Revised Errata Sheet for Draft Method 1633 February 8, 2022

On September 2, 2021, EPA's Office of Water published a draft of EPA Method 1633 for per- and polyfluoroalkyl substances (PFAS) in eight different environmental media, including wastewater, surface water, groundwater, and soils. The draft method has been validated in a single laboratory and is the subject of a multi-laboratory method validation study being conducted by the Department of Defense. The published method is clearly identified as a draft procedure that is subject to revision. As noted at the front of the method itself, this method **is not required** for Clean Water Act (CWA) compliance monitoring until it has been proposed and promulgated through rulemaking.

Since the publication of the draft procedure, EPA has received comments from various sources, some of which have pointed out minor errors or confusing statements in the draft method document. Given the draft status of the procedure and the upcoming validation study, EPA issued an errata sheet on October 15, 2021 to address the errors identified at that time, rather than reissuing a new draft document. Since then, additional questions have been posed to EPA and therefore, EPA has revised and updated the errata sheet to reflect the new questions and concerns. The changes described in this errata sheet will be incorporated in the next revision of the method that will be released after completion of the multi-laboratory method validation study.

The errata for this draft procedure are summarized in the table below that specifies:

1. Where in the document the change occurs
2. How the current text reads
3. How the corrected text will read, with changes shown in red font
4. The reason for the correction

Rows shaded in green indicate changes made since the October 15, 2021 version was posted.

As with the draft method itself, questions or comments about this errata sheet should be sent using the CWA Analytical Methods website's "CONTACT US" page:

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

Comments can also be addressed to:

CWA Methods Team, Engineering and Analysis Division (4303T)
Attn: Adrian Hanley
Office of Science and Technology
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1200 Pennsylvania Avenue
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Errata for Draft Method 1633, Analysis of Per- and Polyfluoroalkyl Substances (PFAS) in Aqueous, Solid, Biosolids, and Tissue Samples by LC-MS/MS, EPA 821-R-21-001, August 2021			
Text Location	**Current Text**	**Corrected Text**	**Reason for Correction**
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%).	All parts of the SPE manifold must be cleaned between samples with methanolic ammonium hydroxide (1%) and air dried prior to use. Sonication with methanolic ammonium hydroxide (1%) may be used for components that will fit in an ultrasonic bath. Smaller parts, like the needles, adapters, reservoirs, and stopcocks associated with the manifold require rinsing with tap water prior to manual cleaning or sonicating with 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%).	Some SPE manifold components may be too large to fit in commonly used ultrasonic baths. Where size is not a concern, sonication is encouraged as an effective approach to cleaning these parts.
6.3.4	Tissue homogenizer – Pro Scientific PRO400DS homogenizer or equivalent with stainless steel macro-shaft and turbo-shear blade	This text will be deleted	The description of the blade and shaft of Pro Scientific device is incorrect, and the PRO400DS homogenizer has a PTFE bearing that makes it inappropriate for use in a PFAS method. Use of the device itself is never called out in the procedure after this section.
6.9.3	Clear snap cap, PVDC film/white silicone, 11 mm (American Chromatography Supplies Cat # C299-11 or equivalent)	Clear snap cap, polyethylene, 11 mm (Fisher Scientific # 03-375-24E, or equivalent)	Since the initial release of the draft method, the supplier has discontinued the snap cap that was cited. The replacement shown here is currently available and does not contain PTFE.
7.5	Taurodeoxycholic Acid (TDCA) or Sodium taurodeoxychloate 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.	Bile salt interference check standard containing Taurodeoxycholic Acid (TDCA) or Sodium taurodeoxychloate hydrate – (Sigma Aldrich 580221-5GM, or equivalent). This standard is used to evaluate the chromatographic program relative to the risk of an interference from bile salts in tissue samples when using acetonitrile as the mobile phase in the instrument. Prepare solution at a concentration of 1 µg/mL in the same solvent as the calibration standards. If using other mobile phases and analyzing tissues, it will be necessary to evaluate taurochenodeoxycholic acid (TCDCA) and tauroursodeoxycholic acid (TUDCA) as well.	Naming this solution the bile salt interference check standard simplifies later discussions of its use. The potential interference with PFOS from bile salts is affected by the mobile phase used for the LC separation and laboratories choosing to use a mobile phase other than that specified in the draft procedure need to make the adjustments described here. The concentration of the solution has also been lowered so it does not overwhelm the peak for PFOS.

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Text Location	**Current Text**	**Corrected Text**	**Reason for Correction**
8.2.3	Maintain all aqueous samples protected from light at 0 - 6 ºC 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 ºC during transport and be received by the laboratory within 48 hours of collection. The laboratory must confirm that the sample temperature is 0 - 6 ºC upon receipt. Once received by the laboratory, the samples must be stored at ≤ -20 ºC until sample preparation.	Maintain aqueous samples protected from light at 0 - 6 ºC 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 ºC during transport and be received by the laboratory within 48 hours of collection. The laboratory must confirm that the sample temperature is 0 - 6 ºC upon receipt. Once received by the laboratory, the samples may be stored at ≤ -20 ºC, or at 0 - 6 ºC, until sample preparation. However, the allowable holding time for samples depends on the storage temperature, as described in Section 8.5.	The change addresses the discrepancy between Sections 8.2 and 8.5
8.3.2	Maintain solid samples protected from light (in HDPE containers) at 0 - 6 ºC from the time of collection until receipt at the laboratory. The laboratory must confirm that the sample temperature is 0 - 6 ºC upon receipt. Once received by the laboratory, the samples must be stored at ≤ -20 ºC until sample preparation.	Maintain solid samples protected from light (in HDPE containers) at 0 - 6 ºC from the time of collection until receipt at the laboratory. The laboratory must confirm that the sample temperature is 0 - 6 ºC upon receipt. Once received by the laboratory, the samples may be stored at ≤ -20 ºC, or at 0 - 6 ºC, until sample preparation. However, the allowable holding time for samples depends on the storage temperature, as described in Section 8.5.	The change addresses the discrepancy between Sections 8.2 and 8.5
9.1.2.2 (c)	c) A narrative stating reason(s) for the modifications (see Section 1.6)	c) A narrative stating reason(s) for the modifications (see Section 1.5)	Section 1.6 does not exist in the draft method, and Section 1.5 is the correct citation
9.2.2	… 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 x 10n, where n is zero or an integer. Example matrix-specific detection limits are listed in Table 6.	… The minimum level of quantification (ML) can be calculated by multiplying the MDL by 3.18 and rounding the result to the nearest 1, 2 or 5 x 10n, where n is zero or an integer (see the Glossary for alternative derivations). Example matrix-specific method detection limits are listed in Table 6.	Responds to questions about the example values in Table 6. Also see the discussion of Table 6 below.
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.	When establishing the chromatographic conditions, it is important to consider the potential interference of bile salts during analyses of tissue samples. Inject the bile salt interference check standard containing TDCA (see Section 7.5 if the mobile phase is not acetonitrile) during the retention time calibration process and adjust the conditions to ensure that TDCA (or TCDCA and TUDCA) 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.	Naming this solution the bile salt interference check standard also simplifies discussions of its use. The potential interference with PFOS from bile salts is affected by the mobile phase used for the LC separation and laboratories choosing to use a mobile phase other than that specified in the draft procedure need to make the adjustments described here.

Errata for Draft Method 1633, Analysis of Per- and Polyfluoroalkyl Substances (PFAS) in Aqueous, Solid, Biosolids, and Tissue Samples by LC-MS/MS, EPA 821-R-21-001, August 2021			
Text Location	**Current Text**	**Corrected Text**	**Reason for Correction**
10.3.5	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.	The laboratory must analyze a bile salt interference check standard (see Section 7.5) 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 the bile salts (e.g., changing the retention time of the bile salts such that they fall 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	Naming this solution the bile salt interference check standard simplifies these discussions of its use.
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).	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 sample is now ready for solid-phase extraction (SPE) and cleanup (Section 12.0).	Section 11.2 addresses processing of aqueous samples, not extracts.
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.	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 analysis of the bile salt interference check standard in Section 10.3.5 before analyzing any tissue samples.	Making the discussion more generic in the context of the bile salt interference check changes noted above.
13.3, #9	9. TDCA standard (only if tissue samples are being analyzed)	9. Bile salt interference check standard (Section 7.5) (only if tissue samples are being analyzed)	Making the discussion more generic in the context of the bile salt interference check changes noted above.
15.1	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.	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 15.1.4 are met.	There is no subsection 15.1.5 in the draft method and subsection 15.1.4 is the correct citation.

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Text Location	**Current Text**	**Corrected Text**	**Reason for Correction**
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.	If the field sample result does not all meet the criteria stated in Sections 15.1.1 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.	The requirements in subsection 15.1.1 were omitted from the original text.
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, …	If the Q1 area for any compound exceeds the calibration range of the system, dilute a subsample of the sample extract with the methanolic ammonium hydroxide and acetic acid solution in Section 7.1.9 by a factor no greater than 10x and analyze the diluted extract.	The incorrect dilution solvent was called out in the draft procedure.
15.3.1	… 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.	… and analyze the diluted extract. If the responses for each EIS in the diluted extract meet the S/N and retention time requirements in Sections 15.1.1 and 15.1.2, and the EIS recoveries from the analysis of the diluted extract are greater than 5%, then the compounds associated with those EISs may be quantified using isotope dilution. Use the EIS recoveries from the original analysis to select the dilution factor, with the objective of keeping the EIS recoveries in the dilution above that 5% lower limit (i.e., if the EIS recovery of the affected analyte in the undiluted analysis is 50%, then the sample cannot be diluted more than 10:1; if the if the EIS recovery of the affected analyte in the undiluted analysis is 30%, then the sample cannot be diluted more than 6:1). Adjust the compound concentrations, detection limits, and minimum levels to account for the dilution.	The explanation of how to deal with quantification of the analytes in diluted extracts was not clear nor correct.

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Text Location	Current Text	Corrected Text	Reason for Correction
15.3.1 (continued)	None	If the EIS responses in the diluted extract do not meet those S/N and retention time requirements, then the compound cannot be measured reliably by isotope dilution in the diluted extract. In such cases, the laboratory must take a smaller aliquot of any affected aqueous sample and dilute it to 500 mL with reagent water and analyze the diluted 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. If a dilution greater than 10x is indicated, then the laboratory must analyze a diluted aqueous sample or a smaller aliquot of a solid sample.	Addresses samples where the diluted extract analysis does not meet the requirements. This becomes two new paragraphs within Section 15.3.1.
15.4.2.4	Report recoveries of all associated EIS compounds for all field samples and QC standards.	Report recoveries of all associated EIS compounds for all field samples and QC standards. If a sample extract was diluted and analyzed, report the EIS recoveries from both the original analysis and the analysis of the dilution.	Responds to questions from laboratories.
Table 2, PFHxS row	**Quantification Ion Mass** | **Confirmation Ion Mass** 98.9 | 79.9	**Quantification Ion Mass** | **Confirmation Ion Mass** 79.9 | 98.9	The quantitation and confirmation ions were reversed in the table.
Table 2, $^{13}C_4$-PFOS row	**Quantification Ion Mass** | **Confirmation Ion Mass** 98.9 | 79.9	**Quantification Ion Mass** | **Confirmation Ion Mass** 79.9 | 98.9	The quantitation and confirmation ions were reversed in the table.
Table 2, $^{13}C_3$HFPO-DA row	**Parent Ion Mass** 284.9	**Parent Ion Mass** 286.9	The parent ion mass was incorrect in the table.
Table 6	ML values for all three matrix types	New footnote added below Table 6.	Commenters noted that they could not reproduce the Minimum Level (ML) values in the table from the details in the method. An explanatory footnote has been added below the table.

Table 6. MDLs and ML values from the Single-laboratory Validation Study, by Matrix[1]						
	Aqueous (ng/L)		Solid (ng/g)		Tissue (ng/g)	
Compound	MDLs	ML[2]	MDLs	ML[2]	MDLs	ML[2]
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
PFTrDA	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.

[2] The ML values in this table were derived from the concentrations of the lowest calibration standard in Table 4, based on the alternative described in the Glossary, using the nominal sample volume (aqueous) or weight (all other matrices) described in the method.

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.