% peak height) from the leading edge of the peak to a line dropped perpendicularly from the apex.

10.4.2 Half Range for the prediction interval of results (HR_{PIR}):

$$HR_{PIR} = 3.963S$$

Where: S = Standard deviation.

3.963 = Constant value for seven replicates.

10.4.3 Upper and Lower Limits for the Prediction Interval of Result ($PIR = \text{Mean} + HR_{PIR}$):

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

The Upper PIR Limit must be $\leq 150\%$ recovery.

$$\frac{\text{Mean} + HR_{PIR}}{\text{FortifiedConcentration}} \times 100\% \leq 150\%$$

The Lower PIR Limit must be $\geq 50\%$ recovery.

$$\frac{\text{Mean} - HR_{PIR}}{\text{FortifiedConcentration}} \times 100\% \geq 50\%$$

11.0 QUALITY CONTROL AND METHOD PERFORMANCE

11.1 Quality Control

Refer to Appendix B for Quality Control frequency, acceptance criteria and corrective action.

11.2 Method Performance

11.2.1 Method Validation

Refer to Pace® Analytical Services – Corporate SOP ENV-SOP-CORQ-0011 *Method Validation and Instrument Verification*, or equivalent replacement, for these procedures.

11.2.1.1 Method Detection Limit (MDL) Study

Detection limits (DL) and limits of quantitation (LOQ) are established at initial method setup and verified on an on-going basis thereafter. Refer to Pace® Analytical Services – Florida SOP ENV-SOP-ORB1-0124, *Determination of Limit of Detection and Limit of Quantitation*, or equivalent replacement, for these procedures.

Note: If an MRL confirmation data set meets these requirements, a DL may be calculated from the MRL confirmation data, and no additional analyses are necessary.

11.2.1.2 Minimum Reporting Level (MRL) Confirmation

Establish a target concentration for the MRL based on the intended use of the method. In some cases, the MRL may be dictated by USEPA or another regulatory body. Establish an Initial Calibration following the procedure outlined in Section 9.2. The lowest calibration standard used to establish the Initial Calibration (as well as the low-level CCV) must be at or below the concentration of the MRL. Establishing the MRL concentration too low may cause repeated failure of ongoing QC requirements. Validate the MRL following the procedure outlined below.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

11.2.1.2.1 Fortify, extract, and analyze seven replicate LFBs at the proposed MRL concentration. These LFBs must contain all method preservatives described in Section 6.0. Calculate the mean (Mean) and standard deviation for these replicates.

11.2.1.2.2 Determine the Half Range for the Prediction Interval of Results (HRPIR) using the equations in Section 10.4. Confirm that the upper and lower limits meet the upper and lower recovery limits in Appendix B.

11.2.2 Performance Evaluation (PE) Samples

Periodic performance evaluation (PE) samples are analyzed in accordance with Pace® Analytical Services – Florida SOP ENV-POL-CORQ-0002, *PT Policy*, or equivalent replacement.

11.3 Analyst Qualifications and Training

Employees that perform any step of this procedure must have a completed Read and Acknowledgment Statement for this version of the SOP in their training record. In addition, prior to unsupervised (independent) work on any client sample, analysts that prepare or analyze samples must have successful initial demonstration of capability (IDOC) and must successfully demonstrate on-going proficiency on an annual basis. Successful means the initial and on-going DOC met criteria, documentation of the DOC is complete, and the DOC record is in the employee's training file. Refer to Pace® Analytical Services – Florida SOP ENV-SOP-ORB1-0113, *Orientation and Training Procedures*, or equivalent replacement, for more information.

11.3.1 Initial Demonstration of Capability (IDC or IDOC): The IDC must be successfully performed prior to analyzing any Field Samples. Prior to conducting the IDC, the analyst must first generate an acceptable initial calibration and ICV following the procedure outlined in Section 9.2.

11.3.2 Initial Demonstration of Low Background: Before any samples are analyzed, or any time a new lot or brand of SPE materials or solvents are received from a supplier, it must be demonstrated that a MB is reasonably free of any contamination that would prevent the identification or quantitation of target analytes, the IS, or SUR.

11.3.2.1 If the background contamination from SPE (refer to Section 3.0) is sufficient to prevent accurate and precise measurements, the condition must be corrected before proceeding with the IDP.

11.3.2.2 Background contamination (see Section 3.0) must be reduced to an acceptable level before proceeding with the IDP. See Appendix B for Method Blank acceptance criteria. If background contamination is an on-going or intermittent issue, the analyst must not attempt to verify an MRL less than the mean MB concentration + 3σ , or three times the mean MB concentration, whichever is greater.

Note: Although quantitative data below the MRL may not be reliably accurate enough for data reporting, such data are useful in determining

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

an MRL cut off for the method analyte if it is found in the MB. Therefore, blank contamination levels may be estimated by extrapolation, when the concentration is below the lowest calibration standard. This also applies to estimating MB concentrations for daily low background verification.

11.3.2.3 Initial Demonstration of Precision (IDP): Prepare, extract, and analyze four replicate LCSs fortified near the midrange of the initial calibration curve according to the procedure described in Section 9.3. Sample preservatives as described in Section 6.0 must be added to these samples. Refer to Appendix B for frequency, acceptance criteria, and corrective actions.

11.3.2.4 Initial Demonstration of Accuracy (IDA) – Using the same set of replicate data generated for the IDP, calculate average recovery. Refer to Appendix B for frequency, acceptance criteria, and corrective actions.

12.0 DATA REVIEW AND CORRECTIVE ACTION

12.1 Data Review

The data review process of Pace® Analytical Services includes a series of checks performed at different stages of the process by different people to ensure that SOPs were followed, the analytical record is complete, and properly documented, QC criteria were met, proper corrective actions were taken for QC failure and other nonconformance(s), and test results are reported with proper qualification, when necessary.

The review and checks that are performed by the employee performing the task is called primary review.

All data and test results are also peer reviewed.

This process, known as secondary review is performed to verify SOPs were followed, that calibration, instrument performance, and QC criteria were met and/or proper corrective actions were taken, qualitative ID and quantitative measurement is accurate, all manual integrations are justified and documented, and approved in accordance with the Pace® Analytical Services SOP for manual integration, calculations are correct, the analytical record is complete and traceable, and that results are properly qualified.

Lastly, a third-level review, called a completeness check, is performed by reporting or project management staff to verify the test report is complete.

Refer to laboratory SOP ENV-SOP-ORB1-0119, *Data Review Process* for specific instructions and requirements for each step of the data review process.

12.2 Corrective Action

Corrective action is required when QC or sample results are not within acceptance criteria.

Refer to Appendix B for a complete summary of QC, acceptance criteria, and recommended corrective actions for QC associated with this test method.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

If corrective action is not taken or was not successful, the decision/outcome must be documented in the analytical record. The primary analyst has primary responsibility for taking corrective action when QA/QC criteria are not met. Secondary data reviewers must verify that appropriate action was taken and/or that results reported with QC failure are properly qualified.

Corrective action is also required when carryover is suspected and when results are over range.

Samples analyzed after a high concentration sample must be checked for carryover and reanalyzed if carryover is suspected. Carryover is usually indicated by low concentration detects of the analyte in successive samples analyzed after the high concentration sample.

Sample results at concentrations above the upper limit of quantitation must be diluted and reanalyzed. The result in the diluted samples should be within the upper half of the calibration range. Results less than the mid-range of the calibration indicate the sample was over diluted and analysis should be repeated with a lower level of dilution. If dilution is not performed, any result reported above the upper range is considered a qualitative measurement and must be qualified as an estimated value.

13.0 POLLUTION PREVENTION AND WASTE MANAGEMENT

Pace® proactively seeks ways to minimize waste generated during work processes. Some examples of pollution prevention include but are not limited to reduced solvent extraction, solvent capture, use of reusable cycletainers for solvent management, and real-time purchasing.

The EPA requires that laboratory waste management practices comply with all applicable federal and state laws and regulations. Excess reagents, samples, and method process wastes are characterized and disposed of in an acceptable manner in accordance with the Pace® Chemical Hygiene Plan / Safety Manual. Refer to this manual for these procedures.

14.0 MODIFICATIONS

A modification is a change to a reference test method made by the laboratory. For example, changes in stoichiometry, technology, quantitation ions, reagent or solvent volumes, reducing digestion or extraction times, instrument runtimes, etc. are all examples of modifications. Refer to Pace® Analytical Services – Corporate SOP ENV-SOP-CORQ-0011, *Method Validation and Instrument Verification*, or equivalent replacement, for the conditions under which the procedures in test method SOPs may be modified and for the procedure and document requirements.

The analyst is permitted to modify LC columns, LC conditions, evaporation techniques, internal standards or surrogate standards, and MS and MS/MS conditions. Each time such method modifications are made, the analyst must repeat the procedures of the IDC. Modifications to LC conditions should still produce conditions such that co-elution of the method analytes is minimized to reduce the probability of suppression/enhancement effects.

14.1 Approved Modifications

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

Method Reference	Method Procedure	SOP Reference	Modified Procedure	Reason for Deviation
EPA 537.1 Section 11.5	Concentrate the extract at 60-65°C	Section 9.3.10	Concentrate the extract at 55-60°C	Lower temperature is providing better recoveries of compounds and surrogates

15.0 RESPONSIBILITIES

- PAS employees that perform any part this procedure in their work activities must have a signed Read and Acknowledgement Statement (R&A) in their training file for the version(s) of the SOP that were in effect during the time the employee performed the activity.
- PAS supervisors and managers however named, are responsible for training employees on the procedures in this SOP, implementing the SOP in the work area, and monitoring on-going adherence to the SOP in the work area(s) they oversee.
- PAS employees are responsible for following the procedures in this SOP. Unauthorized deviations or departures from this SOP are not allowed except with documented approval from the local QM and only when those deviations do not violate the Pace® Code of Ethics or Professional Conduct (COR-POL-0004) or associated policy and procedure(s). Hand-edits or manual changes to the SOP are not permitted. If a change is desired or necessary, Pace® employees must follow the procedures for document revision specified in corporate SOPs ENV-SOP-CORQ-0015, Document Management and ENV-SOP-CORQ-0016, SOP for Creation of SOP and SWI.

16.0 ATTACHMENTS

- Appendix A: Target Analyte List and Routine LOQ
- Appendix B: QC Summary
- Appendix C: Routine Instrument Operating Conditions
- Appendix D: Example Automated Extractor Run
- Appendix E: Explanation of each step on the Automated Extractors

17.0 REFERENCES

- Pace® Quality Assurance Manual- most current version.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

- TNI Standard, "Volume 1 Management and Technical Requirements for Laboratories Performing Environmental Analysis", Revision 2.1, 2016.
- USEPA, "Manual for the Certification of Laboratories Analyzing Drinking Water"; Fifth Edition, January 2005.
- USEPA, "Supplement 1 to the Fifth Edition of the Manual for the Certification of Laboratories Analyzing Drinking Water"; June 2008
- USEPA, Method 537.1 Version 2.0, "Determination of Selected Per- and Polyfluorinated Alkyl Substances in Drinking water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)", March 2020
- USEPA, "UCMR5 Laboratory Approval Manual"; EPA 815-B-20-007; Office of Water: Cincinnati, OH, December 2020.
- ENV-SOP-CORQ-0006, *Manual Integration*, current version.
- ENV-SOP-CORQ-0011, *Method Validation*, current version.
- ENV-SOP-CORQ-0015, *Document Management*, current version.
- ENV-SOP-CORQ-0016, *SOP for SOP and SWI*, current version.
- ENV-TMP-CORQ-0007, *Quality Manual Template*, current version.
- COR-POL-0004, *Code of Ethics and Professional Conduct*, current version.
- COR-MAN-001, *Pace® Safety Manual*, current version.

18.0 REVISION HISTORY

Revisions Made from Prior Version

Section	Description of Change
ALL	Updated SOP Template
ALL	Removed page breaks
8.1	Added 'HPLC grade or equivalent' to Reagent Water and Methanol
9.3.10	Updated section
14.1	Added Approved Modifications

Document Succession: This version replaces the following documents:

Document Number & Version	Document Title	Effective Date
ENV-SOP-ORB1-0175 v03	EPA 537.1	06/05/23

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

Appendix A: Target Analyte List and Routine LOQ

Table 1: Routine Analyte List and Limits of Quantitation (LOQ)¹

Analyte	Water (µg/L)	UCMR5 MRL (µg/L)
PFOS (Perfluorooctanesulfonic acid)	0.004	N/A
PFOA (Perfluorooctanoic acid)	0.004	N/A
PFHxS (Perfluorohexanesulfonic acid)	0.004	N/A
PFHpA (Perfluoroheptanoic acid)	0.004	N/A
PFNA (Perfluorononanoic acid)	0.004	N/A
PFBS (Perfluorobutanesulfonic acid)	0.004	N/A
PFUnA (Perfluoroundecanoic acid)	0.004	N/A
PFDoA (Perfluorododecanoic acid)	0.004	N/A
PFDA (Perfluorodecanoic acid)	0.004	N/A
PFHxA (Perfluorohexanoic acid)	0.004	N/A
NEtFOSAA (N-ethyl perfluorooctanesulfonamidoacetic acid)	0.004	0.005
NMeFOSAA (N-methyl perfluorooctanesulfonamidoacetic acid)	0.004	0.006
PFTeDA (Perfluorotetradecanoic acid)	0.004	0.008
PFTTrDA (Perfluorotridecanoic acid)	0.004	0.007
HFPO-DA (Hexafluoropropylene oxide dimer acid)	0.004	N/A
ADONA (4,8-dioxa-3H-perfluorononanoic acid)	0.004	N/A
9Cl-PF3ONS (9-chlorohexadecafluoro-3-oxanone-1-sulfonic acid)	0.004	N/A
11Cl-PF3OUdS (11-chloroeicosafluoro-3-oxaundecane-1-sulfonic acid)	0.004	N/A

¹Values in place as of effective date of this SOP. LOQ are subject to change. For the most up to date LOQ, refer to the LIMS or contact the laboratory.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

Appendix B: QC Summary

QC Item	Frequency	Acceptance Criteria	Corrective Action	Qualification (non-UCMR)	UCMR/DoD
Tune Standard	Optimize the product ion for each method analyte if calibration criteria cannot be met and after any major modification or maintenance that breaks the MS/MS vacuum.	Follow the manufacturer's instructions. The vendor software will indicate a successful tune. See Section 0.	Identify and correct source of problem, repeat.	NA	NA
RT Window Position	Once per ICAL and at the beginning of the analytical window.	Position is set using the PAF Check Standard.	NA	NA	NA
RT Window Study	At method set-up and after major instrument maintenance that impacts the RT of the analytes.	Window is ± 0.25 minutes. (Values in place as of effective date of this SOP. RT windows are subject to change. For the most up to date values, contact the laboratory.)	NA	NA	NA
Minimum Reporting Limit Confirmation (MRL)	Fortify, extract, and analyze 7 replicates at the proposed MRL concentration. Calculate the mean, standard deviation and HR_{PIR} . Confirm that the upper and lower limits for the Prediction Interval of Result meet the recovery criteria.	Upper PIR $\leq 150\%$ Lower PIR $\geq 50\%$ The MRL is validated if both the Upper and Lower PIR Limits meet the above criteria.	Identify and correct source of problem, repeat. If still unsuccessful, the MRL has been set too low and must be re-evaluated at a higher concentration.	NA	NA
Peak Asymmetry Factor (PAF)	During the IDOC, with each ICAL, and each time chromatographic changes are made that may affect peak shape.	The first two eluting peaks at a mid-level standard must calculate within the 0.8-1.5 range.	Change initial mobile phase conditions to higher aqueous content, repeat.	NA	NA
Initial Calibration (ICAL)	Prior to sample analysis at instrument set up, after CCV failure, and after major maintenance (eg. cleaning the ion source, replacing filament assemblies, or replacing the LC column).	Must meet one of curve fit options presented in Section 9.0. Calculate the concentration of each analyte as an unknown against its calibration curve. Points $\leq$ MRL: $\pm 50\%$ of the true value	Identify and correct source of problem, repeat. Restricting the range of calibration or selecting an alternate method of calibration may be necessary.	None. Do not proceed with analysis.	None. Do not proceed with analysis.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

QC Item	Frequency	Acceptance Criteria	Corrective Action	Qualification (non-UCMR)	UCMR/DoD
	For any curve fit other than Average RF (RSD), curve must also pass RE test at the low and midpoint calibration standard.	Points > MRL: $\pm 30\%$ of the true value Surrogate: $\pm 30\%$ of the true value Internal standard: $\pm 50\%$ of the ICAL average			
Initial Calibration Verification (ICV or QCS)	With each ICAL, IDOC, and at least quarterly.	All analytes must be within $\pm 30\%$ of the true value (%R).	Identify and correct source of problem, re-analyze. If repeat failure, repeat ICAL. Analysis may proceed if it can be demonstrated that the ICV exceedance has no impact on analytical measurements. For example, the ICV %R is high, CCV is within criteria, and the analyte is not detected in sample(s).	Qualify analytes with ICV out of criteria.	UCMR5: Do not report failed QCS to SDWARS
CCV-Low	Daily, before each analysis batch DoD – CCV must be > Low Cal and $\leq$ midpoint Cal	All analytes must be within 50-150% of the true value. (%R)	Identify source of problem, re-analyze. If repeat failure, repeat ICAL. Analysis may proceed if the CCV %R is high and the analyte is not detected in sample(s). DoD: Reanalysis (2 consecutive passing CCVs) or corrective action must be taken.	Qualify analytes with CCV out of criteria.	UCMR5: Data may be reported only if the CCV %R is high and the analyte is not detected in sample(s). No other qualification is allowed. DoD: qualification is allowed only if samples cannot be reanalyzed.
Continuing Calibration Verification (CCV/CCC)	Daily, after every 10 field samples, and at end of analytical window. DoD – CCV must be > Low Cal and $\leq$ midpoint Cal	All analytes must be within 70-130% of the true value. (%R)	Identify source of problem, re-analyze. If repeat failure, repeat ICAL. Analysis may proceed if the CCV %R is high and the analyte is not detected in sample(s). DoD: Reanalysis (2 consecutive passing CCVs) or corrective action must be taken.	Qualify analytes with CCV out of criteria.	UCMR5: Data may be reported only if the CCV %R is high and the analyte is not detected in sample(s). No other qualification is allowed. DoD: qualification is allowed only if samples

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

QC Item	Frequency	Acceptance Criteria	Corrective Action	Qualification (non-UCMR)	UCMR/DoD
					cannot be reanalyzed.
Internal Standards (IS)	Every field sample, standard and QC sample	Peak area count must be within $\pm 50\%$ of the average peak area calculated during the ICAL and 70-140% of the previous CCV.	Troubleshoot instrument performance. Reanalyze samples.	Qualify outages and explain in case narrative.	Data associated with failing IS in samples or QC may not be reported.
Surrogate (SUR)	Every field sample, standard and QC sample	SUR must be within $\pm 30\%$ of the true value (%R).	Rerun sample. If failure confirms and capping CCV is within criteria re-extract if in hold or report with flags. If surrogates exceed maximum criteria BDL samples may be reported. Surrogate compounds that are not associated with the class of analytes being reported may be excluded from analysis and reporting.	Quality samples with surrogates out of criteria.	Data associated with failing SUR in samples or QC may not be reported.
Reporting Limit Verification (Low-LCS, SRM)	With each extraction batch of no more than 20 samples. Spiked at 1-2 times the MRL.	%R must be within $\pm 50\%$ of the true value.	Reanalyze and/or re-prep batch of samples with new LLCS. Samples may be reported if recovery is high and the analyte is not detected in sample(s).	Qualify samples if out of criteria.	Data associated with any LCS failure may not be reported.
Method Blank (MB / LRB)	With each batch of no more than 20 samples and with each IDOC study. Treated exactly as a sample including addition of sample preservatives.	Analyte must be $< \text{MDL}$ or $< \frac{1}{3}$ of the MRL depending on reporting requirements. DoD: Analyte must be $< \frac{1}{2}$ LOQ, $< 1/10$ sample result, or $< 1/10^{\text{th}}$ regulatory limit, whichever is greater Confirm that potential background contaminants are not interfering with the identification or quantitation of method analytes. Background from contaminants that interfere with the measurement of method analytes must be $< \frac{1}{3}$ of the MRL. Blank	Reanalyze and/or re-prep batch of samples with new MB unless they meet exceptions below: 1) If sample ND, report sample without qualification 2) If sample result $> 10\times$ MB detects and sample cannot be reanalyzed, report sample with appropriate qualifier indicating blank contamination. 3) If sample result $< 10\times$ MB detects, report sample with appropriate qualifier to indicate an estimated value. Client must be alerted and authorize this condition.	Qualify samples with detections described in 2) and 3). If the analyte is detected $> \text{MDL}$ in the MB during the IDOC study, do not qualify. Repeat the study.	UCMR5: Data may not be reported if detections in MB are $> 1/3$ UCMR5 MRL, regardless of sample result DoD: results can only be reported if insufficient sample remains to reprep.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

QC Item	Frequency	Acceptance Criteria	Corrective Action	Qualification (non-UCMR)	UCMR/DoD
		contamination may be estimated by extrapolation if the concentration is below the lowest calibration standard.			
Laboratory Control Sample (LCS / LFB)	With each batch of no more than 20 samples. Rotate the fortified concentrations of the LCS between mid and high amounts based on the ICAL. Treated exactly as a sample including addition of sample preservatives. DoD: spiked at or below midpoint of Cal	%R must be within $\pm 30\%$ of the true value.	Reanalyze and/or re-prepare batch of samples with new LCS. Samples may be reported if recovery is high and the analyte is not detected in sample(s). DoD: Corrective action and reanalysis is required if an analyte fails 2 of 3 consecutive LCS.	Qualify samples with LCS and/or LCSD out of criteria.	UCMR5: NA, only low-LCS spiked at the MRL is required.
Matrix Spike/Spike Duplicate (MS/MSD) (LFSD/LFSDM)	With each batch of no more than 20 samples. Rotate through low (1-2 times the MRL), mid, and high level spike concentrations. Spike at $\geq$ the matrix background concentration. See Duplicate for more MSD information.	Low level spike concentration: %R must be within $\pm 50\%$ of the true value. Mid and High level spike concentration: %R must be within $\pm 30\%$ of the true value.	If MB and LCS meet criteria, then samples may be reported with qualification.	Qualify target analytes on the parent sample.	UCMR5: Recovery criteria do not apply. Do not upload MS/MSD with failing SUR or IS to SDWARS.
Sample Duplicate (DUP)	With each batch of no more than 20 samples. May be a Field Duplicate (FD), LCSD, or MSD. An MSD should be analyzed rather than a FD if method analytes are not routinely observed in field samples. An LCSD should be analyzed when there is not sufficient sample volume for an MSD or FD.	Low level spike (LCSD/MSD) or background (FD) concentration: RPD must be $\leq 50\%$. Mid or High level spike (LCSD/MSD) or background (FD) concentration: RPD must be $\leq 30\%$.	If MB and LCS meet criteria, then samples may be reported with qualification.	Qualify target analytes on the parent sample.	UCMR5: Only MS/MSD used. Precision criteria do not apply. Do not upload MS/MSD with failing IDA/IPS to SDWARS.
Field Blank (FB/FRB)	A field reagent Blank must be included and collected from each sample site. Analyze when an associated field sample contains	All method analytes must be $\leq \frac{1}{2}$ the MRL.	All associated field samples are invalid and must be recollected and reanalyzed unless the	Qualify the associated samples. If the sample result is $< 10\times$ the FRB result	Required for each field sample. Associated sample must be recollected

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

QC Item	Frequency	Acceptance Criteria	Corrective Action	Qualification (non-UCMR)	UCMR/DoD
	any method analyte $\geq$ MRL.		sample results are $>10\times$ the FRB results.	report and narrate only upon client request.	for analytes $>1/3$ UCMR5 MRL.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

Appendix C: Routine Instrument Operating Conditions

Injector	Syringe Size	100 µL		
	Sample Loop vol.	50 µL		
	Injection Volume:	10 µL		
	Needle Wash 1	10% Methanol in water		
	Needle Wash 2	100% Methanol		
Pump	Flow rate	400 uL/min		
	Flow method	Gradient		
	Mobile Phase 1	537 Aqueous Mobile Phase		
	Mobile Phase 2	LCMS Methanol/Acetonitrile 80:20		
	Gradient Program	Time	% Mobile Phase 1	% Mobile Phase 2
		Initial	95	5
		0.3	95	5
		9.00	5	95
		13.00	5	95
		13.10	95	5
		17.00	95	5
Column	Type:	Thermo Accucore C18		
	Model Number:	17126-153030		
	Running temp	40 C		
	Length:	100 mm		
	Diameter:	3.0 mm		
	Particle Size	2.6 um		
	Guard Frit	IDEX 2um SS A-101X		
Nominal Tune Values	Capillary Temp	300 C		
	Vaporizer Temp	300 C		
	Sheath Gas Pressure	40 Arb		
	Aux Gas Pressure	10 Arb		
Values may change with source maintenance	Ion Sweep Gas Pressure	1.0 Arb		
	Spray Voltage +	3500 V		
	Spray Voltage -	3000 V		
	Discharge Current +	4.0 uA		

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

Appendix C: Routine Instrument Operating Conditions (Continued)

Analyte	Precursor Ion (m/z)	Product Ion (m/z)	S-Lens	Collision Energy (v)	Start/Stop Window (min)
PFOS	499	80	113	49	100.0-11.00
PFOA	413	369	65	12	10.00-10.50
PFHxS	399	80	127	41	9.50-10.50
PFHpA	363	319	60	12	9.50-10.50
PFNA	463	419	70	13	10.00-11.00
PFBS	299	80	78	39	8.50-9.50
PFUnA	563	519	55	11	10.60-11.40
PFDoA	613	569	97	20	10.80-11.50
PFDA	513	469	69	14	10.30-11.30
PFHxA	313	269	40	10	9.00-10.10
NEtFOSAA	584	419	130	17	10.60-11.40
NMeFOSAA	570	419	108	18	10.40-11.30
PFTeDA	713	669	110	20	11.10-12.00
PFTrDA	663	619	86	18	11.00-12.00
HFPO-DA	285	169	40	6	9.00-10.00
ADONA	377	251	66	12	9.50-10.50
9Cl-PF3ONS	531	351	120	23	10.30-11.10
11Cl-PF3OUdS	631	451	140	26	10.40-11.50
¹³ C ₂ PFOA (IS)	415	370	63	12	10.00-10.50
¹³ C ₄ PFOS (IS)	503	80	138	46	10.00-11.00
NMeFOSAA-d ₃ (IS)	573	419	108	18	10.40-11.30
¹³ C ₂ -PFHxA (S)	315	270	40	10	8.75-10.10
¹³ C ₂ -PFDA (S)	515	470	69	14	10.30-11.30
NEtFOSAA-d ₅ (S)	589	419	108	18	10.40-11.30
¹³ C ₃ -HFPO-DA (S)	287	169	32	6	9.00-10.00

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

Appendix D: Example Automated Extractor Run.

(This is just an example and can be changed to meet updated extraction practices)

- Elute W2, Solvent 1, 5, 15.0mLs
- Elute W1, Solvent 2, 10, 18.0mLs
- Elute W1, Solvent 2, 10, 5.0mLs
- Add Samp W1, Sample, 10, 300mLs
- Rinse W1, Solvent 2, 10, 7.5mLs
- Rinse W1, Solvent 2, 10, 7.5mLs
- Shake, Time based 30 s
- Air-Purge W1, Air, 10, 5.0
- Add Samp W1, Sample, 10, 4.5mLs
- Blow N2, Time based, 5 min
- Rinse 1, Solvent 1, 5, 4.0mLs
- Rinse 1, Solvent 1, 5, 4.0mLs
- Rinse 1, Solvent 1, 5, 4.0mLs
- Shake, Time based, 10 s
- Collect 1, Sample, 5, 4.5mLs

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-ORB1-0175 v04_EPA 537.1
	Effective Date: 12/08/2023

Appendix E: Explanation of each step on the Automated Extractors

Prime – clean valve and pump with selected solvent(s) Solvent(s) are drawn through valves 1 and 2 into the pumps and then pushed out into the top waste instead of flowing through the SPE columns. This can be used to fill or clean solvent tubing.

Elute – pre-condition or wash cartridge with selected solvent(s) Solvent(s) are pushed through the SPE columns into bottom waste 1 or 2. The difference between Elute and Collect is that Collect will push solvents into Fraction 1 or Fraction 2

Add Sample – load sample onto SPE cartridge Sample(s) are drawn and pushed through the SPE columns and then into bottom waste 1 or 2

Collect – elute with selected solvent(s) and collect eluent in specified fraction tubes Solvent(s) are pushed through the SPE columns to elute analytes into Fraction 1 or Fraction 2. Samples can also be directly collected into Fraction 1 or Fraction 2. This is used when manual rinsing of sample containers is performed and the rinsate needs to be collected

Air-purge – use air to purge residual fluid through columns into fractions/waste or to mix sample Outside air is drawn in and then pushed into sample containers (eg. For mixing internal standards) or delivered through SPE columns into fractions or waste. Air-purge can also be used to mix sample after manually adding internal standards

Blow N2 – Blow nitrogen for a specified amount of time or until user manually triggers next step “Time based” option will turn on nitrogen blowing for the specified amount of time. “Manual resume” option will continue nitrogen blowing until the operator presses “start” to resume. A loud “beep” is triggered when using the “Manual resume” option to get the operator’s attention

Rinse – rinse sample containers and deliver rinsate into fraction tubes or waste

Clean – deliver selected solvent(s) into sample containers followed by optional air purge to clear solvent(s) from liquid lines

Shake – activate resonator for a specified amount of time or until user manually triggers next step

Wait – pause run for a specified amount of time or until user manually triggers next step

Pump-Cln – Rinses pump syringes and plungers with solvent(s) and delivers to fractions or waste

Boost – Moves liquid in the cartridge sorbent back and forth

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services



ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633

Effective Date: 10/14/2024

Management Approval:

Adam Haugerud Approved on 10/14/2024 9:50:27 AM

Aileen Stacks Approved on 10/14/2024 8:39:53 AM

David Randall Approved on 10/11/2024 2:57:32 PM

© 2024, Pace® Analytical Services, LLC (PAS)

1.0 SCOPE & APPLICATION

This standard operating procedure (SOP) specifies the laboratory procedure used by PAS-Minneapolis MN for the determination of the Per- and Polyfluoroalkyl Substances (PFAS) in various matrices using LC-MS/MS by EPA Test Method 1633.

1.1 Applicable Matrices

The matrices that can be tested using this SOP include:

Aqueous Matrices	Solid Matrices	Other Matrices
Non-Potable Water	Soil/Sediment	Biphasic (Aqueous/Solid)
Landfill Leachate	Biosolid	-
TCLP / SPLP Leachate	Tissue	-
Aqueous Film Forming Foam (AFFF)	Articles of Commerce	-
Fluorine-Free Foam (F3)	-	-

1.2 Routine Analyte List and Limits of Detection & Quantitation

This SOP uses abbreviations for the analyte name. A cross-reference of analyte name to CAS# and abbreviation is provided in Appendix A, Table 1.

The routine analyte list and limit of quantitation (LOQ) that can be determined by this SOP for each matrix is provided in Appendix A, Table 2.

Detection limits for each analyte are kept on file by the laboratory's Quality Department.

Unless otherwise specified, nominal limits of detection and the LOQ used during testing are adjusted on a per-sample basis to account for actual sample quantities used, percent moisture, and dilution factors.

1.3 Statutory and Program Specific Requirements

When statutory or regulatory program requirements for EPA Method 1633 differ from the procedures outlined in the main body of this SOP, the program specific requirements are called out in the relevant section of the SOP or are provided in appendices to the SOP. These requirements take precedence over the main SOP and must be adhered to when performing work under that jurisdiction, unless otherwise specified on a project-specific basis.

2.0 SUMMARY OF METHOD

Samples are prepared and extracted using solid phase extraction (SPE). Extracts are cleaned using carbon cleanup prior to analysis by LC-MS/MS in the multiple reaction monitoring (MRM) mode.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633 Effective Date: 10/14/2024

Sample concentrations are determined by isotope dilution or extracted internal standard quantification using isotopically labeled compounds added to the samples before extraction.

This test procedure is based on USEPA Method 1633, January 2024, and includes allowable modifications from the reference method to improve performance.

Refer to Section 14.0 for a detailed list of modifications and their rationale. Supporting documentation to demonstrate equivalency and comparability, when required, is maintained by the laboratory's Quality Department.

3.0 INTERFERENCES

Solvents, reagents, glassware, and other sample processing hardware may yield artifacts and elevated baselines causing misinterpretation of chromatograms. This SOP includes procedures for equipment cleaning to mitigate these interferences.

Interferences co-extracted from samples will vary considerably from source to source, depending on the diversity of the site being sampled. Interfering compounds may be present at concentrations several orders of magnitude higher than the native PFAS. Because low levels of PFAS are measured by this method, elimination of interferences using proper cleanup techniques is essential and these steps are incorporated into the procedural sections of this SOP.

The most frequently encountered interferences are fluoropolymers; however, bile salts (e.g., Taurodeoxycholic Acid [TDCA]) may be present in various matrices, including fish and wastewaters, and can interfere in the chromatography. For this reason, the laboratory procedure includes analysis of a standard (bile interference check) containing TDCA is required when establishing the initial chromatographic conditions and with each analytical sequence.

4.0 DEFINITIONS

The definitions of common laboratory terms are provided in the Glossary section of the Laboratory Quality Manual. Refer to the Quality Manual as needed. Key terms relevant to this procedure include:

- **Articles of Commerce:** Solid consumer and related product materials.
- **Extracted Internal Standard (EIS) Quantification:** The response of the target compound is compared to the response of the isotopically labeled analog of another compound with chemical and retention time similarities.
- **Isotope Dilution (ID) Quantitation:** A means of determining a naturally occurring (native) compound by reference to the same compound in which one or more atoms has been isotopically enriched. The labeled PFAS are spiked into each sample and allow identification and correction of the concentration of the native compounds in the analytical process.
- **Isotopically Labeled Compound:** An analog of a target analyte in the method which has been synthesized with one or more atoms in the structure replaced by a stable (non-radioactive) isotope of that atom. Common stable isotopes used are ¹³C (Carbon-13) or Deuterium (D or ²H). These labeled compounds do not occur in nature, so they can be used for isotope dilution quantitation or other method-specific purposes.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

- **PAS:** Acronym for Pace® Analytical Services, LLC.

5.0 HEALTH & SAFETY

Contact your supervisor or local safety coordinator with questions or concerns regarding safety protocol or safe handling procedures for this procedure.

The following sections provide general health and safety information about chemicals and materials that may be present in the laboratory.

- The toxicity or carcinogenicity of each chemical material used in the laboratory has not been fully established. Each chemical should be regarded as a potential health hazard and exposure to these compounds should be as low as reasonably achievable.
- The laboratory maintains documentation of hazard assessments and OSHA regulations regarding the safe handling of the chemicals specified in each method. Safety data sheets for all hazardous chemicals are available to all personnel. Employees must abide by the health, safety and environmental (EHS) policies and procedures specified in this SOP and in the Pace® Chemical Hygiene / Safety Manual (COR-MAN-0001).
- Personal protective equipment (PPE) such as safety glasses, gloves, and a laboratory coat must be worn in designated areas and while handling samples and chemical materials to protect against physical contact with samples that contain potentially hazardous chemicals and exposure to chemical materials used in the procedure.
- Concentrated corrosives present additional hazards and are damaging to skin and mucus membranes. For procedures that require use of acids, use acids in a fume hood whenever possible with PPE designed for handling these materials. If eye or skin contact occurs, flush with large volumes of water. When working with acids, always add acid to water to prevent violent reactions. For procedures that emit large volumes of solvents (evaporation/concentration processes), these activities must be performed in a fume hood or apparatus that reduces exposure.

6.0 SAMPLE COLLECTION, PRESERVATION, HOLDING TIME & STORAGE

6.1 Sample Collection & Bottle Kits

PAS does not offer sample collection services for this test method. The customer's sampling contractor should collect samples in accordance with a sampling plan and collection procedures appropriate to meet regulatory requirements and data quality objectives for the project.

Refer to Appendix A, Table 3, for sample collection, preservation, and holding time requirements for each matrix.

PAS will provide containers for sample collection per customer request. Refer to laboratory SOP ENV-SOP-MIN4-0009 *Bottle Kit Preparation*, or however named, for general instruction on bottle kit preparation and refer to Appendix A, Table 3, for sample collection, preservation, and holding time requirements for each matrix.

PFAS Bottle Kit Specifications:

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

- If the request includes containers for surface wipes, provide a copy of the Pace® PFAS Wipe Sampling Guide in each sampling kit.
- Kits for aqueous sample kits should include the instruction to fill the container to the gradation marked on the container, or to the shoulder of the container if no gradations are provided.
- If matrix spikes are requested (or required by program), provide additional containers for the collection of MS/MSD and remind the customer to designate the sample for use on the chain-of-custody (COC).

For example, if an MS/MSD is requested on an aqueous matrix, the customer must collect and provide six (6) 500 mL containers, two (2) each for the MS, MSD, and parent sample, plus one (1) 125 mL container for prescreening.

- **AFFF Concentrate:** Customers should notify PAS at the time of request for quote or sampling containers if the matrix of any sample is an AFFF concentrate. Due to the specific handling requirements needed for this matrix, pre-notification ensures that the laboratory is prepared to handle the sample properly to prevent the sample from contaminating the work area and/or other test samples.
- **DoD Projects:** Testing may only be done by DoD accredited laboratories. Ensure sample kits include additional containers to collect sample material for laboratory duplicates and/or matrix spikes as required by DoD QSM or as specified in the Project QAPP (Quality Assurance Project Plan).

6.2 Sample Receipt, Storage and Disposal

Laboratory sample receiving personnel check thermal preservation and compliance with the PAS Sample Acceptance Policy on receipt following the procedures specified in SOP ENV-SOP-MIN4-0008 *Sample Management*, or however named. Refer to the Laboratory Quality Manual for the PAS Sample Acceptance Policy.

Unless otherwise specified by contract, samples and extracts are retained for the timeframe specified in the Pace® standard terms and conditions then disposed of in accordance with Federal, State, and Local regulations.

7.0 EQUIPMENT & SUPPLIES

See Appendix A for a complete list of equipment and supplies needed to perform this procedure.

Substitution of instrumentation, equipment, and supplies is permissible only if the replacement meets the specifications outlined in the SOP. If an alternative item does not meet these specifications, it may impact the procedure's performance, necessitating an update and/or revalidation of the procedure.

Plastic containers must be used for all standard, sample, and extraction preparations due to potential adsorption of analytes onto glass. Do not substitute glass for plastic containers.

7.1 Suitability Checks

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Verify the suitability of SPE cartridges, solvents, cryovials, and autosampler vials on a lot basis by analysis of a method blank; maintain records of the verification for readily available historical access. See Section 9.1.2.1.

8.0 REAGENTS & STANDARDS

See Appendix B for a complete list of standards and reagents needed to perform this procedure and for formulations of standard solutions prepared by the laboratory.

Substitution of reagents and standards from those listed in Appendix B are permissible only if the replacement meets the specifications outlined in the SOP. If an alternative item does not meet these exact same specifications for purity, composition, or concentration, any formulation in this SOP must be modified to match the material used. If the substitution is permanent, the SOP and formulations in this SOP must be updated.

All purchased standard solutions must be received with a Certification of Analysis (COA). The COA must be maintained on-site in laboratory possession; access to a COA via the vendor's website is generally not acceptable to external regulatory bodies because the COA must be traceable to each lot received. Refer to laboratory SOP ENV-SOP-MIN4-0180 *Laboratory Procurement Procedures*, or however named, for instructions for receipt and traceability.

9.0 PROCEDURE

9.1 Equipment & Instrument Setup

9.1.1 Support Equipment

9.1.1.1 Top Loading & Analytical Balance

Prior to use on each day of use, verify the calibration verification check on the analytical and/or top loading balance has been performed with working reference weights that bracket the measurements of weights that will be taken.

Verify the balance calibration is within its annual calibration timeframe. If the calibration sticker is missing or the calibration has expired, do not use the balance and notify the quality department of the situation.

If an alternate balance is used: ensure the balance used has been checked for tolerances that bracket the entire range of weight measurements.

9.1.1.2 Drying Oven

Verify the oven is equipped with a calibrated thermometer prior to use.

If the thermometer is missing a calibration label or the calibration has expired, notify the quality department. Do not use the oven until it is equipped with a calibrated thermometer appropriate for the temperature ranges required for the test and the gradients required to record temperatures of use.

If the thermometer used is assigned a correction factor (CF), record the original observation and the temperature corrected for the CF. If the logbook or electronic

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

log does not have a location to record both temperatures, notify the Quality Department of the gap in traceability.

9.1.1.3 Mechanical Pipettes

Prior to use, check the calibration label of the pipette(s) to verify the accuracy check has not expired. If expired or the calibration label is missing, notify the quality department. Do not use the pipette until its calibration status has been verified.

If a different pipette is used, ensure its calibration has been verified at the volume of use before using the pipette.

9.1.1.4 Refrigerators / Freezers

Ensure samples and extracts, and reagents and standards, if stored refrigerated or frozen, are stored separately and the temperature of the units are monitored daily, including weekends and holidays. Refer to the laboratory's SOP, however named, for monitoring procedures and recording of temperature measurements.

9.1.2 Extraction Equipment

9.1.2.1 Suitability Checks

Verify the suitability of SPE cartridges and other disposable plasticware by lot prior to use.

When a new lot of SPE cartridges are received, a lot check is performed by preparing an LCS and a blank of the new lot using the aqueous preparation procedure and then analyzing the extract via LCMSMS.

These supplies are acceptable for use if no target analytes are detected above ½ the detection limit.

9.1.2.2 Cleaning Procedures

Rinse all glassware prior to use with methanolic ammonium hydroxide (1%) and methanol solvent. Air dry after the solvent rinse.

9.1.2.3 SPE Manifold Setup & Use

Inspect sample manifold ports and transfer tubing (if used) prior to use for signs of wear and/or discoloration. Replace manifold ports and transfer tubing as needed.

Clean all parts of the SPE manifold including the chamber prior to use and between samples with methanolic ammonium hydroxide (1%). Air dry before use.

9.1.3 Analytical Instrumentation

9.1.3.1 Instrument Operating Conditions

Set the instrument operating conditions to the following specifications:

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

LC Gradient					MS Parameters	
Step	Total Time (min)	Flow Rate (μL/min)	A ¹ (%)	B ² (%)	Parameter	Setting/Value
1	0.00	400	80	20	Sample Loop	40 μL
2	7.00		0	100	Injection Volume	3 μL
3	7.01		80	20	Column Oven Temperature	40°C
5	8.50		80	20	RT Window	60-90 sec
					Nebulizer	40 psi
					Sheath Gas Flow	11 L/min
					Capillary Voltage	-2500 V
					Gas Temperature	250°C
					Sheath Gas Temperature	300°C

¹ A: 5% Acetonitrile w/ 2mM Ammonium Acetate.

² B: Acetonitrile.

9.1.3.2 Mass Calibration & Mass Calibration Verification

Perform mass calibration and mass calibration verification following the instrument manufacturer instructions prior to first use and at least annually thereafter to check instrument sensitivity and selectivity. Repeat following major instrument maintenance, movement of the instrument, or when other quality control (QC) failures suggest the need.

Ensure the procedures used for mass calibration and mass calibration verification evaluate an ion range that encompasses the ion range (Q1 and Q2 m/z) of the analytes of interest in this method. If the manufacturer's instructions include options for evaluation of mass resolution, the tightest resolution requirements (typically called unit resolution) must be met.

9.2 Calibration Design

Unless otherwise specified, instrument calibration procedures must comply with the PAS Corporate Policy for *Acceptable Calibration Practices for Instrument Testing* (POL ENV-POL-CORQ-0005). Refer to this policy as needed to learn more about policies and requirements for calibration and calibration verification.

9.2.1 Calibration Model

This SOP includes two (2) calibration models. Each option uses isotopically labeled compounds added to the samples prior to extraction. Option 1 is required for any analytes for which there is a stable isotope analogue.

- **Option 1:** Isotope Dilution Quantification (ID): the response of the target compound is compared to the response of its isotopically labeled analog. Twenty-five (25) target compounds are quantified in this way.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633 Effective Date: 10/14/2024

- **Option 2:** Extracted Internal Standard quantification (EIS): the response of the target compound is compared to the response of the isotopically labeled analog of another compound with chemical and retention time similarities; seventeen (17) target compounds are quantified in this way.

9.2.2 Calibration Design

The design of the curve specified in this SOP includes analysis of nine (9) calibration standards; however, not all standards must be included in the final curve.

More than the minimum number of calibration points specified in the published reference method are analyzed to extend the calibration range and to permit exclusion of the lowest or highest standards when needed; the calibration set is designed to leave enough standards remaining to meet the minimum number required for the curve fit, and to ensure that eliminating standards at the low or high end of the calibration does not alter the established LOQ.

- When linear calibration is used, the calibration curve must minimally include 6 calibration levels, with at least 5 within the quantitation range, and the lowest standard at or below the LOQ.
- When non-linear calibration (Quadratic) is used, the calibration curve must minimally include 7 calibration levels, with at least 6 within the quantitation range, and the lowest standard at or below the LOQ.

Weighting is allowed for linear and non-linear regressions, so long as the weighting is inversely proportional to concentration and is not forced through zero.

When considering the range of quantitation, ensure the LOQ routinely achieves a signal to noise ratio of 3:1, and meets the requirements for the confirmation ion and the ion ratio requirements specified in the published reference method. If a confirmation ion is not specified in the published method for the analyte, the signal to noise ratio must be minimally 10:1.

Each calibration sequence must also include the analysis of a second source standard (ICV); however, this standard is never included in the curve – it is used to evaluate accuracy and/or potential bias of calibration standard preparation and curve fit.

Refer to Appendix B for the formulations for each working calibration standard.

Acceptance criteria for the curve are provided later in this SOP.

9.2.3 Calibration Frequency

Calibrate the instrument at initial setup, when calibration verification (ICV, ISC, CCV) results indicate the calibration is no longer valid, when changes made to the operating conditions may affect calibration performance, after major instrument maintenance, or after any other event that may affect instrument performance, such as moving the instrument.

9.2.4 Calibration Sequence

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Prepare the calibration standards and the ICV following the instructions in Appendix B.

Prepare an instrument blank (IBLK) by adding 4925 µL of the IBLK Mix and 50 µL of EIS and 25 µL NIS to a plastic centrifuge tube; vortex to mix.

Analyze calibration standards L1-L9 in order from lowest to highest concentration.

Analyze the prepared IBLK for the Initial Calibration Blank (ICB) check after the highest concentration calibration standard, followed by the ICV. The ICB is used to check for carryover contamination from the highest point in the calibration range; see Appendix A, Table 7, for acceptance criteria.

9.3 Calibration Evaluation

Evaluate the calibration for linearity, accuracy, sensitivity, and carryover. If any criteria are not met, troubleshoot to identify and correct the problem, then repeat calibration. All criteria must be met before analysis of test samples.

Suggested Actions:

- Perform instrument maintenance and reanalyze the calibration sequence.
- Remake the standards and analyze a new calibration sequence.
- Reduce the range of calibration by excluding the highest concentration calibration standards or the first calibration standard. Since CAL L1 is not routinely included in the curve and CAL 2 is the LOQ, no other standards at the low end may be removed. Removal of standards in the interior of the curve is not permitted unless an obvious error with the standard is observed.

9.3.1 Linearity Evaluation

Use one of the following options to evaluate the linearity of the calibration curve. When using a weighted regression, assess linearity with option 2 below.

- Linear Calibration: calculate the relative standard deviation (RSD) of the Relative Response (RR) or Response Factor (RF) values of the initial calibration standards for each target analyte and isotopically labeled compound. The RSD must be $\leq 20\%$.
- Non-Linear Quadratic (2nd Order Fit): calculate the relative standard error (RSE) for each native compound and EIS compound for all the initial calibration standards that were analyzed. The RSE for all target and EIS compounds must be $\leq 20\%$. See Section 10.4.6 for the equation to calculate RSE. Maintain a record of the calculation and evaluation with the curve for historical recordkeeping and reference.

9.3.2 Initial Calibration Verification (ICV)

To verify the accuracy of the calibration, check the recovery of the ICV. The recovery should be within $\pm 30\%$ of the expected value.

When criteria are not met, it is likely there is a problem with the quality or preparation of the calibration standards and/or the ICV; correct the problem prior to sample analysis.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633 Effective Date: 10/14/2024

Suggested actions include:

- 1) Remake and reanalyze the ICV
- 2) Remake calibration standards and repeat the calibration sequence.
- 3) If instrument maintenance is performed: in addition to reanalyzing the ICV, the calibration sequence may need to be reanalyzed if the conditions under which the acceptability of the calibration curve was evaluated have changed.

9.3.3 Instrument Sensitivity Check (ISC)

Check instrument sensitivity daily at the lowest calibration standard included in the quantitation range (CAL L2).

The signal-to-noise ratio of the ISC must be $\geq 3:1$ for the quantification ions and the confirmation ions, or $\geq 10:1$ if the analyte only has a quantification ion. Recovery of the native and EIS compounds for the ISC must be within 70-130%. If the requirements cannot be met, the problem should be corrected before analyses can proceed.

If an individual target compound recovery in an ISC is above the upper control limit and all associated samples are ND for that compound, the data for those samples may be reported. In such cases, a narrative statement must be included in the report indicating the specific compound result that was biased high in the ISC, and that samples were ND for that compound and thus reportable.

See Appendix A, Table 6, for ion associations for each target analyte.

9.3.4 Bile Salts Interference Check

Verify the bile salt peak (TDCA) is detected > 1 minute outside RT window for PFOS. TDCA is added to multiple calibration standards to facilitate this evaluation.

9.3.5 Initial & Continuing Calibration Blanks (ICB & CCB)

Calibration blanks are used to assess carryover and/or contamination in the instrument system and are included in the initial calibration sequence and each daily analytical sequence thereafter.

In analytical sequences that include calibration, analyze the ICB following the highest concentration ICAL standard before the ICV. In sequences without calibration, analyze a CCB each day prior to sample analysis after the opening CCV.

The concentration of each analyte must be $\leq$ the MDL.

- If contamination is suspected, troubleshoot to identify and eliminate the source.
- If the failure is due to carryover from a prior standard, either reduce the upper range of the calibration or increase column flush time to reduce potential for carryover.

9.3.6 Continuing Calibration Verification (CCV)

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Verify the calibration at the beginning of each analytical sequence, after every ten environmental samples, and at the end of the sequence. The ISC may serve as the daily opening CCV. Do not include quality control (MB, LCS), instrument performance check standards (CCV), sample-specific matrix spikes, MS/MSD, field duplicates (FD), or trip blanks in the count of environmental samples.

Use ICAL Level 5 as the CCV and analyze it under the same conditions as the calibration standards.

The recovery of each target analyte and the EIS compounds for the CCVs must be within 70-130% to meet CCV criteria.

See Appendix A, Table 6, for ion associations for each target analyte.

If criteria are not met, take corrective action and reanalyze any extracts that were analyzed between the last passing CCV and the one that failed, unless the CCV result is above the upper limit and all associated samples are not detected (ND), in which case, reanalysis is not required.

Alternately, immediately analyze two CCVs for confirmation. If both confirmation CCV analyses meet the recovery criteria, proceed with analysis; however, samples that were analyzed after the last passing CCV and before the 2 passing confirmation CCVs must be re-analyzed. If either of the 2 confirmation CCV analyses fails to meet the acceptance criteria, recalibrate the instrument.

9.4 Sample Preparation: General Requirements

9.4.1 Contamination Prevention

To reduce potential contamination or bias, do not use any fluoropolymer articles or task wipes during sample preparation or extraction procedures. Use only HDPE or polypropylene containers; reagents and solvents used for cleaning purposes may be stored in glass containers.

9.4.2 Pre-Screening Procedures

Samples may be pre-screened at the discretion of the laboratory prior to sample preparation and/or analysis. Results of the screen may be used to determine if reduced volume extraction is needed or to target dilution of extracts prior to analysis.

Refer to Appendix C for instructions to prepare screening extracts when screening has been scheduled.

9.4.3 Homogenization, Subsampling, and/or Pre-Preparation Requirements

Different matrices require or may require homogenization, subsampling or pre-preparation as specified in the following subsections before sample preparation and extraction.

9.4.3.1 Aqueous: Non-Potable Water and Landfill Leachate

The sample volume used for preparation and extraction may be subsampled from the original container received when it is known that the sample requires a dilution of > 10X or when EIS criteria cannot be met due to matrix interference.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633 Effective Date: 10/14/2024

When subsampling is required to manage high concentrations, no less than 250 µL of aqueous or leachate samples may be subsampled - if greater dilution is required, serial dilution must be employed. When subsampling is required to manage challenging matrices, no less than 1 mL of samples may be subsampled.

Subsampling must be performed with customer knowledge to confirm the protocol used by the laboratory meets their project-specific requirements.

Refer to Appendix D for subsampling instructions.

9.4.3.2 Aqueous: TCLP or SPLP Leachates

The TCLP or SPLP leachates are generated using the laboratory's routine protocol for leachate generation. The leachate solutions provided by the work area performing this step are used as samples for PFAS extraction and analysis. TCLP/SPLP leachates are prepared following the Landfill Leachate protocol, with nominally 100 mL of sample extracted.

9.4.3.3 Aqueous: AFFF or F3

Samples composed of AFFF, F3, or other matrices KNOWN to require a large dilution prior to extraction will be subsampled consistently, with approximately 0.02 g of material aliquoted for each discrete sample preparation/analysis; each AFFF or F3 concentrate sample must be prepared in duplicate. After creating and documenting the subsample aliquot in a 125 mL HDPE PFAS sample bottle, add approximately 60 mL of reagent water, cap the vessel, and shake vigorously by hand for 20-30 seconds. After adding the reagent water and agitating, allow at least 3 hours for the subsample aliquot to fully dissolve in the water. If dissolution is incomplete after 3 hours, agitate again and wait another 3 hours; consider applying continuous agitation (orbital shaker) if full dissolution of the aliquot into water requires extended time. Following the extended dissolution time, prepared AFFF/F3 samples should be extracted and analyzed following the routine protocols for aqueous sample processing. Aqueous reporting limits will be used, with values adjusted to account for the actual sample aliquot, assuming 1 g/mL; this constitutes a 25,000X nominal adjustment factor for AFFF/F3 reporting limits, relative to nominal aqueous reporting limits.

Pace STRONGLY DISCOURAGES clients from sending AFFF concentrate material for analysis; this protocol is designed for "as-prepared" AFFF materials (i.e., pre-diluted to application concentration) and newer generation 'Fluorine Free Foam' (F3) concentrates. If F3 material (concentrate or as-prepared) or AFFF "as-prepared" material is received, follow the normal routines described in the paragraph above. If an AFFF concentrate material (usually of a thick gel-like consistency) is received for analysis, the above protocol may not be enough of a dilution to protect lab systems; in these cases, the lab shall prepare a serial dilution prior to extraction. First, the protocol above is followed (0.02 g of material fully dissolved into 60 mL water). Then a 6 mL aliquot of the initial aqueous solution is diluted into water, with a final volume of 60 mL (i.e., dilute 6 mL of sample solution into 54 mL of water). This serial dilution constitutes a 250,000X

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633 Effective Date: 10/14/2024

nominal adjustment factor for the AFFF concentrate reporting limits, relative to nominal aqueous reporting limits. Handling of AFFF concentrate should ideally be done in an isolated area outside of the PFAS processing area to prevent gross lab contamination.

9.4.3.4 Solid: Soils and Sediments

If a reduced mass extraction is scheduled, use at least 0.5 g (dry weight) for all solid matrices to ensure the amount used for sample preparation is representative of the total mass of sample collected.

9.4.3.5 Solid: Biosolid

If a reduced mass extraction is scheduled, use at least 0.05 g (dry weight) for all biosolid matrices to ensure the amount used for sample preparation is representative of the total mass of sample collected.

9.4.3.6 Biphasic

If the sample is biphasic and the customer has requested phase separation: centrifuge the sample to separate the phases following the procedure specified in Appendix F, then prepare and extract each phase separately.

9.4.3.7 Articles of Commerce

Articles of commerce typically need to be milled to a fine powder using the protocol specified in Appendix H before sample preparation. This step may not be required when the material is known to be sufficiently homogenous and more than 95% of the material will pass through a 250 µm sieve.

Perform this step as needed and when specified on a project basis.

9.4.3.8 Tissue

Homogenization of tissue matrices is performed on behalf of all PAS locations by the PAS laboratory located in Green Bay, Wisconsin (PAS-Green Bay) in accordance with their local SOP ENV-SOP-GBAY-0129 Sample Homogenization Compositing and Sub-Sampling. PAS personnel may access this SOP via “Qualtrax” (AKA Ideagen Quality Management), the electronic document management system (eDMS) used by PAS for document management, control, and distribution.

If a reduced mass extraction is scheduled, use at least 0.2 g for all tissue matrices to ensure the amount used for sample preparation is representative of the total mass of sample collected.

9.5 Percent Solids Determination

9.5.1 Aqueous Matrices

A percent solids determination is not conducted for aqueous matrices. Instead, the laboratory visually assesses the sample matrix during sample preparation and employs alternative strategies to mitigate the effect of any solids present.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

9.5.2 Solid Matrices (excluding tissues and Articles of Commerce)

Percent solids must be determined on all solid and biosolid samples prior to preparation and analysis to determine the sample mass used for extraction in accordance with laboratory SOP ENV-SOP-MIN4-0055 *Determination of Percent Moisture by ASTM D2974-07*, or however named. Percent solids are logged-in and determined by the same personnel that normally generate this data daily. The percent solids analysis will follow the Method ASTM D2974-07 protocol.

Percent solids are not determined for tissue, surface wipes or the solid phase of a biphasic sample. These matrices are always reported on a wet weight basis.

9.6 Sample Preparation: Procedure

9.6.1 Aqueous Matrices

Do not filter samples at any point prior to sample preparation.

Unless a reduced volume extraction is necessary, use the following amounts for sample preparation and analysis:

Routine Extraction Volume for Aqueous Matrices¹

Non-Potable Water	Landfill Leachate	TCLP/SPLP Leachate	AFFF or F3
500 mL	100 mL	100 mL	0.02 g

¹ Nominal amount on which LOQ in Table 2 is derived. When different sample volumes are used, detection limits and the LOQ are adjusted for the actual amount used.

- For NPW: Use the entire volume from one (1) of the 500 mL containers provided.
- Landfill Leachates: Use entire volume from one (1) of the 125 mL containers provided.
- TCLP/SPLP Leachate: Subsample 100 mL of the filtered leachate generated by pouring the leachate from the container into a 125 mL HDPE PFAS sample bottle. Use the gradations on the bottle as a measuring guide.
- AFFF or F3: Subsample 0.02 g of the material received into a 125 mL HDPE PFAS sample bottle and dilute in 60 mL of reagent water; see Section 9.4.3.3 for the complete pre-extraction preparation protocol.

Create separate preparation batches for aqueous matrices that use different initial volumes because the volume used for the MB and LCS/LLCS must match the amount used for samples.

Preparation Steps:

- 1) Remove the sample container from storage. Pull additional containers for each MS/MSD and for the laboratory duplicate, when a duplicate is requested or required, and an additional container has been provided to perform the analysis.

DoD Accredited Laboratories: If the matrix is AFFF or F3, a duplicate must be performed with each sample. Confirm that there is enough sample available to take the duplicate from the same container.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633 Effective Date: 10/14/2024

- 2) Invert the sample container 3-4 times to homogenize the sample and then allow the sample to settle.
- 3) Visually inspect each settled sample for settled or suspended solids. Consider any visible indications of particulate matter including settled solids collected on the bottom of the container, cloudiness and/or dark color of the sample, suspended solids within the sample, increased viscosity, etc. If uncertain, seek a second opinion from another analyst, team lead, or supervisor; these matrix characteristics may require reduced-volume extraction.
 - a) If potential interferences other than settled solids are observed, take action to mitigate the impacts on data quality. Examples of other sample characteristics that could necessitate precautionary measures with a field sample (i.e., prepare from reduced volume) include presence of standing foam in the sample after shaking, thick/viscous matrix, samples from known highly contaminated sites/projects, client notes indicating potential high concentrations, and knowledge of potential hazards to safety, equipment performance, or process integrity due to sample composition. In such cases, confer with a team lead or supervisor to determine the best approach. Use the largest volume possible (i.e., the smallest reduction possible) when performing a reduced-volume extraction to manage challenging matrix characteristics; use no less than 1 mL of sample.
 - b) If none of these conditions are observed or if the solids observed are less than ½ cm in depth, proceed to Step 4.
 - c) If settled solids are observed at a depth greater than ½ cm, skip Section 9.6.1 and follow the preparation and extraction instructions specified in Appendix E.
 - d) If settled or suspended solids appear to be more than 5% of the total volume of the sample:
 - Recap the container and shake to homogenize; uncap the container.
 - Using a pipet calibrated to quantitatively measure 1 mL, transfer 1 mL of sample into a pre-labeled 50 mL centrifuge tube.
 - Add 40 mL of reagent water to the tube.
 - Record 1 mL for the volume used for sample preparation and extraction.
 - Proceed with Step 5 after initial weight has been determined for all samples in the batch.
- 4) Weigh the container+sample on the top-loading balance and record the weight measurement in the preparation record to the nearest 0.1 g. This measurement will be used later to calculate the actual volume of sample extracted.
- 5) Prepare a method blank (MB), laboratory control sample (LCS), and low-level laboratory control sample (LLLCS) by measuring the volume of blank matrix specified into a 500 mL HPDE container. For TCLP / SPLP, prepare 2 MB.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

- NPW – Use 500 mL of reagent water.
 - Landfill Leachate – Use 100 mL of reagent water.
 - TCLP / SPLP Leachate – Use 100 mL of TCLP or SPLP blank solution for the LCS, LLCs and Leachate MB. Prepare another MB using 100 mL of reagent water.
 - AFFF / F3 – Can be batched with NPW; if batched separately, prepare batch QC following NPW routines.
- 6) Quantitatively add 128 µL of the PDS-10X standard mix to LLCs and 80 µL of 1633PDS mix to the LCS.
 - 7) Add 40 µL of EIS solution to each container (all samples and QC).
 - 8) If the batch includes matrix spikes, add 80 µL of 1633PDS mix to each matrix spike.
 - 9) Cap each container and invert to mix.
 - 10) Check the pH of each container using a pH indicator strip to verify the pH is 6.5 ± 0.5 . If necessary, adjust the pH with 50% formic acid or ammonium hydroxide (or with 5% formic acid and 3% aqueous ammonium hydroxide). Do not insert the strip into the sample container: use a disposable pipet to transfer a small amount of sample onto the pH indicator strip instead.
 - 11) Proceed to Section 9.7 for extraction and extract cleanup.

9.6.2 Solid Matrices (Non-Tissue)

Unless a reduced mass extraction is necessary, use the following amounts for sample preparation and analysis:

Routine Target Mass for Solid Matrices¹

Soil/Sediment	Biosolid	Articles of Commerce
5 g dry weight	0.5 g dry weight	2 g

¹ Nominal amount on which LOQ in Table 2 is derived. When different sample mass is used, detection limits and the LOQ are adjusted for actual amount used.

Do not combine biosolids in the same preparation batch as soils/sediments or articles of commerce due to the difference in amount of material used.

Skip Steps 2-4 if the sample matrix is a surface wipe or article of commerce.

- 1) Remove the sample from storage.
- 2) Using a wooden tongue depressor, mix each sample in its original jar by stirring from the bottom of the container to the top and in a circular motion along the sides of the jar, breaking particles up by pressing against the side of the container (to <1 mm). The homogenized sample will be even in color and have no separate layers. Store the homogenized material in its original container. If it is not practical to mix the sample within its container, transfer the sample to a larger container to mix.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

- 3) Check the LIMS to obtain the percent solids result for each sample in the batch.
- If the test results are not available, check to see if a percent solids analysis is included in the work order; if not, notify the Project Manager (PM) they will need to add the test to the log-in.
 - If test results are not available because the department performing the test has not completed the determination, notify your supervisor or contact the leader of the department performing the test to obtain the status. Preparation may not begin until this information is available.
 - If the percent solids result is <50%, notify the PM and request that the matrix in the work order be changed to biosolid; prepare the sample as a biosolid.
 - If the percent solids result is <5%, notify the PM and request the matrix be changed to landfill leachate; prepare the sample as a landfill leachate.

If none of these scenarios apply, proceed to Step 4.

- 4) Determine the target mass for extraction for soils/sediments and biosolids by dividing the target weight for sample preparation by the percent solids in decimal form.

For example, if a soil sample is 90% percent solids, the mass used for extraction must be within the range of 5.4-5.8 g.

- a) Convert 90% percent solids to decimal form by dividing 90 by 100. ($90/100 = 0.9$)
- b) Divide 5 g (target mass) by the percent solids in decimal form ($5/0.9 = 5.555...$)
- c) Round 5.5555 to the same significance of measurement as the top loading balance¹ (5.6).
- d) Add ± 0.2 g to the target result of 5.6 g to obtain the target weight range of 5.4 to 5.8 g.

¹ For biosolids, use an analytical balance and convert the result to the significance of the analytical balance (0.01 g)

- 5) Place a 50 mL polypropylene centrifuge tube on the balance and tare the balance.

Measure the target weight of sample into the tube so that the weight is within the range determined in the prior step. Record the amount used to the nearest 0.1 g (top loading) or 0.01 g (analytical).

NOTE: Surface wipes should have been received in a 50 mL centrifuge tube. If the wipe was received in a different container, transfer the wipe to a 50 mL centrifuge tube using clean forceps.

If matrix spikes or a laboratory duplicate were requested or are required, prepare separate containers for each.

If the sample is extremely dry, wet the sample with 2.5 g of reagent water, which is the same amount always used to wet the clean matrix used for the MB and LCS.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633 Effective Date: 10/14/2024

- 6) Prepare the MB, LCS and LLLCS using Ottawa Sand wetted with reagent water by adding the clean matrix and water to a 50 mL polypropylene centrifuge tube.
 - Soil/Sediment – Use 5 g Ottawa Sand and 2.5 g of reagent water
 - Biosolid – Use 0.5 g of sand and 0.25 g of reagent water
 - Surface Wipe – Use a new ghost wipe from a pre-tested lot
 - Articles of Commerce – Can be batched with soil/sediment; if batched separately, prepare batch QC following soil/sediment routines.
- 7) Quantitatively add 160 µL of the PDS-10X standard mix to LLLCS and 80 µL of 1633PDS mix to the LCS.
- 8) If the batch includes matrix spikes, add 80 µL of 1633PDS mix to each matrix spike sample.
- 9) Add 40 µL of EIS solution to each container (all samples and QC)
- 10) Using a vortex mixer, vortex the tube to disperse the spike solution and allow to equilibrate for approximately 30 minutes. Alternatively, shake by hand.
- 11) First Extraction: Add 15 mL of 0.3% methanolic ammonium hydroxide to each centrifuge tube. Vortex to disperse, then shake for 30 minutes on a variable speed mixing table. Next, centrifuge each tube for 10 minutes at 3000 rpm (~2000 RCF) then transfer the supernatant to a clean, labeled 125 mL HDPE sample bottle.
- 12) Second Extraction: Add 15 mL of 0.3% methanolic ammonium hydroxide to the remaining solid sample in each centrifuge tube. Vortex to disperse, then shake for 30 minutes on a variable speed mixing table. Centrifuge each tube at 3000 rpm (~2000 RCF) for 10 minutes and decant the supernatant from the second extraction into the same container with the supernatant from the first extraction.
- 13) Adjust the extraction solution to 60 mL volume using reagent water. A validated comparison bottle may be used to make this adjustment.
- 14) Check the pH of each sample solution using a pH indicator strip to verify the pH is 4.0 ± 0.5 . If necessary, adjust the pH with 50% formic acid or ammonium hydroxide (or with 5% formic acid and 3% aqueous ammonium hydroxide). Do not insert the strip into the sample container: use a disposable pipet to transfer a small amount of sample onto the pH indicator strip instead.
- 15) Proceed to Section 9.7 for extraction and extract cleanup.

9.6.3 Tissue

Thaw the homogenized tissue before starting the preparation procedure.

- 1) Place a 50 mL polypropylene centrifuge tube on a top loading balance and tare the balance. Weigh $2 \text{ g} \pm 0.2 \text{ g}$ of homogenized sample into the tube. Record the weight measurement to the nearest 0.1 g. After each sample in the batch has been subsampled, reseal the containers and return to frozen storage.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

- 2) If matrix spikes or a laboratory duplicate were requested or are required, prepare separate containers for each.
- 3) Prepare the MB, LCS and LLLCS using a reference tissue matrix.
- 4) Quantitatively add 160 µL of the PDS-10X standard mix to the LLLCS and 80 µL of 1633PDS mix to the LCS.
- 5) If the batch includes matrix spikes, add 80 µL of 1633PDS mix to each matrix spike sample.
- 6) Add 40 µL of EIS solution to each container (all samples and QC).
- 7) Using a vortex mixer, vortex the tube to disperse the spike solution and allow to equilibrate for approximately 30 minutes. Alternatively, shake by hand.
- 8) 8) First Extraction: Add 15 mL of 0.05M KOH in methanol to each tube. Vortex to disperse then place tubes on a variable speed mixing table to shake for 1 hour. Centrifuge the tube at 3000 rpm (~2000 RCF) for 10 minutes and collect the supernatant in a clean, labeled 125 mL HDPE sample bottle.
- 9) 9) Second Extraction: Add 10 mL of acetonitrile to remaining tissue in the centrifuge tube, vortex to mix and disperse the tissue. Sonicate for 30 minutes. Centrifuge each tube at 3000 rpm (~2000 RCF) for 10 minutes and collect the supernatant, adding it to the same container with the supernatant from the first extraction.
- 10) Adjust the extract solution to 75 mL volume using reagent water. A validated comparison bottle may be used to make this adjustment.
- 11) Check the pH of each sample solution using a pH indicator strip to verify the pH is 4.0 ± 0.5 . If necessary, adjust the pH with 50% formic acid or ammonium hydroxide (or with 5% formic acid and 3% aqueous ammonium hydroxide). Do not insert the strip into the sample container: use a disposable pipet to transfer a small amount of sample onto the pH indicator strip instead.
- 12) Proceed to Section 9.7 for extraction and extract cleanup.

9.7 SPE Extraction & Carbon Cleanup

Use the same procedure for all matrices.

- 1) Prepare an SPE cartridge for each sample and QC by packing clean silanized glass wool into the bottom of a WAX SPE cartridge barrel, to a height of ~1 cm. Ensure the glass wool is not densely packed to allow for proper sample flow.
- 2) Set up the vacuum manifold with one prepared WAX SPE cartridge, with a reservoir adaptor and reservoir stacked on each cartridge, ensuring all connections are tight. If using plastic transfer lines instead of reservoirs, ensure the connecting caps are snug in each cartridge and the transfer lines are placed within the appropriate sample. Label each reservoir or cartridge tube with the associated sample ID.
- 3) Pre-condition the cartridges with 15 mL of 1% methanolic ammonium hydroxide followed by 5 mL of 0.3M formic acid. Perform each rinse separately. Do not allow the WAX SPE to go dry. Discard the wash solvents.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633 Effective Date: 10/14/2024

- 4) Pour the sample into the reservoir taking care to avoid splashing while loading; or, if using transfer lines, begin opening the stopcocks on the vacuum manifold. Do not pipette the sample in this step. Adjust the vacuum/stopcocks to pass each sample through the cartridge at a rate of approximately 5-10 mL/min; discard the eluate. Retain the empty sample bottle for solvent rinsing.

If the SPE cartridge becomes clogged during sample transfer, refer to Appendix G for troubleshooting steps and mitigation strategies.

- 5) Rinse the walls of the reservoir twice with 5 mL reagent water, followed by 5 mL of 1:1 Methanol:0.1M Formic acid. Pass each rinse through the cartridge using vacuum. If using sample transfer lines for loading, add the 3 rinses to the original sample container and load each rinse through the transfer lines to the cartridge, one reagent at a time. Discard the rinse solutions.
- 6) Dry the cartridges by pulling air through them for ~ 5 minutes.
- 7) While the WAX cartridges are drying, condition the carbon cartridges (GCB) by rinsing with two aliquots of 5 mL of MeOH. Perform this step on a separate vacuum manifold or on the unused ports of the same manifold as the WAX cartridges.

Insert the narrow end of a 5 mL or 10 mL plastic pipette tip into the top of the GCB cartridge and use it like a reservoir for the rinse solvent. Apply a light vacuum to pull the rinse solvent through the cartridge; discard the rinsates. Do not allow the GCB cartridges to go completely dry during/after this step. Remove the pipette tip from the GCB cartridge and discard.

- 8) When the WAX cartridges are sufficiently dry, remove each WAX cartridge and reservoir out of its manifold port and stack it on a conditioned GCB cartridge by inserting the outlet of the WAX cartridge into the inlet of the GCB cartridge. Place the dual cartridge-reservoir stack back in the same manifold port that the WAX cartridge originally occupied. This cartridge stacking should be done one cartridge at a time to avoid mixing up manifold locations.

If using transfer lines: follow the same cartridge stacking process but ensure that the transfer line connecting caps are snug in each cartridge and the transfer lines are placed within the appropriate sample bottle after stacking the two cartridges.

- 9) Place a clean collection tube in the manifold rack for each cartridge so that the extract delivery needles are not touching the walls of the tubes. Resume vacuum and ensure a proper seal is achieved. DO NOT add NIS to these collection tubes.
- 10) Rinse the inside of each sample container using 5 mL of 1% methanolic ammonium hydroxide. Then pour the rinse from each sample container to transfer the rinse to the associated SPE reservoir to wash the walls of the reservoir. If using sample transfer lines, transfer the container rinse from each sample container to its associated cartridge using vacuum. Whether using reservoirs or transfer lines, allow the elution solvent to soak the SPE sorbent for 2 minutes, then use vacuum to pull the elution solvent through the cartridge and into the collection tubes in a slow, dropwise manner.
- 11) Aqueous samples: air dry the empty sample container after the rinse is transferred. Weigh the empty container with the cap on and subtract that from the weight of the bottle with sample, determined in Section 9.6.1, step 4.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

12) Add 25 µL of concentrated acetic acid and 20 µL of NIS mix to each collection tube and vortex to mix. Transfer a portion of each extract into an autosampler vial for LC-MS/MS analysis. Cap the collection tubes containing the remaining extract and store the extract at 0 - 6°C.

13) Proceed to Section 9.8 for analysis.

9.8 Analysis

Analyze the extracts by LC-MS/MS in multiple reaction monitoring (MRM) mode, with unit mass resolution. Allow the extracts to warm to room temperature prior to aliquoting into autosampler vials and loading on the instrument.

On each day of analysis:

- 1) Flush the column with 100% Acetonitrile for at least 30 minutes to clear any accumulated impurities and interferences from the sample pathway and to equilibrate the system. Open the purge valve(s) on the pumps for the first ~1 minute of flush time and/or use the “purge” function available as part of the pump functionality and purge the autosampler injector flow path using the “purge” function, if available.
- 2) Equilibrate the column by flushing the column for ~15 mins with 50:50 Mobile Phase A: Mobile Phase B followed by ~15 minutes of 98:2 Mobile Phase A: Mobile Phase B.
- 3) Verify system pressure is stable.
- 4) Bring standards and samples to room temperature, vortex to homogenize, then transfer a portion to an autosampler vial in preparation for analysis.
- 5) Following any initial priming injections on days when a calibration is not needed, an ISC and an instrument blank (IBLK) will be analyzed to demonstrate system suitability and the absence of system contamination prior to sample analysis.
- 6) After acceptable ISC and IBLK results are confirmed, load and submit extracted samples for analysis, typically starting with batch QC with field samples following. A CCV and CCB must be analyzed after every 10 (or fewer) field samples.

Arrange standards and samples in an autosampler to run in sequence as follows:

Order of Sequence with ICAL	Order of Sequence w/o ICAL
Cal L1-9 in order of lowest to highest	Opening ISC/CCV
IBLK	IBLK
ICV	MB
Follow same sequence as w/o ICAL, starting at MB	LL LCS
-	LCS
-	Samples (10 or fewer)
-	CCV
-	CCB
-	Samples (10 or fewer)

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Order of Sequence with ICAL	Order of Sequence w/o ICAL
-	Closing CCV

¹ The closing CCV analysis may be used as the opening solution for the next analytical sequence when criteria for the opening CCV are also met.

10.0 DATA ANALYSIS & CALCULATIONS

Data is reduced using the acquisition software provided by the instrument manufacturer.

10.1 Retention Time (RT) Window

Establish RT Windows for all analytes, EIS and NIS with each ICAL event and at the beginning of each analytical sequence thereafter. Analytical conditions must be set to allow a separation of at least 1 minute between TDCA and the retention time window of PFOS. If this is not achieved the problem must be corrected. The RT window used must be of sufficient width to detect earlier-eluting branched isomers.

- In sequences that include an initial calibration curve (ICAL) use the midpoint standard of the curve to set the RT for analytes, EIS and NIS.
- In sequences that do not include ICAL, use the beginning CCV retention times to set the RT for analytes, EIS and NIS.

10.2 Qualitative Identification

The presence of target analytes, EIS, and NIS compounds are qualitatively confirmed when the criteria in Sections 10.2.1. through 10.2.5. are met.

If identification criteria are not met and, based on analyst discretion, the analyte may be present, the presence of a target analyte may be reported. In such cases, data will be reported with a qualifier alerting the data user that qualitative identification could not be confirmed because the detection did not meet the method-required criteria but based on analyst discretion, the compound may be present and should be considered an estimated value.

If the criteria listed below are not met for the standards, the laboratory must stop analysis of samples and correct the issue.

10.2.1 Integration

The response of all isomers in the quantitative standards should be used to define integration parameters and ion ratios.

In samples, the total integrated response should include the branched isomer peaks that have been identified in the quantitative standard, if present. For compounds with identified branched and linear isomer peaks, the reported concentration is the sum of all isomers. If standards (either quantitative or qualitative) containing a mix of isomers are not available for purchase for a particular compound, only the linear isomer can be identified and quantitated in samples.

10.2.1.1 Manual Integration

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Manual integration is sometimes necessary to correct inaccurate automated integrations but must never be used to meet QC criteria or to substitute for proper instrument maintenance and/or method set-up. To ensure that all manual integrations are justified and proper, all manual integrations must be performed, documented, reviewed, and approved in accordance with corporate SOP ENV-SOP-CORQ-0006, *Manual Integration*. Refer to this SOP for guidance on manual integration techniques and required procedures.

10.2.2 Retention Time (RT)

The RTs for the target analytes, EIS, and NIS compounds must fall within ± 0.4 minutes of the predicted retention times from the midpoint standard of the ICAL or initial daily CCV, whichever was used to establish the RT window position for the analytical batch. All branched isomer peaks identified in the calibration standards also must fall within 0.4 minutes of the retention times set for the analytical batch.

For all target analytes with exact corresponding isotopically labeled analogs, target analytes must elute within ± 0.1 minutes of the associated EIS compound.

10.2.3 Signal-to-Noise Ratio (S/N)

Peak responses of the quantitation and confirmation ions must be at least three times the background noise level (S/N 3:1); if there is no confirmation ion, S/N must be $>10:1$.

If the S/N ratio is not met in a standard or QC sample due to high background noise, correct the problem (e.g., perform instrument maintenance).

If the S/N ratio is not met in a blank or sample, then detection of the analyte cannot be confirmed; the analyte is “not detected”.

10.2.4 Ion Ratio Criteria

For any compound that meets S/N and RT criteria, the ratio of the total (branched and linear isomer) quantification ion response to the total (branched and linear isomer) confirmation ion response – referred to as the Ion Ratio – must fall within $\pm 50\%$ of the ratio observed in the mid-point ICAL standard.

The ion ratio requirement does not apply for PFPrA, PFBA, PFPeA, NMeFOSE, NEtFOSE, PFMPA, or PFMBA because suitable secondary transitions are unavailable (not detectable or inadequate S/N).

10.2.5 Detection Limit

The calculated concentration (see Section 10.4) of the target analyte must be greater than the adjusted detection limit (DL) to be reported. Quantified results below the DL cannot be confirmed and will be reported as “not detected.”

10.3 Quantitative Analysis

Test results are quantified by automated data reduction software using the equations in Section 10.4.

10.4 Calculations

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633 Effective Date: 10/14/2024

All data is reduced by automated software, no manual calculations are performed for this test.

10.4.1 Target Analyte Concentration

Concentrations of the target analytes are determined with respect to the extracted internal standard (EIS) which is added to the sample prior to extraction (see Appendix A, Table 6). Other equations may be used if the laboratory demonstrates that those equations produce the same numerical result as produced by the equation below:

$$\text{Concentration (ng/L or ng/g)} = \frac{\text{Area}_N M_{EIS}}{\text{Area}_{EIS} (\overline{RR} \text{ or } \overline{RF})} \times DF \times \frac{V_f}{V_i}$$

Where: Area_N = The measured area of the quantitation ion for the unlabeled PFAS target analyte.

Area_{EIS} = The measured area of the quantitation ion for the EIS compound.

M_{EIS} = The expected concentration of the EIS in the extract (ng/L).

$\overline{RR}$ = Average response ratio used to quantify target compounds by the isotope dilution method.

$\overline{RF}$ = Average response factor used to quantify target compounds by the extracted internal standard method.

DF = Extract Dilution Factor; if no extract dilution was performed, $DF=1$.

V_f = Final extract volume (L).

V_i = Initial sample volume (L) or weight (g).

10.4.2 EIS Concentration

The EIS compounds are quantitated with respect to an NIS, as shown in Appendix A Table 6, using the response factors from the most recent initial calibration. Other equations may be used if the laboratory demonstrates that those equations produce the same numerical result as produced by the equation below:

$$\text{Concentration (ng/L or ng/g)} = \frac{\text{Area}_{EIS} M_{NIS}}{\text{Area}_{NIS} \overline{RF}_S} \times DF \times \frac{V_f}{V_i}$$

Where: Area_{EIS} = The measured area of the quantitation ion for the EIS compound.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633 Effective Date: 10/14/2024

$Area_{NIS}$ = The measured area of the quantitation ion for the associated NIS compound.

M_{NIS} = The expected concentration of the NIS in the extract (ng/L).

RF_S = Average response factor used to quantify the EIS compound by the non-extracted internal standard method.

DF = Dilution Factor; if no extract dilution was performed, DF=1.

V_f = Final extract volume (L).

V_i = Initial sample volume (L) or weight (g).

10.4.3 ICAL Relative Response (RR)

Used to calculate the response ratio (RR) for each target analyte calibrated by isotope dilution quantitation.

$$RR = \frac{Area_t M_{EIS}}{Area_{EIS} M_t}$$

Where: $Area_t$ = Measured area of the quantitation ion for the target analyte.

$Area_{EIS}$ = Measured area of the quantitation ion for the corresponding labeled PFAS used as the EIS in the calibration standards.

M_{EIS} = Concentration of the isotopically labeled PFAS used as the EIS in the calibration standards (ng/L).

M_t = Concentration of the target analyte in the calibration standard (ng/L).

10.4.4 ICAL Response Factor (RF): Target Analyte

Used to calculate the response factor (RF) for each target analyte calibrated by extracted internal standard quantitation.

$$RF = \frac{Area_t M_{EIS}}{Area_{EIS} M_t}$$

Where: $Area_t$ = Measured area of the quantitation ion for the target analyte.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633 Effective Date: 10/14/2024

$Area_{EIS}$ = Measured area of the quantitation ion for the corresponding labeled PFAS used as the EIS in the calibration standards.

M_{EIS} = Concentration of the isotopically labeled PFAS used as the EIS in the calibration standards (ng/L).

M_t = Concentration of the target analyte in the calibration standard (ng/L).

10.4.5 ICAL Response Factor (RF): EIS

$$RF_S = \frac{Area_{EIS}M_{NIS}}{Area_{NIS}M_{EIS}}$$

Where: $Area_{EIS}$ = Measured area of the quantitation ion for the EIS compound in the calibration standard.

$Area_{NIS}$ = Measured area of the quantitation ion for the NIS compound in the calibration standard.

M_{NIS} = Concentration of the NIS compound in the calibration standard (ng/L).

M_{EIS} = Concentration of the EIS compound in the calibration standard (ng/L).

10.4.6 ICAL Relative Standard Error (RSE)

Used to assess linearity of quadratic curve fits.

$$RSE = 100 \times \sqrt{\frac{\sum_{i=1}^n \frac{[x'_i - x_i]^2}{x_i}}{n - p}}$$

Where: x_i = Nominal concentration (true value) of each calibration standard.

x'_i = Measured concentration of each calibration standard.

n = Number of standard levels in the curve.

p = Type of curve (2 = linear, 3 = quadratic).

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

10.4.7 Other Calculations

For common calculations such as percent recovery and relative percent difference, refer to laboratory SOP ENV-SOP-MIN4-0171 *Laboratory Calculations*, or however named.

10.5 Reporting Specifications

Unless otherwise specified on a project basis, report final test results as follows:

Matrix	Units	Comment
Aqueous: All Matrices	ng/L	If alternate protocols are used to manage matrix characteristics or high concentrations, narrate sample data accordingly.
Soils/Sediments & Biosolids	ng/g dry weight	Test results are adjusted for percent moisture. Include the test information from percent solids determination in the test report.
Biphasic: Solid phase	ng/g wet weight basis	
Tissue	ng/g wet weight basis	
Article of Commerce	ng/g as received	Indicate if cryomilling was performed before preparation.

Adjust the DL and LOQ for sample volume or mass and any dilution factor before using it as the threshold value for reporting quantitative test results. Unless otherwise specified, test results between the DL and the LOQ, when reported, are qualified as estimated.

The recovery of associated EIS compounds for each target analyte must be included for each sample in the test report, regardless of the type of deliverable provided.

If the sample was diluted, include the DF in the test report to identify the factor by which the final test results were adjusted. Results for any analyte found in a sample or extract that has been diluted will be reported at the least dilute level for which the measured concentration is within the calibration range (e.g., above the LOQ for the analyte and below the highest calibration standard) and in which isotopically labeled compound recoveries are within their respective QC acceptance criteria. This may require reporting results for some analytes in the same sample from different analyses.

For compounds with identified branched and linear isomer peaks, the reported concentration is the sum of all isomers. If standards (either quantitative or qualitative) containing a mix of isomers are not available for purchase for a particular compound, only the linear isomer can be identified and quantitated in samples.

11.0 QUALITY CONTROL & METHOD PERFORMANCE

11.1 Method Performance Validation

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633 Effective Date: 10/14/2024

The performance of this SOP was verified in accordance with the PAS corporate SOP for Method Validation and Instrument Verification. Records of verification are kept by the Quality Department in accordance with the PAS policy for Record Management (ENV-POL-0013).

Detection limits (DL or MDL, including LOD for DoD) and limits of quantitation (LOQ) were established in accordance with laboratory SOP ENV-SOP-MIN4-0162 *Determination of LOD and LOQ*, or however named, during initial method setup.

These limits are continuously verified and updated in accordance with USEPA procedures for determining method detection limits, as well as the requirements specified in the TNI Standard and DoD/DOE QSM, as applicable. Customers should be informed that the laboratory is obligated to update these limits when verification checks reveal a change in sensitivity or method performance, regardless of customer approval.

11.1.1 Initial Precision and Recovery (IPR)

To verify the performance of the laboratory's SOP against the specifications of the test method, a demonstration of capability (DOC) must be conducted for each matrix following the procedure outlined in Section 9.2.1 of EPA Method 1633. Refer to the method published in January 2024 by the USEPA Office of Water (Document EPA 821-R-24-001) for detailed instructions. In summary, the lab must follow the SOP protocol to extract and analyze four (4) samples prepared as LCS, along with at least one MB, for each matrix to be tested. The average percent recovery and RSD of the target analyte results are tabulated and compared to method acceptance criteria.

The results of the IPR must be within the accuracy and precision limits specified in Appendix A, Table 10. There are no IPR mean or RSD criteria for EIS compounds; %REC of each IPR sample must meet the criteria in Appendix A, Table 9.

11.2 Analyst Demonstration of Capability (DOC)

Before conducting unsupervised (independent) work on any client samples, analytical personnel (including sample preparation) must demonstrate their ability to achieve accurate and precise test results using the procedures specified in this SOP (IDOC) and redemonstrate this capability on an annual basis thereafter (ODOC).

Refer to the laboratory's Quality Manual and to laboratory SOP ENV-SOP-MIN4-0165 *Laboratory Training Procedures*, or however named, for more information about the requirements and procedure for IDOC and CDOC.

The average recovery of the target analytes in the DOC samples must be within the accuracy limits for LCS specified in Appendix A, Table 8, and the RSD must be within the precision limits for IPR specified in Appendix A, Table 10. There are no mean or RSD criteria for EIS compounds in the DOC; %REC of each DOC sample must meet the criteria in Appendix A, Table 9.

The IPR study may also suffice as IDOC for the analyst who performed the study.

11.3 Matrix Impact (MS/MSD)

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Matrix spikes are used to assess potential impact of matrix interference on the accuracy or precision of the procedure on samples of the same or similar matrix – samples of similar matrix are usually classified as samples of the same matrix type collected from the same sampling site.

According to EPA Method 1633, matrix spikes (MS/MSD) generally are not required for methods that employ isotopic dilution because the recovery of the EIS compounds spiked into every sample reflects the effect the matrix has on recovery of target compounds.

Therefore, matrix spikes are not required for compliance monitoring. However, the EPA method acknowledges that some projects or programs may be interested in this information.

Customers interested in using information from matrix spikes to understand potential matrix impact on their samples must request MS/MSD, provide additional sample material to the laboratory to perform the test, and identify which sample to use on the chain-of-custody (COC).

If no customer has requested MS/MSD on any sample in the preparation batch, an MS/MSD is not performed.

NOTE for DoD Projects: Some DoD QSM versions require 1 MS/MSD pair per preparation batch. Customers performing work that requires compliance with a version of the QSM that requires MS/MSD must provide additional sample material for the laboratory to perform this test.

11.4 Sampling Impact (Field Duplicate)

A field duplicate (FD) refers to the testing of a replicate container of sample collected with all other samples from a sampling event and used to measure precision of the field sampling technique.

Customers requiring a field duplicate must request this analysis and provide a separate container for analysis. The FD must be logged in as a routine environmental sample; it may not be subsampled from the same container of any other sample.

11.5 Quality Control (QC)

The laboratory prepares and analyzes the following QC at the frequency specified in the table below. Refer to Appendix A, Table 7, for acceptance criteria and requirements for corrective action(s) and/or data qualification.

Item ¹	Acronym	Frequency
Mass Calibration	N/A	Annually and as needed.
Mass Calibration Verification	N/A	After mass calibration.
Initial Calibration	ICAL	Initially at instrument setup, then as needed.
Initial Calibration Verification	ICV	With each ICAL, prior to sample analysis.
Instrument Blank	IBLK/ICB	Daily prior to analysis and after high concentration standards.
Instrument Sensitivity Check	ISC	Daily prior to analysis.
Continuing Calibration Verification	CCV	At the beginning (ISC) and every 10 samples.
Continuing Calibration Blank	CCB	After each bracketing CCV.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Item ¹	Acronym	Frequency
RT Window	RTW	During ICAL and at the beginning of each analytical sequence.
Bile Check	TDCA	With each ICAL and daily with each CCV.
Method Blank	MB	1 per batch of 20 or fewer samples.
Laboratory Control Samples	LCS & LLLCS	1 pair per batch of 20 or fewer samples.
Laboratory Duplicate	DUP	See Section 11.5.1.

¹ Extracted (EIS) and Non-extracted Internal Standards (NIS) are included in each QC item.

11.5.1 Laboratory Duplicate (AKA Matrix Duplicate)

A laboratory duplicate is a duplicate preparation and analysis of a sample whose aliquot was taken from the same container as the parent sample and used to measure precision.

EPA Method 1633 does not require laboratory duplicates for NPDES compliance monitoring, but the method acknowledges that a duplicate may be needed on a project-specific basis. Some accreditation programs, such as DoD ELAP, require a duplicate for each sample of certain matrices, such as AFFF or F3.

To support project needs, the laboratory will perform a duplicate when the customer has requested this measure, designated which sample from their sampling event to use as the duplicate on the chain-of-custody (COC), and have provided sufficient additional sample material for the test.

However, customers should be advised that when the duplicate is taken from a different container, which is necessary for certain matrices, the duplicate effectively becomes a field duplicate and the results for precision are inclusive of potential bias from sampling error.

Instructions for DUP to Laboratory Personnel by Matrix:

- **NPW** – Laboratory duplicates must be prepared from a separate container because the entire sample is consumed during routine preparation and extraction and subsampling is only allowed for primary dilutions of samples expected to have high concentrations of target analytes, or to manage sample matrix characteristics.

If a duplicate is requested and no sample has been designated for use, confer with the PM to determine which sample to use and verify that more than two (2) 500 mL containers were received. If two or less containers were received; there is insufficient sample volume available to perform the duplicate – notify the PM of the sampling nonconformity. If more than two (2) 500 mL containers were received, use one (1) for the parent sample and one (1) for the duplicate.

Do not perform reduced volume extraction to provide a DUP.

- **Surface Wipes** – The entire sample is consumed during preparation and extraction. Given the nature of the matrix and collection technique, this sample matrix is not suitable for analysis of a laboratory duplicate.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633 Effective Date: 10/14/2024

If a duplicate is requested, notify the PM that the matrix is not suitable for this determination and cancel the request.

- **AFFF/F3, Soil/Sediment, Biosolid, Articles of Commerce, and Tissue** – A laboratory duplicate is performed when requested by the customer and when there is enough material to take the duplicate aliquot from the same container. Note that DoD QSM 5.4/6.0 requires a laboratory duplicate on a sample basis for AFFF and F3 matrices. Thus, for DoD project work, customers must provide sufficient mass for each sample to meet this program requirement.

11.5.2 Extracted Internal Standards (EIS)

To assess method performance on the sample matrix, the laboratory must spike all samples and QC with the EIS solution. Analyze each sample according to the procedures in this SOP. Compute the percent recovery of the EIS compound concentration using the NIS quantitation method; see Appendix A, Table 9, for acceptance criteria.

If the recovery of any EIS compound is outside of the acceptance limits in the original, undiluted analysis, prepare and analyze a limited extract dilution and/or a re-extraction using a reduced sample amount.

Prior to calculating EIS percent recovery, the calculated concentration of the EIS are always adjusted to account for any extract dilutions prepared for analysis.

11.5.3 Non-extracted Internal Standard (NIS)

To assess method performance, the laboratory must spike all sample extracts with the NIS solution prior to analysis. Percent recovery of the NIS compound is calculated by comparing the measured response in each analysis to the average response of the same compound measured in the ICAL.

Calculate the ratio of the NIS peak areas from the QC and field samples relative to the mean area of the corresponding NIS in the most recent initial calibration to check for possible bad injections of NIS solution or loss of instrument sensitivity. If the NIS areas are low for all the field samples and QC samples in the analytical batch, it suggests a loss of instrument sensitivity, while low areas in only some field or QC samples suggests a possible bad injection. See Appendix A, Table 9, for acceptance criteria.

11.5.4 Sample Dilutions

Samples with quantified target analytes exceeding the upper calibration range must be diluted and reanalyzed to bring the result within the calibration range. However, if it is known that the analytes are not compounds of concern for the project, test results may be reported over the calibration range, if they are qualified to disclose the situation and documented client consent for reporting this way is obtained.

If the extract dilution required is greater than 10X, repeat sample preparation using a reduced sample amount, guided by the magnitude of the original over-range results.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

The response of the EIS in the diluted extract must meet the S/N and RT requirements for qualitative identification and must be within the limits specified in Appendix A, Table 9. If EIS criteria are not met, then the compound cannot be measured reliably in the diluted extract, and sample preparation and analysis must be repeated with a smaller subsample.

The amount used for re-extraction should be consistent with the dilution applied to the original extract. For example, if a 10X dilution was needed for an aqueous sample, 50 mL of sample should be subsampled for re-prep and extraction. (Divide the original amount of sample prepared by 10 – this is the sample amount.)

12.0 DATA REVIEW & CORRECTIVE ACTION

12.1 Data Review

Review all data and test results before releasing the test report to ensure that the analytical record is complete and properly documented, criteria were met, appropriate corrective actions were taken for QC failures or other nonconformances, and test results are properly qualified when necessary.

Initial review, known as primary review, is done by the individual who performed the task.

Review of the data and test results done by someone other than the person who performed the task is known as secondary review, and it is done by a qualified peer or supervisor. Secondary review is done to ensure any errors missed during primary review are caught and corrected before the test report is delivered. Secondary review is also done to ensure that SOPs were followed, calibration and instrument performance met specifications, QC criteria were satisfied, and/or proper corrective actions were implemented, qualitative identification and quantitative measurements are accurate, any manual integrations are justified, documented, and approved per the PAS Corporate SOP for manual integration, calculations are accurate, and the analytical record is complete and traceable, with final test results properly qualified.

In addition to primary and secondary review, a completeness check is performed by project management personnel to confirm that the test report is complete and ready for release.

Refer to laboratory SOP ENV-SOP-MIN4-0092 *Data Review Process*, or however named, for specific instructions and requirements for each step of the data review process.

12.2 Corrective Action

Corrective action is required when QC or sample results fall outside the acceptance criteria. For a complete summary of QC parameters, acceptance criteria, and recommended corrective actions associated with this test method, refer to Appendix A, Table 7. The primary analyst has primary responsibility for taking corrective action when QA/QC criteria are not met. Secondary data reviewers must verify that appropriate action was taken and/or that results reported with QC failure are properly qualified.

Corrective action is also required when instrumental carryover contamination is suspected. Samples analyzed after a sample with a detection above the working calibration range must be checked for carryover and reanalyzed if carryover is suspected. If field sample analyte

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

concentrations exceed the highest calibration standard and the same analytes in the following field sample or in consecutive following field samples also exceed the MB acceptance criteria, the affected samples should be reanalyzed using a fresh aliquot of the sample extract.

If corrective actions are not taken or are unsuccessful, the decision and outcome must be documented in the analytical record and test results qualified to alert the end data user of the nonconformity.

13.0 POLLUTION PREVENTION & WASTE MANAGEMENT

Refer to laboratory SOP ENV-SOP-MIN4-0098 *Waste Handling and Management*, or however named, for waste handling and management practices pertaining to the waste streams created at this facility.

Pace® proactively seeks ways to minimize waste generated during work processes. Some examples of pollution prevention include but are not limited to reduced solvent extraction, solvent capture, use of reusable cycletainers for solvent management, and real-time purchasing.

The EPA requires that laboratory waste management practices comply with all applicable federal and state laws and regulations. Excess reagents, samples, and method process wastes are characterized and disposed of in an acceptable manner in accordance with the Pace® Chemical Hygiene Plan / Safety Manual. Refer to this manual for these procedures.

14.0 MODIFICATIONS

The procedures in this SOP have been modified from the reference method to improve overall performance. Supporting documentation to demonstrate equivalency and comparability, when required, is maintained by the Quality Department.

Modification	Section of Reference Method Modified	Justification for Modification	Validation of Modification
Changed EIS quantitation association for PFMPA, 7:3FTCA, ADONA, 9Cl-PF3ONS, & 11Cl-PF3OUdS	Section 20, Table 10	Improved performance (allowable modification), more appropriate correlation	MDL, IPR, PT
Updated product ion used for measurement of 13C4-PFOA from m/z 172 to m/z 372	Section 20, Table 10	Match ion transition used for EIS 13C8-PFOA and native PFOA	MDL, IPR, PT
Increased the size of the SPE sorbent bed to 250 mg and tube size to 20 mL	Section 6.7.1	Improved performance (allowable modification); wider cartridge improves sample loading, reduces chances of clogs	MDL, IPR, PT
Increased flow rate to allow up to 10 mL/min	Section 12.1.4	Comparable and validated performance	MDL, IPR, PT
Omit TSS determination for aqueous samples; utilize centrifugation to minimize clogging of SPE	Section 11.1 & 11.2.4	Allowable modifications per method	Not applicable.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Modification	Section of Reference Method Modified	Justification for Modification	Validation of Modification
Solid matrix extraction: eliminated 3 rd liquid-solid extraction step and omitted concentration step	Section 11.3.4, 11.3.6, and 11.3.9	Improved performance & reduced sample processing time	MDL, IPR, PT
Use pre-packed GCB cartridges; omit syringe filtration	All references to carbon cleanup	Improved performance, reduced sample processing time, & ease of use	MDL, IPR, PT
Tissue matrix extraction: reduced shake time, eliminated 3 rd liquid-solid extraction, and omitted concentration step	Section 11.4.4, 11.4.6, 11.4.8	Comparable and validated performance	MDL, IPR, PT

15.0 RESPONSIBILITIES

PAS employees that perform any part this procedure in their work activities must have a signed Read and Acknowledgement Statement (R&A) in their training file for the version(s) of the SOP that were in effect during the time the employee performed the activity.

PAS supervisors and managers, however named, are responsible for training employees on the procedures in this SOP, implementing the SOP in the work area, and monitoring on-going adherence to the SOP in the work area(s) they oversee.

PAS employees are responsible for following the procedures in this SOP. Unauthorized deviations or departures from this SOP are not allowed except with documented approval from the local QM and only when those deviations do not violate the Pace® Code of Ethics or Professional Conduct (COR-POL-0004) or associated policy and procedure(s). Hand-edits or manual changes to the SOP are not permitted. If a change is desired or necessary, Pace® employees must follow the procedures for document revision specified in corporate SOPs ENV-SOP-CORQ-0015, *Document Management* and ENV-SOP-CORQ-0016, *SOP for Creation of SOP and SWI*.

16.0 ATTACHMENTS

- Appendix A: Tables
 - Table 1: Analyte Name / Abbreviation / CAS Number Cross-Reference Table
 - Table 2: Routine Analyte List and LOQ by Matrix
 - Table 3: Sample Collection, Preservation, and Holding Time Requirements
 - Table 4: Required Instruments, Equipment, and Supplies
 - Table 5: Prepared Concentration of Working Calibration Standards
 - Table 6: Identification and Quantitation Ions by Analyte, EIS, and NIS
 - Table 7: QC Acceptance Criteria, Corrective Action and Data Qualification Requirements
 - Table 8: LCS/LLCS Acceptance Limits
 - Table 9: EIS and NIS Acceptance Limits

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

- Table 10: IPR Limits for Method Validation
- Appendix B: Reference Materials – Reagents, Standards & Formulations
 - Table 1: Purchased Reagents
 - Table 2: Purchased Standard Solutions
 - Table 3: Laboratory Prepared Reagent Formulation Table
 - Table 4: Laboratory Prepared Intermediate Standard Solutions
 - Table 5: Laboratory Prepared Working Standard Solutions
- Appendix C: Sample Screening Protocol
- Appendix D: Subsampling Procedure for Aqueous Matrices
- Appendix E: Aqueous Sample Preparation – Settled Solids Depth > ½ cm
- Appendix F: Biphasic Sample Separation Procedure
- Appendix G: SPE Clog Mitigation Procedure
- Appendix H: Particle Size Reduction by Cryomill for Articles of Commerce

17.0 REFERENCES

- ENV-POL-CORQ-0005, *Acceptable Calibration Practices*, current version.
- ENV-SOP-CORQ-0006, *Manual Integration*, current version.
- ENV-SOP-CORQ-0011, *Method Validation and Instrument Verification*, current version.
- ENV-SOP-CORQ-0015, *Document Management*, current version.
- ENV-SOP-CORQ-0016, *Standard Operating Procedures and Standard Work Instructions*, current version.
- COR-POL-0004, *Code of Ethics and Professional Conduct*, current version.
- COR-MAN-001, *Pace® Safety Manual*, current version.
- ENV-MAN-MIN4-0001, *Quality Manual*, current version.
- TNI Standard, “Volume 1 Management and Technical Requirements for Laboratories Performing Environmental Analysis”, current version.
- EPA. Method 1633, Analysis of Per- and Polyfluoroalkyl Substances (PFAS) in Aqueous, Solid, Biosolids, and Tissue Samples by LC-MS/MS. January 2024.
- DoD-EDQW and DOE-DQW. DoD/DOE Quality Systems Manual (QSM) for Environmental Laboratories, Version 6.0, 2021 – Table B-24.
- Standard Method for the Examination of Water and Wastewater, 23rd Edition, 2017 – Total, Fixed, and Volatile Solids in Solid and Semisolid Samples, ASTM D2974-07.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

- Articles of Commerce: 3M, STANDARD TEST METHOD, Determination of PFOA in articles of commerce: disassembly, disjointment, and mechanical preparation of solid samples, sample preparation of aqueous polymer systems, sample extraction, and LC-MS quantitative analysis, June 2020.

18.0 REVISION HISTORY

Revisions Made from Prior Version

Section	Description of Change
All	New document.

Document Succession: This version replaces the following document(s):

Document Number	Version	Document Title	Effective Date:
N/A	N/A	N/A	N/A

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633 Effective Date: 10/14/2024

Appendix A: Tables

Table 1: Analyte Name / Abbreviation / CAS Number Cross-Reference Table

Analyte Name	Abbreviation	CAS #
Perfluoropropanoic acid	PFPrA	422-64-0
Perfluorobutanoic acid	PFBA	375-22-4
Perfluoropentanoic acid	PFPeA	2706-90-3
Perfluorohexanoic acid	PFHxA	307-24-4
Perfluoroheptanoic acid	PFHpA	375-85-9
Perfluorooctanoic acid	PFOA	335-67-1
Perfluorononanoic acid	PFNA	375-95-1
Perfluorodecanoic acid	PFDA	335-76-2
Perfluoroundecanoic acid	PFUnA	2058-94-8
Perfluorododecanoic acid	PFDoA	307-55-1
Perfluorotridecanoic acid	PFTTrDA	72629-94-8
Perfluorotetradecanoic acid	PFTeDA	376-06-7
Perfluorobutanesulfonic acid	PFBS	375-73-5
Perfluoropentanesulfonic acid	PFPeS	2706-91-4
Perfluorohexanesulfonic acid	PFHxS	355-46-4
Perfluoroheptanesulfonic acid	PFHpS	375-92-8
Perfluorooctanesulfonic acid	PFOS	1763-23-1
Perfluorononanesulfonic acid	PFNS	68259-12-1
Perfluorodecanesulfonic acid	PFDS	335-77-3
Perfluorododecanesulfonic acid	PFDoS	79780-39-5
1H,1H, 2H, 2H-Perfluorohexane sulfonic acid	4:2 FTS	757124-72-4
1H,1H, 2H, 2H-Perfluorooctane sulfonic acid	6:2 FTS	27619-97-2
1H,1H, 2H, 2H-Perfluorodecane sulfonic acid	8:2 FTS	39108-34-4
Perfluorooctanesulfonamide	PFOSA	754-91-6
N-methyl perfluorooctanesulfonamide	NMeFOSA	31506-32-8
N-ethyl perfluorooctanesulfonamide	NEtFOSA	4151-50-2
Bis (trifluoromethylsulfonyl)amine	TFSI	82113-65-3
N-methyl perfluorooctanesulfonamidoacetic acid	NMeFOSAA	2355-31-9
N-ethyl perfluorooctanesulfonamidoacetic acid	NEtFOSAA	2991-50-6
N-methyl perfluorooctanesulfonamidoethanol	NMeFOSE	24448-09-7
N-ethyl perfluorooctanesulfonamidoethanol	NEtFOSE	1691-99-2
Hexafluoropropylene oxide dimer acid (GenX)	HFPO-DA	13252-13-6
4,8-Dioxa-3H-perfluorononanoic acid	ADONA	919005-14-4

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633 Effective Date: 10/14/2024

Analyte Name	Abbreviation	CAS #
Perfluoro-3-methoxypropanoic acid	PFMPA	377-73-1
Perfluoro-4-methoxybutanoic acid	PFMBA	863090-89-5
Nonafluoro-3,6-dioxaheptanoic acid	NFDHA	151772-58-6
9-Chlorohexadecafluoro-3-oxanonane-1-sulfonic acid	9Cl-PF3ONS	756426-58-1
11-Chloroeicosafluoro-3-oxaundecane-1-sulfonic acid	11Cl-PF3OUdS	763051-92-9
Perfluoro(2-ethoxyethane)sulfonic acid	PFEESA	113507-82-7
3-Perfluoropropyl propanoic acid (FPrPA)	3:3 FTCA	356-02-5
2H,2H,3H,3H-Perfluorooctanoic acid (FPePA)	5:3 FTCA	914637-49-3
3-Perfluoroheptyl propanoic acid (FHpPA)	7:3 FTCA	812-70-4
Extracted Internal Standards (EIS)		
Perfluoro-n-[13C3] propanoic acid	13C3-PFPrA	NA
Perfluoro-n-[13C4] butanoic acid	13C4-PFBA	NA
Perfluoro-n-[13C5] pentanoic acid	13C5-PFPeA	NA
Perfluoro-n-[1,2,3,4,6-13C5] hexanoic acid	13C5-PFHxA	NA
Perfluoro-n-[1,2,3,4-13C4] heptanoic acid	13C4-PFHpA	NA
Perfluoro-n-[13C8] octanoic acid	13C8-PFOA	NA
Perfluoro-n-[13C9] nonanoic acid	13C9-PFNA	NA
Perfluoro-n-[1,2,3,4,5,6-13C6] decanoic acid	13C6-PFDA	NA
Perfluoro-n-[1,2,3,4,5,6,7-13C7] undecanoic acid	13C7-PFUnA	NA
Perfluoro-n-[1,2-13C2] dodecanoic acid	13C2-PFDoA	NA
Perfluoro-n-[1,2-13C2] tetradecanoic acid	13C2-PFTeDA	NA
Perfluoro-1-[2,3,4-13C3] butanesulfonic acid	13C3-PFBS	NA
Perfluoro-1-[1,2,3-13C3] hexanesulfonic acid	13C3-PFHxS	NA
Perfluoro-1-[13C8] octanesulfonic acid	13C8-PFOS	NA
1H,1H,2H,2H-Perfluoro-1-[1,2-13C2] hexanesulfonic acid	13C2-4:2FTS	NA
1H,1H,2H,2H-Perfluoro-1-[1,2-13C2] octanesulfonic acid	13C2-6:2FTS	NA
1H,1H,2H,2H-Perfluoro-1-[1,2-13C2] decanesulfonic acid	13C2-8:2FTS	NA
Perfluoro-1-[13C8] octanesulfonamide	13C8-PFOSA	NA
N-methyl-D3-perfluoro-1-octanesulfonamide	D3-NMeFOSA	NA
N-ethyl-D5-perfluoro-1-octanesulfonamide	D5-NEtFOSA	NA
N-methyl-D3-perfluoro-1-octanesulfonamidoacetic acid	D3-NMeFOSAA	NA
N-ethyl-D5-perfluoro-1-octanesulfonamidoacetic acid	D5-NEtFOSAA	NA
N-methyl-D7-perfluorooctanesulfonamidoethanol	D7-NMeFOSE	NA
N-ethyl-D9-perfluorooctanesulfonamidoethanol	D9-NEtFOSE	NA
Tetrafluoro-2-heptafluoropropoxy-13C3-propanoic acid	13C3-HFPO-DA	NA

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Analyte Name	Abbreviation	CAS #
NIS		
Perfluoro-n-[2,3,4-13C3] butanoic acid	13C3-PFBA	NA
Perfluoro-n-[1,2-13C2]hexanoic acid	13C2-PFHxA	NA
Perfluoro-n-[1,2,3,4-13C4]octanoic acid	13C4-PFOA	NA
Perfluoro-n-[1,2,3,4,5-13C5] nonanoic acid	13C5-PFNA	NA
Perfluoro-n-[1,2-13C2]decanoic acid	13C2-PFDA	NA
Perfluoro-1-hexane[18O2]sulfonic acid	18O2-PFHxS	NA
Perfluoro-n-[1,2,3,4-13C4] octanesulfonic acid	13C4-PFOS	NA

Table 2: Routine Analyte List and LOQ¹ by Matrix

Abbreviation	CAS #	Aqueous (NPW) (ng/L)	Leachate (Landfill/TCLP/ SPLP) (ng/L)	Solids (Soil/Sediment) (ng/g)	Biosolid (ng/g)	Tissue (ng/g)
PFPPrA	422-64-0	6.4	32	0.8	8	2.0
PFBA	375-22-4	6.4	32	0.8	8	2.0
PFPeA	2706-90-3	3.2	16	0.4	4	1.0
PFHxA	307-24-4	1.6	8	0.2	2	0.5
PFHpA	375-85-9	1.6	8	0.2	2	0.5
PFOA	335-67-1	1.6	8	0.2	2	0.5
PFNA	375-95-1	1.6	8	0.2	2	0.5
PFDA	335-76-2	1.6	8	0.2	2	0.5
PFUnA	2058-94-8	1.6	8	0.2	2	0.5
PFDoA	307-55-1	1.6	8	0.2	2	0.5
PFTTrDA	72629-94-8	1.6	8	0.2	2	0.5
PFTeDA	376-06-7	1.6	8	0.2	2	0.5
PFBS	375-73-5	1.6	8	0.2	2	0.5
PFPeS	2706-91-4	1.6	8	0.2	2	0.5
PFHxS	355-46-4	1.6	8	0.2	2	0.5
PFHpS	375-92-8	1.6	8	0.2	2	0.5
PFOS	1763-23-1	1.6	8	0.2	2	0.5
PFNS	68259-12-1	1.6	8	0.2	2	0.5
PFDS	335-77-3	1.6	8	0.2	2	0.5
PFDoS	79780-39-5	1.6	8	0.2	2	0.5
4:2 FTS	757124-72-4	6.4	32	0.8	8	2.0

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Abbreviation	CAS #	Aqueous (NPW) (ng/L)	Leachate (Landfill/TCLP/ SPLP) (ng/L)	Solids (Soil/Sediment) (ng/g)	Biosolid (ng/g)	Tissue (ng/g)
6:2 FTS	27619-97-2	6.4	32	0.8	8	2.0
8:2 FTS	39108-34-4	6.4	32	0.8	8	2.0
PFOSA	754-91-6	1.6	8	0.2	2	0.5
NMeFOSA	31506-32-8	1.6	8	0.2	2	0.5
NEtFOSA	4151-50-2	1.6	8	0.2	2	0.5
TFSI	82113-65-3	3.2	16	0.4	4	1.0
NMeFOSAA	2355-31-9	1.6	8	0.2	2	0.5
NEtFOSAA	2991-50-6	1.6	8	0.2	2	0.5
NMeFOSE	24448-09-7	16	80	2.0	20	5.0
NEtFOSE	1691-99-2	16	80	2.0	20	5.0
HFPO-DA	13252-13-6	6.4	32	0.8	8	2.0
ADONA	919005-14-4	6.4	32	0.8	8	2.0
PFMPA	377-73-1	3.2	16	0.4	4	1.0
PFMBA	863090-89-5	3.2	16	0.4	4	1.0
NFDHA	151772-58-6	3.2	16	0.4	4	1.0
9Cl-PF3ONS	756426-58-1	6.4	32	0.8	8	2.0
11Cl-PF3OUdS	763051-92-9	6.4	32	0.8	8	2.0
PFEESA	113507-82-7	3.2	16	0.4	4	1.0
3:3 FTCA	356-02-5	8	40	1.0	10	2.5
5:3 FTCA	914637-49-3	40	200	5.0	50	12.5
7:3 FTCA	812-70-4	40	200	5.0	50	12.5

¹ LOQ effective on SOP effective date; values are subject to change. If you are an external party using this SOP for reference, please contact the laboratory for the most up-to-date information.

Table 3: Sample Collection, Preservation¹ and Holding Time Requirements

Matrix	Container per Sample	Minimum Sample Size ²	Preservation	Holding Time Collection to Prep	Holding Time Prep to Analysis
Aqueous: Non-potable Water (NPW)	Linerless HDPE 2 x 500 mL + 1 x 125 mL ³	500 mL	Option 1: 0-6°C Option 2: ≤20°C	Option 1: 28 Days Option 2: 90 Days	28 Days
Aqueous: Landfill Leachate	Linerless HDPE 2 x 125 mL	100 mL	Option 1: 0-6°C Option 2: ≤20°C	Option 1: 28 Days Option 2: 90 Days	28 Days
Aqueous: TCLP or SPLP	See TCLP / SPLP SOP(s): Leachates are generated using methods for TCLP and/or SPLP and then processed per this SOP for PFAS.				

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Matrix	Container per Sample	Minimum Sample Size ²	Preservation	Holding Time Collection to Prep	Holding Time Prep to Analysis
Solid: Soil / Sediment	Linerless Polypropylene 1 x 3 oz/90 mL wide mouth container	5 g	Option 1: 0-6°C Option 2: ≤20°C	Option 1: 90 Days Option 2: 90 Days	28 Days
Solid: Biosolid	Linerless HDPE 1x 125 mL	0.5 g	Option 1: 0-6°C Option 2: ≤20°C	Option 1: 90 Days Option 2: 90 Days	28 Days
Tissue: Unhomogenized Fish	Sampling team should wrap the fish in aluminum foil or insert into food-grade polyethylene tubing or a resealable zip-top bag for shipment to the lab.	Whole Fish or Fillet (for homogenization)	≤20°C	45 Days	N/A
Tissue: Homogenized Fish	Linerless Polypropylene 1 x 3 oz/90 mL wide mouth container	2 g	≤20°C	90 Days	28 Days
Solid: Articles of Commerce	PFAS Free Container (HDPE bottle, resealable zip-top bag)	2 g	0-6°C	365 Days	28 Days
Aqueous: AFFF/F3	Linerless HDPE 1 x 125 mL or 1 x 90 mL	0.02 g	0-6°C	28 Days	28 Days

¹ Preservation at time of collection and on receipt at laboratory. Unless otherwise specified, samples are stored after receipt in the laboratory under the same conditions as preservation until time of preparation or analysis.

² Nominal amount of sample required for each discrete test. Solid and biosolid sample amounts reflect dry weight.

³ Used for pre-screening analyses.

Table 4: Required Instruments, Equipment, and Supplies

Item	Specification	Vendor	Model	Catalog #
LC MS/MS	Tandem Quadrupole Mass Spectrometer	Agilent	6495C	N/A
Analytical Column	2.1 x 50 mm (1.8 µm)	Agilent	Zorbax RRHD Eclipse Plus	959757-902
Guard Cartridge/Column	2.1 x 5 mm, 1.8 µm	Agilent	Zorbax Eclipse Plus C18	821725-901
Trap / Delay Colum	4.6 x 30 mm	Agilent	Infinity Lab PFC Delay Column	5062-8100
Oven	Stable Temperature of 105±5°C	Varied	Varied	Varied

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Item	Specification	Vendor	Model	Catalog #
Analytical Balance	Measurement to 0.0001g	Varied	Varied	Varied
Top Loading Balance	Measurement to 0.01g	Varied	Varied	Varied
Ultrasonic Mixer (Sonicator)	Time controlled	Varied	Varied	Varied
Vortex Mixer	Multi-Tube	Fisher Scientific	Fisherbrand™ Digital Multi-Tube Vortexer	02-215-542
	Alternative Option: Single Tube	Fisher Scientific	Fisherbrand™ Pulsing Vortex Mixer	02215375
Mixing Table	Variable Speed	Fisher Scientific	3Multi-Plate Shaker	88-861-021
Centrifuge	3000 RPM (~2000 RCF)	VWR Sigma- Aldrich	Beckman Allegra 6 Hettich Rotanta 460	See Catalog
Centrifuge Bottles	Sterile, Polypropylene, Conical, 500mL	Fisher Scientific	Corning™ Polypropylene Centrifuge Tubes, Sterile	431123
Vacuum Manifold	Designed for SPE Cartridges	Sigma-Aldrich	Visiprep™ SPE	57265
Manifold Liners	Compatible with SPE manifold	Millipore Sigma	Disposable Liners for Visiprep DL	57059
		Restek	Quick Replace Liners, PTPE or Resprep QR Vacuum Manifolds	28310
Vacuum Pump	Vacuum of ~10-15 inches of mercury	MilliporeSigma,	WP6111560 115V,60 HZ, 3.5A,	See Catalog
Vacuum Tubing	1/4"ID, 5/8" OD, 3/16" Wall	Varied	N/A	Various
SPE Cartridges	Designed for PFAS	Phenomenex (locally approved)	Strata-X-AW 33uM Polymeric Weak Anion-PFAS 250mg/20m Tubes	8B-S541-FEG (formerly CSO-9216)
SPE Reservoirs	60mL, polypropylene	Phenomenex	Empty SPE Tubes 60cc	AHO-7189
Adapter Caps	To fit 12, 20, 60mL SPE tubes	MilliporeSigma	Cartridge Adapter	57267
Adjustable-volume Mechanical Pipettes	10uL to 10mL	Various	Various	Various
Disposable Pipette Tips	HDPE or Polypropylene: volume of 10uL to 5mL	Various	Various	Various
Solvent Evaporator	Nitrogen Evaporation	Various	Automated: II	415001
Carbon Cartridges	Graphitized Carbon Black Solid Phase Extraction Absorbent	Phenomenex	Strata GCB, 25mg/1mL Pass Through Cartridge	8B-S528-CAJ
Glass Wool	Silanized	Millipore Sigma	Supelco Glass Wool, Salinized	20411
Water System	Capable of producing Type I water	Millipore	Synergy	Varies
pH Indicator Strips	Range 0-14, Sensitivity 0.5 pH unit 0 to 9, 1 pH unit 9-14	MilliporeSigma	Whatman Panepha™	WHA0360005
Bottles	HDPE or Polypropylene with linerless caps of same material	QEC (Pace Approved)	90 mL	6214-003PPGBC-P
			125 mL	6213-U004BC-P

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl
	Substances by EPA 1633
Effective Date: 10/14/2024	

Item	Specification	Vendor	Model	Catalog #
Tubes	HDPE or Polypropylene with linerless caps of same material	Fisher Scientific	250 mL	6212-Q008BC-P
			500 mL	6212-Q016/BC-150 PACE
			Falcon™ 15 mL conical	05-527-90
			Falcon™ 50 mL conical	14-432-22
Containers	Narrow Mouth HDPE	Thermo	4 mL	2002-9125
			8 mL	2002-9025
			15 mL	2002-9050
	Polyethylene	Grainger	Cryogenic Vial 2 mL	6EMV1
	Microcentrifuge Tube	Fisher	1.5 mL	05-408-129
Screw Top Vials	Polypropylene vial	Agilent	250 µL	5190-2243
Screw Top Cap	9mm clear propylene, thin polypropylene-silicone septa	Agilent	Screw Style Cap	5191-8151
Single-step Filter Vials	0.2 µm nylon membrane	Restek	SINGLE StEP® Standard Filter Vials	25891
Surface Wipe	Pre-wetted with methanol	Environmental Express (locally approved)	Ghost Wipes for Methamphetamine testing	SC5000
Ottawa Sand	20-30 Mesh Ottawa Sand	Fisher	Ottawa Sand 20-30 Mesh For Cement Testing	S23-3
Reference Tissue	Homogenized Cod Fillet	Varies	N/A	N/A
Cryogenic Mill	25 mm grinding ball	Retsch	CryoMill	20.749.0001
Scissors or Heavy Plate Shears	Stainless Steel	Varies	Varies	Varies
Cleaning Brushes	Various Size	Varies	Varies	Varies
Polishing Cloths				
Cotton Balls / Swabs				

Table 5: Prepared Concentration of Working Calibration Standards (ng/mL)¹

Compound	L1	L2 ²	L3	L4	L5 ³	L6	L7	L8	L9
Perfluoroalkyl carboxylic acids									
PFPrA	0.4	0.8	2	10	20	50	100	200	250
PFBA	0.4	0.8	2	10	20	50	100	200	250
PFPeA	0.2	0.4	1	5	10	25	50	100	125
PFHxA	0.1	0.2	0.5	2.5	5	12.5	25	50	62.5
PFHpA	0.1	0.2	0.5	2.5	5	12.5	25	50	62.5

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Compound	L1	L2 ²	L3	L4	L5 ³	L6	L7	L8	L9
PFOA	0.1	0.2	0.5	2.5	5	12.5	25	50	62.5
PFNA	0.1	0.2	0.5	2.5	5	12.5	25	50	62.5
PFDA	0.1	0.2	0.5	2.5	5	12.5	25	50	62.5
PFUnA	0.1	0.2	0.5	2.5	5	12.5	25	50	62.5
PFDaA	0.1	0.2	0.5	2.5	5	12.5	25	50	62.5
PFTTrDA	0.1	0.2	0.5	2.5	5	12.5	25	50	62.5
PFTeDA	0.1	0.2	0.5	2.5	5	12.5	25	50	62.5
Perfluoroalkyl sulfonic acids									
PFBS	0.089	0.178	0.444	2.22	4.44	11.1	22.2	44.4	55.5
PFPeS	0.094	0.188	0.470	2.35	4.70	11.8	23.5	47.0	58.8
PFHxS	0.091	0.182	0.456	2.28	4.56	11.4	22.8	45.6	57.0
PFHpS	0.095	0.190	0.476	2.38	4.76	11.9	23.8	47.6	59.5
PFOS	0.093	0.186	0.464	2.32	4.64	11.6	23.2	46.4	58.0
PFNS	0.096	0.192	0.480	2.40	4.80	12.0	24.0	48.0	60.0
PFDS	0.096	0.193	0.482	2.41	4.82	12.1	24.1	48.2	60.3
PFDoS	0.097	0.194	0.484	2.42	4.84	12.1	24.2	48.4	60.5
Fluorotelomer sulfonic acids									
4:2FTS	0.375	0.750	1.87	9.37	18.7	46.9	93.7	187	234
6:2FTS	0.380	0.761	1.90	9.51	19.0	47.6	95.1	190	238
8:2FTS	0.384	0.768	1.92	9.6	19.2	48	96	192	240
Perfluorooctane sulfonamides									
PFOSA	0.1	0.2	0.5	2.5	5	12.5	25	50	62.5
NMeFOSA	0.1	0.2	0.5	2.5	5	12.5	25	50	62.5
NEtFOSA	0.1	0.2	0.5	2.5	5	12.5	25	50	62.5
Perfluoromethane sulfonamide									
TFSI	0.2	0.4	1	5	10	25	50	100	125
Perfluorooctane sulfonamidoacetic acids									
NMeFOSAA	0.1	0.2	0.5	2.5	5	12.5	25	50	62.5
NEtFOSAA	0.1	0.2	0.5	2.5	5	12.5	25	50	62.5
Perfluorooctane sulfonamide ethanols									
NMeFOSE	1	2	5	25	50	125	250	500	625
NEtFOSE	1	2	5	25	50	125	250	500	625
Per- and polyfluoroether carboxylic acids									
HFPO-DA	0.4	0.8	2	10	20	50	100	200	250
ADONA	0.378	0.756	1.89	9.45	18.9	47.25	94.5	189	236

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Compound	L1	L2 ²	L3	L4	L5 ³	L6	L7	L8	L9
PFMPA	0.2	0.4	1	5	10	25	50	100	125
PFMBA	0.2	0.4	1	5	10	25	50	100	125
NFDHA	0.2	0.4	1	5	10	25	50	100	125
Ether sulfonic acids									
9CI-PF3ONS	0.373	0.746	1.87	9.33	18.7	46.7	93.3	187	233
11CI-PF3OUDS	0.377	0.754	1.89	9.43	18.9	47.2	94.3	189	236
PFEESA	0.178	0.357	0.892	4.46	8.92	22.3	44.6	89.2	112
Fluorotelomer Carboxylic Acids									
3:3FTCA	0.5	1	2.5	12.5	25	62.5	125	250	313
5:3FTCA	2.5	5	12.5	62.5	125	313	625	1250	1560
7:3FTCA	2.5	5	12.5	62.5	125	313	625	1250	1560
Extracted Internal Standards (EIS)									
13C3-PFPrA	10	10	10	10	10	10	10	10	10
13C4-PFBA	10	10	10	10	10	10	10	10	10
13C5-PFPeA	5	5	5	5	5	5	5	5	5
13C5-PFHxA	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5
13C4-PFHpA	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5
13C8-PFOA	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5
13C9-PFNA	1.25	1.25	1.25	1.25	1.25	1.25	1.25	1.25	1.25
13C6-PFDA	1.25	1.25	1.25	1.25	1.25	1.25	1.25	1.25	1.25
13C7-PFUnA	1.25	1.25	1.25	1.25	1.25	1.25	1.25	1.25	1.25
13C2-PFDoA	1.25	1.25	1.25	1.25	1.25	1.25	1.25	1.25	1.25
13C2-PFTeDA	1.25	1.25	1.25	1.25	1.25	1.25	1.25	1.25	1.25
13C3-PFBS	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5
13C3-PFHxS	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5
13C8-PFOS	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5
13C2-4:2FTS	5	5	5	5	5	5	5	5	5
13C2-6:2FTS	5	5	5	5	5	5	5	5	5
13C2-8:2FTS	5	5	5	5	5	5	5	5	5
13C8-PFOSA	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5
D3-NMeFOSA	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5
D5-NEtFOSA	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5
D3-NMeFOSAA	5	5	5	5	5	5	5	5	5
D5-NEtFOSAA	5	5	5	5	5	5	5	5	5
D7-NMeFOSE	25	25	25	25	25	25	25	25	25

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Compound	L1	L2 ²	L3	L4	L5 ³	L6	L7	L8	L9
D9-NEtFOSE	25	25	25	25	25	25	25	25	25
13C3-HFPODA	10	10	10	10	10	10	10	10	10
Non-Extracted Internal Standards (NIS)									
13C3-PFBA	5	5	5	5	5	5	5	5	5
13C2-PFHxA	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5
13C4-PFOA	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5
13C5-PFNA	1.25	1.25	1.25	1.25	1.25	1.25	1.25	1.25	1.25
13C2-PFDA	1.25	1.25	1.25	1.25	1.25	1.25	1.25	1.25	1.25
18O2-PFHxS	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5
13C4-PFOS	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5

¹ Concentration of each target analyte, EIS, and NIS in each calibration level when the formulations specified in Appendix B are used to prepare the working standards.

² L2 is used as the ISC and verifies the LOQ.

³ L5 is used as the CCV.

Table 6: Identification and Quantification Ions by Analyte, EIS, NIS

Abbreviation	CAS #	Ion Mass			Quantification Reference
		Parent	Quantification	Confirmation	
PFPrA	422-64-0	163	119	NA	13C3-PFPrA
PFBA	375-22-4	213	169	NA	13C4-PFBA
PFPeA	2706-90-3	263	219	69	13C5-PFPeA
PFHxA	307-24-4	313	269	119	13C5-PFHxA
PFHpA	375-85-9	363	319	169	13C4-PFHpA
PFOA	335-67-1	413	369	169	13C8-PFOA
PFNA	375-95-1	463	419	219	13C9-PFNA
PFDA	335-76-2	513	469	219	13C6-PFDA
PFUnA	2058-94-8	563	519	269	13C7-PFUnA
PFDaA	307-55-1	613	569	319	13C2-PFDaA
PFTTrDA	72629-94-8	663	619	169	13C2-PFDaA
PFTeDA	376-06-7	713	669	169	13C2-PFTeDA
PFBS	375-73-5	299	80	99	13C3-PFBS
PFPeS	2706-91-4	349	80	99	13C3-PFHxS
PFHxS	355-46-4	399	80	99	13C3-PFHxS
PFHpS	375-92-8	449	80	99	13C8-PFOS

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Abbreviation	CAS #	Ion Mass			Quantification Reference
		Parent	Quantification	Confirmation	
PFOS	1763-23-1	499	80	99	13C8-PFOS
PFNS	68259-12-1	549	80	99	13C8-PFOS
PFDS	335-77-3	599	80	99	13C8-PFOS
PFDoS	79780-39-5	699	80	99	13C8-PFOS
4:2 FTS	757124-72-4	327	307	81	13C2-4:2FTS
6:2 FTS	27619-97-2	427	407	81	13C2-6:2FTS
8:2 FTS	39108-34-4	527	507	81	13C2-8:2FTS
PFOSA	754-91-6	498	78	478	13C8-PFOSA
NMeFOSA	31506-32-8	512	219	169	D3-NMeFOSA
NEtFOSA	4151-50-2	526	219	169	D5-NEtFOSA
TFSI	82113-65-3	280	78	147	13C3-HFPODA
NMeFOSAA	2355-31-9	570	419	483	D3-NMeFOSAA
NEtFOSAA	2991-50-6	584	419	526	D5-NEtFOSAA
NMeFOSE	24448-09-7	616	59	NA	D7-NMeFOSE
NEtFOSE	1691-99-2	630	59	NA	D9-NEtFOSE
HFPO-DA	13252-13-6	285	169	185	13C3-HFPODA
ADONA	919005-14-4	377	251	85	13C4-PFH _p A
PFMPA	377-73-1	229	85	NA	13C4-PFBA
PFMBA	863090-89-5	279	85	NA	13C5-PFP _e A
NFDHA	151772-58-6	295	201	85	13C5-PFH _x A
9Cl-PF3ONS	756426-58-1	531	351	533→353	13C8-PFOS
11Cl-PF3OuDS	763051-92-9	631	451	633→453	13C8-PFOS
PFEESA	113507-82-7	315	135	83	13C5-PFH _x A
3:3FTCA	356-02-5	241	177	117	13C5-PFP _e A
5:3FTCA	914637-49-3	341	237	217	13C5-PFH _x A
7:3FTCA	812-70-4	441	317	337	13C9-PFNA
Extracted Internal Standards (EIS)					
13C3-PFPrA	NA	166	121	NA	13C3-PFBA
13C4-PFBA	NA	217	172	NA	13C3-PFBA
13C5-PFP _e A	NA	268	223	NA	13C2-PFH _x A
13C5-PFH _x A	NA	318	273	NA	13C2-PFH _x A
13C4-PFH _p A	NA	367	322	NA	13C2-PFH _x A
13C8-PFOA	NA	421	376	NA	13C4-PFOA
13C9-PFNA	NA	472	427	NA	13C5-PFNA

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Abbreviation	CAS #	Ion Mass			Quantification Reference
		Parent	Quantification	Confirmation	
13C6-PFDA	NA	519	474	NA	13C2-PFDA
13C7-PFUnA	NA	570	525	NA	13C2-PFDA
13C2-PFDoA	NA	615	570	NA	13C2-PFDA
13C2-PFTeDA	NA	715	670	NA	13C2-PFDA
13C3-PFBS	NA	302	80	NA	18O2-PFHxS
13C3-PFHxS	NA	402	80	NA	18O2-PFHxS
13C8-PFOS	NA	507	80	NA	13C4-PFOS
13C2-4:2FTS	NA	329	81	NA	18O2-PFHxS
13C2-6:2FTS	NA	429	81	NA	18O2-PFHxS
13C2-8:2FTS	NA	529	81	NA	18O2-PFHxS
13C8-PFOSA	NA	506	78	NA	13C4-PFOS
D3-NMeFOSA	NA	515	219	NA	13C4-PFOS
D5-NEtFOSA	NA	531	219	NA	13C4-PFOS
D3-NMeFOSAA	NA	573	419	NA	13C4-PFOS
D5-NEtFOSAA	NA	589	419	NA	13C4-PFOS
D7-NMeFOSE	NA	623	59	NA	13C4-PFOS
D9-NEtFOSE	NA	639	59	NA	13C4-PFOS
13C3-HFPO-DA	NA	287	169	NA	13C2-PFHxA
Non-Extracted Internal Standards (NIS)					
13C3-PFBA	NA	216	172	NA	NA
13C2-PFHxA	NA	315	270	NA	NA
13C4-PFOA	NA	417	372	NA	NA
13C5-PFNA	NA	468	423	NA	NA
13C2-PFDA	NA	515	470	NA	NA
18O2-PFHxS	NA	403	84	NA	NA
13C4-PFOS	NA	503	80	NA	NA

Table 7: QC Acceptance Criteria, Corrective Action and Data Qualification Requirements

QC Item	Acceptance Criteria	Corrective Action	Qualification
Mass Calibration	Must meet manufacturer's acceptance criteria.	Identify and correct source of problem, repeat Mass Calibration.	None. Do not proceed with analysis.
Mass Calibration Verification	Must meet manufacturer's acceptance criteria.	Identify and correct source of problem, repeat Mass Calibration.	None. Do not proceed with analysis.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

QC Item	Acceptance Criteria	Corrective Action	Qualification
Initial Calibration (ICAL)	Must meet linearity requirement specified in Section 9.3.1 for type of curve fit used.	Identify and correct source of problem, repeat.	None. Do not proceed with analysis.
Initial Calibration Verification (ICV)	All analytes must be within $\pm 30\%$ of their true values. (%R)	Identify and correct source of problem, re-analyze. If repeat failure, repeat ICAL.	If corrective action is not taken, qualify or disclose nonconformity for data user to assess impact on project samples.
Calibration Blank (ICB)	Non-DoD: No targets detected greater than MDL. DoD: No targets detected greater than $\frac{1}{2}$ LOQ	Identify and correct source of contamination or performance issue. Alternately, re-calibrate using a lower concentration standard for the highest point.	None. Do not proceed with analysis.
RT Window Position	Set position using the mid-point of the ICAL on the day ICAL is performed; otherwise, the opening CCV is used. RT Window is ± 60 secs from RT position.	NA	NA
Instrument Sensitivity Check (ISC)	All native and isotopically labelled compounds within $\pm 30\%$ recovery.	Identify and correct source of problem and reanalyze ISC. If problem persists, repeat ICAL.	None. Do not proceed with analysis.
Bile Salts Check	Bile Salt peak detected >1 min outside RT window for PFOS.	Identify and correct source of problem, then reevaluate Bile Salts separation from PFOS.	None. Do not proceed with analysis.
Continuing Calibration Verification (CCV)	All native and isotopically labelled compounds within $\pm 30\%$ recovery.	See Section 9.3.6 for required corrective actions based on circumstance.	Qualify test results associated with CCV failure.
Instrument Blank (IBLK) / Continuing Calibration Blanks (CCB)	Non-DoD: No targets detected $> MDL$. DoD: No targets detected $> \frac{1}{2}$ LOQ	Identify and correct source of contamination or performance issue. Reanalyze IBLK.	No samples shall be analyzed until opening IBLK has met acceptance criteria, unless the samples to be analyzed will not report the compound(s) detected in the IBLK.
Extracted Internal Standards (EIS)	See Table 9.	If batch QC is acceptable, reanalyze to confirm failure. If confirmed, re-extract from reduced sample amount and reanalyze sample, if possible. If failure persists, no further action is needed.	Qualify outages
Non-extracted Internal Standards (NIS)	NIS areas in all samples must be within 50 – 200% of the average area of the corresponding NIS in the ICAL.	Troubleshoot instrument performance. Reanalyze samples.	Qualify outages

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633 Effective Date: 10/14/2024

QC Item	Acceptance Criteria	Corrective Action	Qualification
Method Blank (MB)	Non-DOD: no targets detected greater than the LOQ or 1/10 of the concentration detected in associated sample(s), whichever is greatest. DoD: No targets detected > ½ LOQ	If IBLK is acceptable, reanalyze MB to confirm. If confirmed, re-extract and reanalyze associated impacted samples if sufficient amount remains and samples are within holding time; otherwise qualify associated test results.	Qualify outages
LCS/LLCS	%R: See Table 8	If the most recent ISC/CCV is acceptable, reanalyze LCS to confirm. If low-failure results are confirmed, re-extract and reanalyze associated samples (if sufficient sample remains). If insufficient sample remains for reprep, narrate and report data associated with low-failure LCS. If high-failure results are confirmed and sample(s) is ND for failing compound, narrate and report sample data.	Qualify outages
MS/MSD	%R: See Table 8 RPD ≤30%		Qualify outages
Laboratory Duplicate	RPD from parent sample ≤30%		Qualify outages.

Table 8: LCS / LLLCS Acceptance Limits (%R)

Analyte	CAS #	Aqueous/Leachate	Solid: All Matrices	Tissue
PFPrA	422-64-0	70-140	70-140	60-140
PFBA	375-22-4	70-140	70-140	60-140
PFPeA	2706-90-3	65-135	60-150	60-145
PFHxA	307-24-4	70-145	65-140	60-160
PFHpA	375-85-9	70-150	65-145	60-145
PFOA	335-67-1	70-150	70-150	60-150
PFNA	375-95-1	70-150	70-155	60-145
PFDA	335-76-2	70-140	70-155	60-150
PFUnA	2058-94-8	70-145	70-155	60-155
PFDoA	307-55-1	70-140	70-150	60-140

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Analyte	CAS #	Aqueous/Leachate	Solid: All Matrices	Tissue
PFTTrDA	72629-94-8	65-140	65-150	60-150
PFTTeDA	376-06-7	60-140	65-150	60-140
PFBS	375-73-5	60-145	65-145	60-150
PFPeS	2706-91-4	65-140	55-160	60-145
PFHxS	355-46-4	65-145	60-150	60-155
PFHpS	375-92-8	70-150	65-155	60-140
PFOS	1763-23-1	55-150	65-160	60-160
PFNS	68259-12-1	65-145	55-140	45-140
PFDS	335-77-3	60-145	40-155	50-140
PFDoS	79780-39-5	50-145	25-160	25-140
4:2 FTS	757124-72-4	70-145	60-150	55-150
6:2 FTS	27619-97-2	65-155	55-200	60-150
8:2 FTS	39108-34-4	60-150	70-150	60-170
PFOSA	754-91-6	70-145	70-140	60-150
NMeFOSA	31506-32-8	60-150	70-155	60-160
NEtFOSA	4151-50-2	65-145	70-140	60-150
TFSI	82113-65-3	70-140	70-140	60-140
NMeFOSAA	2355-31-9	50-140	65-155	60-145
NEtFOSAA	2991-50-6	70-145	65-165	60-145
NMeFOSE	24448-09-7	70-145	70-140	40-180
NEtFOSE	1691-99-2	70-135	70-135	60-205
HFPO-DA	13252-13-6	70-140	70-145	60-145
ADONA	919005-14-4	65-145	70-160	55-165
PFMPA	377-73-1	55-140	30-140	25-145
PFMBA	863090-89-5	60-150	60-150	60-150
NFDHA	151772-58-6	50-150	60-155	60-180
9Cl-PF3ONS	756426-58-1	70-155	70-150	60-170
11Cl-PF3OUdS	763051-92-9	55-160	45-160	50-170
PFEESA	113507-82-7	70-140	70-140	50-150
3:3 FTCA	356-02-5	65-130	45-130	30-140
5:3 FTCA	914637-49-3	70-135	60-130	60-160
7:3 FTCA	812-70-4	50-145	60-150	60-200

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl
	Substances by EPA 1633
Effective Date: 10/14/2024	

Table 9: EIS and NIS Acceptance Limits (%R)

Labeled Compound	Aqueous	Leachate	Solid:	Biosolid	Tissue
13C3-PFPrA	5-130	5-130	8-130	5-130	5-130
13C4-PFBA	5-130	5-130	8-130	5-130	5-130
13C5-PFPeA	40-130	40-130	35-130	35-130	10-185
13C5-PFHxA	40-130	40-130	40-130	40-130	25-170
13C4-PFHpA	40-130	40-130	40-130	40-130	25-150
13C8-PFOA	40-130	40-130	40-130	40-130	25-150
13C9-PFNA	40-130	40-130	40-130	40-145	35-185
13C6-PFDA	40-130	40-130	40-130	40-130	30-150
13C7-PFUnA	30-130	40-130	40-130	40-130	30-180
13C2-PFDoA	10-130	35-130	40-130	40-130	35-180
13C2-PFTeDA	10-130	25-130	20-130	10-160	20-160
13C3-PFBS	40-135	40-130	40-135	40-150	25-190
13C3-PFHxS	40-130	40-130	40-130	40-140	35-175
13C8-PFOS	40-130	40-130	40-130	40-130	40-160
13C2-4:2FTS	40-200	40-220	40-165	40-300	30-300
13C2-6:2FTS	40-200	40-170	40-215	40-300	35-300
13C2-8:2FTS	40-300	40-145	40-275	40-300	40-365
13C8-PFOSA	40-130	40-130	40-130	20-140	25-180
D3-NMeFOSA	10-130	40-130	10-130	20-130	5-130
D5-NEtFOSA	10-130	35-130	10-130	20-130	5-130
D3-NMeFOSAA	40-170	35-130	40-135	30-150	30-250
D5-NEtFOSAA	25-135	30-130	40-150	20-140	30-235
D7-NMeFOSE	10-130	20-130	20-130	25-130	5-160
D9-NEtFOSE	10-130	20-130	15-130	20-130	5-130
13C3-HFPODA	40-130	40-130	40-130	40-130	20-185
13C3-PFBA	50-200	50-200	50-200	50-200	50-200
13C2-PFHxA	50-200	50-200	50-200	50-200	50-200
13C4-PFOA	50-200	50-200	50-200	50-200	50-200
13C5-PFNA	50-200	50-200	50-200	50-200	50-200
13C2-PFDA	50-200	50-200	50-200	50-200	50-200
18O2-PFHxS	50-200	50-200	50-200	50-200	50-200
13C4-PFOS	50-200	50-200	50-200	50-200	50-200

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl
	Substances by EPA 1633
Effective Date: 10/14/2024	

Table 10: IPR Limits for Method Verification

Analyte	CAS #	Aqueous/Leachate		Solid: All Matrices		Tissue	
		%R	RPD	%R	RPD	%R	RPD
PFPPrA	422-64-0	70-140	20	70-140	20	70-140	20
PFBA	375-22-4	70-135	21	70-140	17	70-135	20
PFPeA	2706-90-3	70-135	23	70-140	26	70-130	20
PFHxA	307-24-4	70-135	24	70-135	23	70-140	20
PFHpA	375-85-9	70-135	28	70-140	21	70-140	20
PFOA	335-67-1	65-155	27	70-140	23	70-130	25
PFNA	375-95-1	70-140	28	65-145	24	70-140	20
PFDA	335-76-2	65-140	26	70-145	26	70-135	20
PFUnA	2058-94-8	70-135	29	70-145	26	70-135	20
PFDaA	307-55-1	70-130	21	70-145	25	70-135	20
PFTTrDA	72629-94-8	60-145	29	55-160	26	55-160	20
PFTeDA	376-06-7	70-145	27	70-145	24	70-140	25
PFBS	375-73-5	70-140	23	60-145	25	70-145	20
PFPeS	2706-91-4	70-135	25	65-140	29	70-150	20
PFHxS	355-46-4	70-135	27	65-145	28	70-140	25
PFHpS	375-92-8	70-140	30	70-140	27	70-145	20
PFOS	1763-23-1	70-140	29	70-135	27	70-135	30
PFNS	68259-12-1	70-135	29	70-140	27	60-130	20
PFDS	335-77-3	70-135	30	50-150	31	55-135	20
PFDoS	79780-39-5	45-135	35	40-140	40	25-145	45
4:2 FTS	757124-72-4	70-135	27	70-135	27	65-140	30
6:2 FTS	27619-97-2	70-135	32	60-160	50	70-140	25
8:2 FTS	39108-34-4	70-140	33	70-140	27	70-150	25
PFOSA	754-91-6	70-135	22	70-140	19	70-140	20
NMeFOSA	31506-32-8	70-135	30	65-145	26	70-140	30
NEtFOSA	4151-50-2	70-130	26	70-135	19	70-140	40
TFSI	82113-65-3	70-140	20	70-140	20	70-140	20
NMeFOSAA	2355-31-9	65-140	32	65-145	31	70-140	20
NEtFOSAA	2991-50-6	70-135	28	60-150	31	70-140	25
NMeFOSE	24448-09-7	70-135	29	70-140	19	60-150	20
NEtFOSE	1691-99-2	70-130	21	70-135	17	70-145	40
HFPO-DA	13252-13-6	70-135	23	70-140	25	70-140	25
ADONA	919005-14-4	70-135	23	70-155	26	70-145	20

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Analyte	CAS #	Aqueous/Leachate		Solid: All Matrices		Tissue	
		%R	RPD	%R	RPD	%R	RPD
PFMPA	377-73-1	60-140	23	70-140	25	70-145	40
PFMBA	863090-89-5	65-145	27	55-145	33	70-140	20
NFDHA	151772-58-6	65-140	37	45-145	27	70-155	30
9CI-PF3ONS	756426-58-1	70-145	30	65-135	23	70-140	20
11CI-PF3OUdS	763051-92-9	50-150	35	50-135	31	65-140	20
PFEESA	113507-82-7	70-135	25	70-140	20	70-145	25
3:3 FTCA	356-02-5	70-130	23	45-155	32	55-130	25
5:3 FTCA	914637-49-3	70-130	24	70-135	28	70-145	20
7:3 FTCA	812-70-4	55-130	34	70-145	39	70-155	20

END OF APPENDIX

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Appendix B: Reference Materials – Reagents, Standards & Formulations

Table 1: Purchased Reagents

Name	Grade	Vendor	Catalog	Storage
Acetic Acid	ACS	Fisher	A38500	Room Temperature
Acetonitrile ¹	UPLC	Fisher	A996-4	Room Temperature
Ammonium Acetate	LC/MS	Fisher	A639-500	Room Temperature
Aqueous Ammonium Acetate	5M	Fisher	AM9071	0-6°C (refrigerated)
Ammonium Hydroxide	Certified ACS+	Fisher	A669-500	Room Temperature
Formic Acid	>96% purity	Fisher	147932500 t	Room Temperature
Methanol	HPLC, 99.9% purity	Fisher	A454-4	Room Temperature
Potassium Hydroxide	Certified ACS	Fisher	P250-500 o	Room Temperature
Liquid Nitrogen	>95% Purity	Linde	NILC-230-22	Room Temperature

¹ Verify Acetonitrile, Methanol, and Laboratory Generated Regent Water by lot/batch before use.

Table 2: Purchased Standard Solutions¹

Name	Vendor	Catalog	Purpose
EPA 1633 Native PFAS Mix	Wellington Laboratories	EPA-1633STK	Primary Source Standard
PFPrA Native Stock (50 µg/mL)	Cambridge Isotope Laboratories	ULM-11323-A-S	Primary Source Standard
TFSI Native Stock (100 µg/mL)	AccuStandard	PFOS-030S	Primary Source Standard
WatR™ Pollution PFAS in Wastewater ²	ERA	404	Second Source Standard
Mass-labelled PFAS Extraction Standard Mix	Wellington Laboratories	MPFAC-HIF-ES	EIS
13C3-PFPrA Stock (50 µg/mL)	Cambridge Isotope Laboratories	CLM-11324-A-S	EIS
Mass-labelled PFAS Injection Standard Mix	Wellington Laboratories	MPFAC-HIF-IS	NIS
Neat Taurodeoxycholic Acid (TDCA)	Cayman Chemical	15935	Bile Salt Interference

¹ Unless other storage conditions are specified by the manufacturer, store the purchased standard solutions in the dark at 0-6°C, in tightly sealed screw-capped vials. Place a mark on the vial at the level of the solution after each use so that solvent loss by evaporation can be detected. Replace the solution if solvent loss has occurred. Properly discard or remove from the work area unused material on the expiration date to prevent inadvertent use of expired material.

² ERA item # 404 is not prepared with identical concentrations in each lot; therefore, the ICV formulation will need to be reviewed and potentially adjusted with each new lot of this mix. The formulation below was designed based on ERA item# 404, lot# P336-404.

Purchased standard solutions must be received with a Certificate of Analysis (COA), which must be retained on-site in the laboratory and be traceable to each corresponding lot received. Refer to laboratory SOP ENV-SOP-MIN4-0180 *Laboratory Procurement Procedures*, or however named, for the procedures for standard receipt and traceability.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Substitution of reagents and standards from those listed in these tables is permissible only if the replacement meets the specifications outlined in the SOP. If an alternative item does not meet these exact same specifications for purity, composition, or concentration, any formulation in this SOP must be modified to match the material used. If the substitution is permanent, the SOP and formulations in this SOP must be updated.

Prepared Reagents & Standard Solution Formulations:

Plastic containers must be used for all standard, sample, and extraction preparations due to potential adsorption of analytes onto glass. Do not substitute glass for plastic containers.

18O-mass labeled perfluoroalkyl sulfonates may undergo isotopic exchange with water under certain conditions, which lowers the isotopic purity of the standards over time - monitor standards for degradation and replace as needed.

Modifications to these formulations are not permitted unless the formulation of the parent component changes, in which case the formulation must be adjusted accordingly.

Table 3: Laboratory Prepared Reagent Formulation Table

Name	Component	Amount	Volume	Expires
Mobile Phase A: 2mM ammonium acetate in 95:5 water/acetonitrile	Ammonium Acetate	0.154 g	1 L	60 Days
	Acetonitrile	50 mL		
	Reagent Water	950 mL		
Instrument Blank Mix (IBLK Mix): Methanol with 4% Water, 1% Ammonium Hydroxide, and 0.625% Acetic Acid	Methanol	94.4 mL	100 mL	30 Days
	Ammonium Hydroxide	3.3 mL		
	Acetic Acid	0.625 mL		
	Reagent Water	1.67 mL		
Acetic Acid (0.1%)	Acetic Acid	1 mL	1 L	90 Days
	Reagent Water	999 mL		
Ammonium Hydroxide (3%): Aqueous	Ammonium Hydroxide	10 mL	100 mL	90 Days
	Reagent Water	90 mL		
Ammonium Hydroxide (0.3%): Methanolic	Ammonium Hydroxide	1 mL	100 mL	30 Days
	Methanol	99 mL		
Ammonium Hydroxide (1%): Methanolic	Ammonium Hydroxide (30%)	3.3 mL	100 mL	30 Days
	Methanol	96.7 mL		
Ammonium Hydroxide (2%): Methanolic	Ammonium Hydroxide (30%)	6.6 mL	100 mL	30 Days
	Methanol	93.4 mL		
Formic Acid (0.1 M): Aqueous	Formic Acid	3.77 mL	1 L	2 Years

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Name	Component	Amount	Volume	Expires
	Regent Water	996.33 mL		
Formic Acid (0.3 M): Aqueous	Formic Acid	11.31 mL	1 L	2 Years
	Regent Water	988.69 mL		
Formic Acid (5% v/v): Aqueous	Formic Acid	5 mL	100 mL	2 Years
	Regent Water	95 mL		
Formic Acid (50% v/v): Aqueous	Formic Acid	50 mL	100 mL	2 Years
	Regent Water	50 mL		
1:1 Methanol:0.1M Formic acid	Methanol	50 mL	100 mL	2 Years
	Formic Acid (0.1M)	50 mL		
Potassium Hydroxide (0.05M): Methanolic	Potassium Hydroxide	3.3 g	1 L	90 Days
	Methanol	1 L		
Reagent Water (ASTM 1)	Laboratory Generated			2 Years

Table 4: Laboratory Prepared Intermediate Standard Solutions

Name	Component	Amount	Volume	Storage	Expires
TDCA Stock (2000 µg/mL)	Neat TDCA	10 mg	5 mL	<0°C	1 Year
	Methanol	5 mL			
1633PDS (Concentrations vary)	EPA-1633STK	5000 µL	10 mL	0-6°C	30 Days or with each new lot of parent standard
	PFPPrA stock	100 µL			
	TFSI stock	25 µL			
	Methanol	4875 µL			
PDS-10X (Concentrations vary)	1633PDS	300 µL	3 mL	0-6°C	30 Days or with each new lot of parent standard
	Methanol	2700 µL			
EIS Mix (Concentrations vary)	MPFAC-HIF-ES	5000 µL	10 mL	0-6°C	30 Days
	13C3-PFPPrA stock	200 µL			
	Methanol	4800 µL			

Table 5: Laboratory Prepared Working Standard Solutions

Name	Component	Amount	Volume	Storage	Expires
ICV	ERA-404 Mix	200 µL	4 mL	0-6°C	30 Days
	EIS Mix	40 µL			
	MPFAC-HIF-IS	20 µL			

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl
	Substances by EPA 1633
Effective Date: 10/14/2024	

Name	Component	Amount	Volume	Storage	Expires
	IBLK Mix	3740 µL			
CAL L1	PDS-10X	16 µL	2 mL	0-6°C	30 Days
	EIS Mix	20 µL			
	MPFAC-HIF-IS	10 µL			
	IBLK Mix	1954 µL			
CAL L2	PDS-10X	80 µL	5 mL	0-6°C	30 Days
	EIS Mix	50 µL			
	MPFAC-HIF-IS	25 µL			
	TDCA Stock (2000 µg/mL)	25 µL			
	IBLK Mix	4820 µL			
CAL L3	PDS-10X	80 µL	2 mL	0-6°C	30 Days
	EIS Mix	20 µL			
	MPFAC-HIF-IS	10 µL			
	IBLK Mix	1890 µL			
CAL L4	1633PDS	40 µL	2 mL	0-6°C	30 Days
	EIS Mix	20 µL			
	MPFAC-HIF-IS	10 µL			
	IBLK Mix	1930 µL			
CAL L5	1633PDS	200 µL	5 mL	0-6°C	30 Days
	EIS Mix	50 µL			
	MPFAC-HIF-IS	25 µL			
	TDCA Stock (2000 µg/mL)	25 µL			
	IBLK Mix	4700 µL			
CAL L6	1633PDS	200 µL	2 mL	0-6°C	30 Days
	EIS Mix	20 µL			
	MPFAC-HIF-IS	10 µL			
	IBLK Mix	1770 µL			
CAL L7	1633PDS	400 µL	2 mL	0-6°C	30 Days
	EIS Mix	20 µL			
	MPFAC-HIF-IS	10 µL			
	IBLK Mix	1570 µL			
CAL L8	1633PDS	800 µL	2 mL	0-6°C	30 Days
	EIS Mix	20 µL			
	MPFAC-HIF-IS	10 µL			
	IBLK Mix	1170 µL			

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Name	Component	Amount	Volume	Storage	Expires
CAL L9	1633PDS	1000 µL	2 mL	0-6°C	30 Days
	EIS Mix	20 µL			
	MPFAC-HIF-IS	10 µL			
	TDCA Stock (2000 µg/mL)	10 µL			
	IBLK Mix	960 µL			

END OF APPENDIX

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633 Effective Date: 10/14/2024

Appendix C: Sample Screening Protocol

Samples that are known or suspected to contain high concentrations of target analytes may be pre-screened to determine if a reduced volume or mass should be used for extraction or if an extract dilution should be performed prior to initial analysis. Other pre-screening procedures may be used.

This screen protocol may be used for all matrices included in this SOP, except surface wipes because there is insufficient representative sample to perform the screen procedure.

Aqueous Matrices (Non-Potable Water, Landfill Leachate, TCLP/SPLP Leachate)

Always take the aliquot for screening from the 125 mL container; do not take a subsample from the sample container to be used for sample preparation or analysis (except in the case of TCLP/SPLP).

- 1) Mix the sample to ensure it is homogenous. Measure 2 mL of sample into a 15 mL centrifuge tube.
- 2) Add 2 mL of methanol, 40 µL of EIS Mix and 20 µL MPFAC-HIF-IS to the sample and vortex to mix.
- 3) Filter 1 mL of the sample through 0.2-µm membrane filter into a microvial.
- 4) Analyze the screen extract and use the test result to determine if a reduced volume should be extracted or if the extract from the routine procedure should be diluted prior to analysis.

Soil/Sediment, Biosolid, Tissue, and Articles of Commerce

- 1) Measure 1.0 g (± 0.1) of sample into a 50 mL polypropylene centrifuge tube.
- 2) Add 20 mL of 0.3% methanolic ammonium hydroxide to the sample. Vortex to mix for 10 min. Let the sample sit to settle and/or centrifuge to produce a clear extract.
- 3) Filter using a 0.2 µm filter vial:
 - (i) Add 40 µL of EIS mix to a clean filter vial (chamber).
 - (ii) Add 400 µL of clear extract from step 2 to the vial. Carefully vortex to mix.
 - (iii) Use filter/plunger to filter the extract.
- 4) Transfer 30 µL of extract to a ~300 µL polypropylene microvial and dilute to 300 µL with 0.3% methanolic ammonium hydroxide. Add 1.5 µL of MPFAC-HIF-IS to the resulting solution.
- 5) Analyze the screen extract and use the test result to determine if a reduced volume should be extracted or if the extract from the routine procedure should be diluted prior to analysis.

END OF APPENDIX

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633 Effective Date: 10/14/2024

Appendix D: Subsampling Procedure for Aqueous Matrices

Due to the propensity of some target analytes to stratify within the sample matrix or to adhere to walls of the sample container, subsampling of aqueous samples may only be done on a project-specific basis, with client approval.

In this context, subsampling refers to the taking of an aliquot of sample from the container provided to the laboratory for extraction in lieu of using the entire sample volume collected. This is sometimes necessary when high concentrations of target analytes are expected in the sample or for other project-specific reasons.

Subsampling Procedure for Non-Potable Water (NPW), Landfill Leachate, TCLP/SPLP

- 1) Determine the volume (mL) for extraction based on screen results or project specifications. The amount subsampled must be at least 250 µL. If additional dilutions are needed, these must be made by serial dilution of the sample.
- 2) Place an uncapped, HDPE bottle onto the weigh pan of a top loading balance and tare the balance. Select an appropriately sized bottle for the volume of sample to be aliquoted. Regardless of volume subsampled, the sample aliquot should be diluted to at least 50 mL prior to SPE.
- 3) Gently invert the sample to mix 3-4 times then transfer the sample into the HDPE bottle until the target volume is achieved, assuming sample density of 1 g/mL.
 - If foam forms during inversion and more than 5 mL is required, pour the sample to transfer, but avoid the transfer of foam.
 - If foam forms and less than 5 mL is needed, use a pipette to draw sample from ½ cm below the foam.
 - If no foam forms, the transfer may be done by pouring or by pipette.
- 4) Fill the bottle with reagent water.
- 5) Proceed to Section 9.6 and prepare the sample using the same stepwise procedure used for aqueous samples.

Subsampling Procedure for AFFF and F3 and Other Challenging Aqueous Matrices

The following protocol is designed for “as prepared” AFF materials and newer generation F3 concentrates and other challenging matrices, such as viscous or cloudy matrices, or those with standing foam. Consult with technical specialist or supervisor to determine when to use this protocol.

If concentrated AFF (thick gel-like matrix) was received, notify the supervisor to determine where to perform the subsampling procedure. Subsampling of the concentrate must be done in an isolated area to prevent the material from contaminating the work area and other samples.

- 1) Prepare 2 samples for all matrices designated AFFF or F3.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

- 2) Place an uncapped 125 mL HDPE bottle onto the weigh pan of a top loading balance and tare the balance.
- 3) Measure 0.02 g of sample into the HDPE bottle. Record the actual mass subsampled.
- 4) Add approximately 60 mL of reagent water to the bottle, cap the bottle.
- 5) Shake by hand vigorously for 20-30 seconds.
- 6) Set the sample aside for at least 3 hours for complete dissolution.
- 7) Check the sample after 3 hours to verify dissolution is complete. If full dissolution has not occurred, place the sample bottle on an orbital shaker for 15 minutes, then allow the sample to settle for 3 hours.
- 8) If the sample is an AFFF concentrate gel, go to step 9; otherwise, proceed to Section 9.6 and prepare the sample using the same stepwise procedure used for aqueous samples.
- 9) Dilute the AFFF concentrate subsample solution by quantitatively measuring 6 mL of the 60 mL solution into a new 125 mL HDPE container and dilute to 60 mL with reagent water. The serial dilution, when the above protocol is followed without deviation, represents a 250,000X primary dilution. Proceed to Section 9.6 and extract the prepared sample using the same stepwise procedure used for aqueous samples.

END OF APPENDIX

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Appendix E: Aqueous Sample Preparation – Settled Solids Depth > ½ cm

As allowed by EPA Method 1633 PAS uses alternate strategies in lieu of the total suspended solids (TSS) determination to mitigate solid materials or other interferences present in aqueous matrices.

Use this procedure in lieu of Section 9.6.1 (steps 1-9) when settled solids in the sample appear to be of a depth > ½ cm and less than 5% of the total volume.

- 1) Label a 500 mL conical centrifuge bottle with the sample ID. Set the labeled tube in a rack and remove the cap.
- 2) Spike each sample with 40 µL of EIS Mix in its original sample container.
- 3) If the sample is a MS or MSD, add 80 µL of 1633PDS to each.

NOTE: The MS/MSD or laboratory duplicate must be processed the same way as the parent sample and vice versa.

- 4) Cap the container and vigorously shake to mix.
- 5) Pour the sample into the labeled centrifuge bottle, taking care to transfer all the solids present in the original sample bottle to the centrifuge bottle. Tightly cap the centrifuge bottle when done.
- 6) Place the tubes in the centrifuge rotor, ensuring that the carousel is symmetrically balanced. Close the top and centrifuge at 3000 RPM (~2000 RCF) for 6 minutes. Record the start and end time in the preparation log.
- 7) Decant the centrifuged sample back into its original container, taking care to avoid transfer of the settled solids.
- 8) Weigh the original sample container + decanted sample and record the weight measurement in the preparation log.
- 9) Extract the decanted sample using the procedure in Section 9.6.1, starting with step 10. If the SPE cartridge begins to clog during sample transfer, refer to Appendix G for troubleshooting steps and mitigation strategies.
- 10) When the SPE cartridges have been dried and the carbon cartridges added to the SPE stack, rinse the original sample bottle following the protocol in Step 10 of Section 9.7, but do not start elution.
- 11) Add 5mL of 1% methanolic ammonium hydroxide to each centrifuge bottle and swirl to rinse. If the condensed solids become re-suspended during this step, centrifuge the container to separate.
- 12) Using a pipet, transfer the rinsates from the original sample container and the centrifuge bottle to the associated sample's cartridge/reservoir stack. Elute the cartridge with both rinsate volumes; allow the elution solvent to soak the SPE sorbent for 2 minutes, then use vacuum to pull the elution solvent through the cartridge and into the collection tubes in a slow, dropwise manner.

If using sample transfer lines, add the rinsate from the centrifuge bottle to the original sample bottle for transfer to the cartridge and then elute as above.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

- 13) Add 5 mL of 1% methanolic ammonium hydroxide directly to the SPE reservoir of all batch QC samples (MB/LCS/LLCS) and elute with the normal 5 mL bottle rinse described in Step 10 of Section 9.7.
- 14) Concentrate the extract to ~3.5 mL using nitrogen blowdown at an approximate flow rate of 1.2 L/min. Adjust to final volume to 4 mL with 1% methanolic ammonium hydroxide using a validated comparison vial.
- 15) Weigh the empty sample bottle with cap and record the weight measurement. Subtract this weight from the initial container+sample weight from Step 8 to determine initial sample volume.
- 16) Add 25 µL of concentrated acetic acid and 20 µL of NIS mix to each extract and vortex to mix. Transfer a portion of each extract into an autosampler vial for LC-MS/MS analysis. Cap the collection tubes containing the remaining extracts and store at 0 - 6 °C until analysis.
- 17) Document in the preparation log which samples in the prep batch were centrifuged prior to SPE; these samples must be identified in the test report to alert the customer and data user of the deviation from routine protocol.
- 18) Record any other observations or anomalies observed during the centrifugation or extraction process, particularly if any issues were encountered during the sample loading step of the SPE process.

END OF APPENDIX

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Appendix F: Biphasic Sample Separation Procedure

Use this procedure when the customer has requested separation of a biphasic sample into aqueous and solid phases.

- 1) Verify the log-in has been updated to specify sample preparation for each matrix. Be sure the log-in for the solid fraction does not include percent solids determination: the solid phase of a biphasic sample is reported on a wet weight basis.
- 2) Label a 500 mL conical centrifuge bottle with the sample ID. Set the labeled bottle in a rack and remove the cap.
- 3) Vigorously shake the sample in its original container then completely transfer the entire sample to the corresponding centrifuge bottle and cap the centrifuge bottle.
- 4) Place the centrifuge bottle(s) in the centrifuge rotor, ensuring that the carousel is symmetrically balanced. Close the top and centrifuge at 3000 RPM (~2000 RCF) for 6 minutes. Record the start and end time in the preparation log.
- 5) Transfer the liquid portion of sample back into the original sample container; avoid transferring solid material as much as possible.
- 6) Tare a top loading balance then weigh the original container + liquid sample and record this measurement to the nearest 0.1 g on the preparation log as initial weight for the aqueous phase.
- 7) Proceed to Section 9.6.1 to add the sample to a routine NPW preparation batch and complete the preparation process starting with Step 7.
- 8) Label a 50 mL centrifuge tube with the sample ID.
- 9) Place the tube on a top loading balance and tare the balance.
- 10) Using a wooden tongue depressor, transfer the solid material from the 500 mL centrifuge bottle to the 50 mL container.
- 11) Record the amount transferred to the nearest 0.1 g on the preparation log as initial weight, targeting 5 g for preparation.

END OF APPENDIX

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633 Effective Date: 10/14/2024

Appendix G: SPE Clog Mitigation Procedure

This appendix provides instructions on what to do when an SPE cartridge has become clogged.

Indications that an SPE cartridge may be or will become clogged include discoloration of the cartridge frit or absorbent bed, coloration or sedimentation on the bottom of the reservoir, and a slowing of the sample loading rate.

When these conditions are observed do not add any more sample volume to the reservoir of the clogging cartridge. The goal is to stop the process before the cartridge seizes up completely, so that a sample extract can still be collected from the cartridge.

- If there is >10 mL remaining in the reservoir, close the manifold port to stop sample loading. After all other samples have finished loading, carefully remove the cartridge-adaptor-reservoir assembly and transfer the remaining sample volume back into the original sample container.
- If there is <10 mL remaining, allow the sample to continue to load. If the sample has not finished after all other samples have passed through their cartridge, close all other ports except for the sample loading, then apply more vacuum pressure to pull the sample through. Do this step one at a time for each clogged SPE. If it is obvious that the cartridge has seized, do not attempt this step. Instead, remove the cartridge and return the remaining sample volume to its original container.

The extraction process may continue as follows; if QA acceptance criteria are not met for the resulting extract, the sample should be re-extracted with reduced volume.

- Replace the cartridge from the abandoned sample and finish the SPE elution process (Step 5 from Section 9.7) to collect what extract is available from the SPE. If the SPE clogs completely, abandon the elution process; the sample must be diluted and the extraction repeated.
- To obtain the volume of sample used for preparation and extraction, reweigh the empty container and subtract this weight from the original container+sample weight measurement.
- Record in the preparation log that SPE extraction could not be completed with full sample volume.

Alternatively,

- Option 1: Reprepare and re-extract the sample following the centrifuge procedure in Appendix E. This option may be appropriate when centrifugation was not originally performed on the sample that clogged.
- Option 2: Reprepare the sample using a reduced volume. No less than 1 mL of homogenized sample may be used for extraction in these cases.
- Option 3: With client approval, re-log the sample as a biosolid matrix and prepare following the biosolid matrix extraction protocols, targeting 0.5 g of sample. In this case, the sample results will generally not be corrected for dry weight.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

- Option 4: With client approval, re-log and reprepare the sample as two separate phases (aqueous and solid). Attempt phase separation of the aqueous and solid phases of the sample following Appendix F and, if successful, prepare as two separate samples.
- Option 5: Test procedure is not applicable. This may be the only option when the matrix interference and/or the modification to the procedure become so extreme that the resulting test result is unreliable.

In all cases, the protocol followed must be recorded, disclosed to the customer, and test results adjusted for actual amounts used.

END OF APPENDIX

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

Appendix H: Particle Size Reduction by Cryomill for Articles of Commerce

Clients may submit solid samples of various consumer and related product materials for analysis of PFAS. To effectively mobilize any PFAS that may be present within the structure of the material, samples must be milled to a fine powder using a cryogenic mill (Cryomill). Use this procedure to mill articles of commerce to a fine powder prior to sample preparation, when needed.

Examples of common articles requiring milling prior to extraction and analysis are molded articles, coated wire, textiles, paper, leather, food, and LDPE and polypropylene.

Sample Processing Procedure

- 1) Remove the sample from storage.
- 2) If possible, use scissors or heavy plate shears to roughly cut the material into smaller pieces, preferably to a size ~10 X 10 mm. The optimum size depends on the equipment used for milling.
- 3) Use a Retsch Cryomill (ball mill) with automatic liquid nitrogen feed to mill the material to ~ 100 um in diameter using the specifications in the table below:

Sample Type ¹	Cycles	Preset	Frequency (Hz)	PCT ² (minutes)	MT ³ (minutes)	ICT ⁴ (minutes)
Elastomers (FKM, FFKM, FEPM, and agglomerates)	1	1	30	Auto	2	NA
Compacted Cake	5	NA	30	Auto	2	2
Melt Extruded Pellets and Molded Articles	7	2	30	Auto	4	2
Coated Wire	3	3	30	Auto	2	2
Textiles, Paper, Leather	3	4	25	10	2.5	0
Food (meat, cheese)	1	5	25	8	2	NA
LDPE and PP	2	6	25	10	2	2

¹ Use Feed Mass of 5g for most applications

² PCT = Processing Time

³ MT: Milling Time

⁴ ICT: In-Between Cooling Time

- 4) Make sure to follow the User Manual for installing and removing the grinding jar. This includes the two lock rings on the left side of the mechanism.
 - (i) Make sure outer LOCK RINGS are loosened.
 - (ii) Screw in the grinding jar (from the right side of the mechanism). Tighten with black plastic tightening aid.

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.

Test Method Standard Operating Procedure (SOP): Pace® Analytical Services

	ENV-SOP-MIN4-0205 v00_Determination of Per- and Polyfluoroalkyl Substances by EPA 1633
	Effective Date: 10/14/2024

- (iii) Tighten two LOCK RINGS: Outside ring tightened first (hand-tighten only); inner ring tightened second (snug), using the tool.
 - (iv) Perform grinding cycle.
 - (v) Loosen those two LOCK RINGS: Inside ring loosened first, outer ring loosened second. To loosen either ring, the tool will be needed because of icing.
 - (vi) Unscrew grinding jar.
 - (vii) When cryomill is not in use, make sure to replace the cooling jacket seal.
- 5) When the milling process is complete, sweep the milled material into a labeled 50 mL centrifuge tube.
 - 6) Immediately proceed to Section 9.6.2 for sample preparation or freeze the sample at a temperature of $\leq -10^{\circ}\text{C}$ until preparation.
 - 7) Clean the mill between each sample to avoid cross-contamination (see procedure below).

Mill Cleaning Procedure

- 1) Remove any residual material with DI water and a brush. If needed, also use a clean polish cloth or cotton ball/swab.
- 2) Rinse all parts with methanol followed by DI, using an ultrasonic bath as needed with each rinse.
- 3) Repeat Steps 1-2 until clean.

END OF APPENDIX

The distribution of printed or electronic copies of this document outside of the PAS electronic document management system (eDMS) is not controlled or tracked. PAS employees must ensure any copy used for daily work, if not accessed directly from PAS eDMS, is the current version of the document.