BS ISO 21675:2019

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Water quality — Determination of perfluoroalkyl and polyfluoroalkyl substances (PFAS) in water — Method using solid phase extraction and liquid chromatography-tandem mass spectrometry (LC-MS/MS)

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National foreword

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Water quality — Determination of perfluoroalkyl and polyfluoroalkyl substances (PFAS) in water — Method using solid phase extraction and liquid chromatography-tandem mass spectrometry (LC-MS/MS)

Qualité de l'eau — Détermination des substances d'alkyle perfluorés et polyfluorés (SPFA) dans l'eau — Méthode par extraction en phase solide et chromatographie liquide et spectrométrie de masse en tandem (CL-SM/SM)

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Foreword

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The procedures used to develop this document and those intended for its further maintenance are described in the ISO/IEC Directives, Part 1. In particular, the different approval criteria needed for the different types of ISO documents should be noted. This document was drafted in accordance with the editorial rules of the ISO/IEC Directives, Part 2 (see www.iso.org/directives).

Attention is drawn to the possibility that some of the elements of this document may be the subject of patent rights. ISO shall not be held responsible for identifying any or all such patent rights. Details of any patent rights identified during the development of the document will be in the Introduction and/or on the ISO list of patent declarations received (see www.iso.org/patents).

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For an explanation of the voluntary nature of standards, the meaning of ISO specific terms and expressions related to conformity assessment, as well as information about ISO's adherence to the World Trade Organization (WTO) principles in the Technical Barriers to Trade (TBT), see www.iso.org/iso/foreword.html.

This document was prepared by Technical Committee ISO/TC 147, *Water quality*, Subcommittee SC 2, *Physical, chemical and biochemical methods*.

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Water quality — Determination of perfluoroalkyl and polyfluoroalkyl substances (PFAS) in water — Method using solid phase extraction and liquid chromatography-tandem mass spectrometry (LC-MS/MS)

WARNING — Persons using this document should be familiar with normal laboratory practice. This document does not purport to address all of the safety problems, if any, associated with its use. It is the responsibility of the user to establish appropriate safety and health practices.

IMPORTANT — It is absolutely essential that tests conducted in accordance with this document be carried out by suitably qualified staff.

1 Scope

This document specifies a method for the determination of selected perfluoroalkyl and polyfluoroalkyl substances (PFAS) in non-filtrated waters, for example drinking water, natural water (fresh water and sea water) and waste water containing less than 2 g/l solid particulate material (SPM) using liquid chromatography-tandem mass spectrometry (LC-MS/MS). The compounds monitored by this method are typically the linear isomers. The group of compounds determined by this method are representative of a wide variety of PFAS. The analytes specified in Table 1 can be determined by this method. The list can be modified depending on the purpose for which the method is intended. The lower application range of this method can vary depending on the sensitivity of the equipment used and the matrix of the sample. For most compounds to which this document applies ≥0,2 ng/l as limit of quantification can be achieved. Actual levels can depend on the blank levels realized by individual laboratory.

The applicability of the method to further substances, not listed in Table 1, or to further types of water is not excluded, but is intended to be validated separately for each individual case.

NOTE 1 PFAS is used in this document to describe the analytes monitored. Many of the compounds in Table 1 are perfluoroalkyl and are also considered polyfluoroalkyl substances.

NOTE 2 The linear PFAS isomers are specified in this document. The branched isomers can be present in environmental samples, especially for PFOS. Annex E provides an example of an analytical approach to the chromatographic and spectroscopic separation of individual isomers.

2 Normative references

The following documents are referred to in the text in such a way that some or all of their content constitutes requirements of this document. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies.

ISO 3696, *Water for analytical laboratory use — Specification and test methods*

ISO 5667-1, *Water quality — Sampling — Part 1: Guidance on the design of sampling programmes and sampling techniques*

ISO 5667-3, *Water quality — Sampling — Part 3: Preservation and handling of water samples*

ISO 21253-1, *Water quality — Multi-compound class methods — Part 1: Criteria for the identification of target compounds by gas and liquid chromatography and mass spectrometry*

3 Terms and definitions

For the purposes of this document, the following terms and definitions apply.

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— ISO Online browsing platform: available at https://www.iso.org/obp

— IEC Electropedia: available at http://www.electropedia.org/

3.1
perfluoroalkyl and polyfluoroalkyl substances
PFAS
commonly used international abbreviation for organic compounds with replacement of most or all hydrogen atoms by fluorine in the aliphatic chain structure

Note 1 to entry: The term is used in the broader sense for per- and polyfluoroalkyl substances (PFAS), and per- and polyfluorinated compounds (PFC) as well.

4 Principle

The analytes listed in Table 1 are extracted from the water sample by solid-phase extraction using a weak anion exchange sorbent followed by solvent elution and determination by liquid chromatography-tandem mass-spectrometry.

The user should be aware that each analyte has its own specific optimum conditions and therefore modification of the analyte list could require the specification of additional conditions for each additional parameter.

Table 1 — Analytes determinable by this method

Analyte	IUPAC[a] name	Formula	Abbreviation	CAS-RN[b]
Perfluoro-*n*-butanesulfonic acid	1,1,2,2,3,3,4,4,4-Nonafluorobutane-1-sulfonic acid	$C_4HF_9O_3S$	PFBS	375-73-5
Perfluoro-*n*-hexanesulfonic acid	1,1,2,2,3,3,4,4,5,5,6,6,6-Tridecafluorohexane-1-sulfonic acid	$C_6HF_{13}O_3S$	PFHxS	355-46-4
Perfluoro-*n*-heptanesulfonic acid	1,1,2,2,3,3,4,4,5,5,6,6,7,7,7-Pentadecafluoroheptane-1-sulfonic acid	$C_7HF_{15}O_3S$	PFHpS	375-92-8
Perfluoro-*n*-octanesulfonic acid	1,1,2,2,3,3,4,4,5,5,6,6,7,7,8,8,8-Heptadecafluorooctane-1-sulfonic acid	$C_8HF_{17}O_3S$	PFOS	1763-23-1
Perfluoro-*n*-decanesulfonic acid	1,1,2,2,3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,10-Henicosafluorodecane-1-sulfonic acid	$C_{10}HF_{21}O_3S$	PFDS	335-77-3
Perfluorooctanesulfonamide	1,1,2,2,3,3,4,4,5,5,6,6,7,7,8,8,8-Heptadecafluoro-1-octanesulfonamide	$C_8H_2F_{17}NO_2S$	FOSA	754-91-6
N-methyl perfluorooctanesulfonamide	1,1,2,2,3,3,4,4,5,5,6,6,7,7,8,8,8-Heptadecafluoro-N-methyl-1-octanesulfonamide	$C_9H_4F_{17}NO_2S$	*N*-MeFOSA	31506-32-8
N-ethyl perfluorooctanesulfonamide	N-Ethyl-1,1,2,2,3,3,4,4,5,5,6,6,7,7,8,8,8-heptadecafluorooctane-1-sulfonamide	$C_{10}H_6F_{17}NO_2S$	*N*-EtFOSA	4151-50-2
N-methyl perfluorooctanesulfonamidoacetic acid	2-[1,1,2,2,3,3,4,4,5,5,6,6,7,7,8,8,8-Heptadecafluorooctylsulfonyl(methyl)amino] acetic acid	$C_{11}H_6F_{17}NO_4S$	*N*-MeFOSAA	2355-31-9
N-ethyl perfluorooctanesulfonamidoacetic acid	2-[Ethyl(1,1,2,2,3,3,4,4,5,5,6,6,7,7,8,8,8-heptadecafluorooctylsulfonyl)amino] acetic acid	$C_{12}H_8F_{17}NO_4S$	*N*-EtFOSAA	2991-50-6
6:2 Fluorotelomer sulfonic acid	3,3,4,4,5,5,6,6,7,7,8,8,8-Tridecafluorooctane-1-sulfonic acid	$C_8H_5F_{13}O_3S$	6:2 FTSA	27619-97-2
8:2 Fluorotelomer sulfonic acid	3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,10-Heptadecafluorodecane-1-sulfonic acid	$C_{10}H_5F_{17}O_3S$	8:2 FTSA	39108-34-4

[a] IUPAC: International Union of Pure and Applied Chemistry.

[b] CAS-RN: Chemical Abstract Services Registry Number.

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Table 1 *(continued)*

Analyte	IUPAC[a] name	Formula	Abbreviation	CAS-RN[b]
9-Chlorohexadecafluoro-3-oxanonane-1-sulfonic acid	2-(6-Chloro-1,1,2,2,3,3,4,4,5,5,6,6-dodecafluorohexoxy)-1,1,2,2-tetrafluoroethanesulfonic acid	$C_8HClF_{16}O_4S$	9Cl-PF3ONS	73606-19-6
Perfluoro-*n*-butanoic acid	2,2,3,3,4,4,4-Heptafluorobutanoic acid	$C_4HF_7O_2$	PFBA	375-22-4
Perfluoro-*n*-pentanoic acid	2,2,3,3,4,4,5,5,5-Nonafluoropentanoic acid	$C_5HF_9O_2$	PFPeA	2706-90-3
Perfluoro-*n*-hexanoic acid	2,2,3,3,4,4,5,5,6,6,6-Undecafluorohexanoic acid	$C_6HF_{11}O_2$	PFHxA	307-24-4
Perfluoro-*n*-heptanoic acid	2,2,3,3,4,4,5,5,6,6,7,7,7-Tridecafluoroheptanoic acid	$C_7HF_{13}O_2$	PFHpA	375-85-9
Perfluoro-*n*-octanoic acid	2,2,3,3,4,4,5,5,6,6,7,7,8,8,8-Pentadecafluorooctanoic acid	$C_8HF_{15}O_2$	PFOA	335-67-1
Perfluoro-*n*-nonanoic acid	2,2,3,3,4,4,5,5,6,6,7,7,8,8,9,9,9-Heptadecafluorononanoic acid	$C_9HF_{17}O_2$	PFNA	375-95-1
Perfluoro-*n*-decanoic acid	2,2,3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,10-Nonadecafluorodecanoic acid	$C_{10}HF_{19}O_2$	PFDA	335-76-2
Perfluoro-*n*-undecanoic acid	2,2,3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,11,11,11-Henicosafluoroundecanoic acid	$C_{11}HF_{21}O_2$	PFUnDA	2058-94-8
Perfluoro-*n*-dodecanoic acid	2,2,3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,11,11,12,12,12-Tricosafluorododecanoic acid	$C_{12}HF_{23}O_2$	PFDoDA	307-55-1
Perfluoro-*n*-tridecanoic acid	2,2,3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,11,11,12,12,13,13,13-Pentacosafluorotridecanoic acid	$C_{13}HF_{25}O_2$	PFTrDA	72629-94-8
Perfluoro-*n*-tetradecanoic acid	2,2,3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,11,11,12,12,13,13,14,14,14-Heptacosafluorotetradecanoic acid	$C_{14}HF_{27}O_2$	PFTeDA	376-06-7
Perfluoro-*n*-hexadecanoic acid	2,2,3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,11,11,12,12,13,13,14,14,15,15,16,16,16-Hentriacontafluorohexadecanoic acid	$C_{16}HF_{31}O_2$	PFHxDA	67905-19-5
Perfluoro-*n*-octadecanoic acid	2,2,3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,11,11,12,12,13,13,14,14,15,15,16,16,17,17,18,18,18-Pentatriacontafluorooctadecanoic acid	$C_{18}HF_{35}O_2$	PFOcDA	16517-11-6
8:2 Fluorotelomer unsaturated carboxylic acid	3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,10-Hexadecafluorodec-2-enoic acid	$C_{10}H_2F_{16}O_2$	8:2 FTUCA	70887-84-2
8:2 Polyfluoroalkyl phosphate diester	Bis(3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,10-heptadecafluorodecyl) hydrogen phosphate	$C_{20}H_9F_{34}O_4P$	8:2 diPAP	678-41-1
Hexafluoropropylene oxide dimer acid	2,3,3,3-Tetrafluoro-2-(1,1,2,2,3,3,3-heptafluoropropoxy)propanoic acid	$C_6HF_{11}O_3$	HFPO-DA	13252-13-6
4,8-Dioxa-3H-perfluorononanoic acid	2,2,3-Trifluoro-3-[1,1,2,2,3,3-hexafluoro-3-(trifluoromethoxy)propoxy] propanoic acid	$C_7H_2F_{12}O_4$	DONA	919005-14-4

[a] IUPAC: International Union of Pure and Applied Chemistry.

[b] CAS-RN: Chemical Abstract Services Registry Number.

5 Interferences

5.1 Interferences with sampling and extraction

Sample bottles (7.1) shall consist of materials that do not contaminate or change the composition of the sample during sample storage. All types of fluoropolymer plastics, including polytetrafluoroethylene (PTFE) and fluoroelastomer materials, shall be avoided during sampling, sample storage and extraction.

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Sample bottles (7.1) shall be checked for possible background contamination before use. If background contamination is suspected or detected in sample bottles (7.1), then wash sample bottles (7.1) with water (6.1) and methanol (6.6) prior to use. To avoid cross contamination, the sample bottles (7.1) should only be used once. The use of intermediate sample tubes (7.6) and vials (7.10) should be limited in the overall process to avoid contamination of loss by sorption. To avoid losses resulting from adsorption of target analytes to the wall of sample bottle (7.1) and reservoir column (7.4), extract all of the sample from the sample bottle (7.1) and rinse the wall of sample bottle (7.1) and reservoir column (7.4) with methanol (6.6).

Commercially available adsorbent materials often vary in quality or activity. Considerable batch-to-batch differences in quality and selectivity of these materials are possible. The recovery of a single substance may also vary with respect to its concentration. Therefore, check analyte recovery periodically at different concentrations and whenever new batches/lots of reagents or labware are used (12.1).

5.2 Interferences with LC-MS/MS

Substances with similar retention times that can produce ions with similar mass to charge ratios (m/z) to those produced by the analytes of interest may interfere with the determination.

These interferences may lead to incompletely resolved signals and/or additional signals in the mass chromatograms of target substances. Depending on their levels in the sample, such substances may affect the accuracy and precision of the results. The chromatographic separation is different with the LC column (see Annex C for examples). As long as the peak of interest can be separately integrated from interferences, it may be used.

Matrix interferences may be caused by contaminants that are co-extracted from the samples. The extent of matrix interferences varies considerably, depending on the nature of the samples. In drinking water and ground water, matrix interferences are usually negligible, whereas waste water and sea water matrices can be affected by matrix interferences that lead to ionization suppression or enhancement resulting in bias or reduced sensitivity of the method. As long as the required limits of quantification can be achieved in samples, samples can be diluted to minimize matrix effects.

Interferences arising directly from analytical instruments can be significant for unmodified commercial LC systems because many parts are made of PTFE and other fluoropolymers. It is necessary to check for possible blank contamination from the individual parts, such as tubing, solvent inlet filters, valve seals and the degassing equipment, and replace these with materials such as stainless steel and polyetheretherketone (PEEK), where possible.

NOTE Background contamination can arise from within the instrument. A delay column can be attached between the solvent mixer and injection valve to chromatographically resolve these background contaminants from the instrument and/or mobile phases from the target analytes.

The LC-vial caps shall be free of fluoropolymer material. Efforts should be taken to minimize background levels in procedural blank materials such that the procedural blank, including the instrumental blank, is at least 10-fold below the reporting limit.

6 Reagents

Whenever possible, use certified or analytical-grade reagents or residue free-analytical grade reagents stored in glass or polypropylene containers with metal or polypropylene lined caps. Avoid using reagents with fluoropolymer lined caps and check contamination levels of target substances using repeated blank determinations. Carry out additional cleaning or conditioning steps to ensure background levels are minimized, if necessary.

6.1 Water, blank-value free, e.g. complying with grade 1 as specified in ISO 3696.

Purified laboratory water can be used, but should be confirmed to be free of PFAS. The quality of water is checked by the same procedure given in 9.3.

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6.2 Acetic acid, $w(CH_3COOH)$ = 99,9 % mass fraction (999 g/kg).

6.3 Acetonitrile, CH_3CN.

6.4 Ammonia solution, $w(NH_3)$ = 25 % mass fraction (250 g/kg).

6.5 Ammonium acetate, $w(CH_3COONH_4)$ = 97 % mass fraction (970 g/kg).

6.6 Methanol, CH_3OH, blank-value free.

NOTE The quality of methanol is checked by evaporating 10 ml of methanol with a gentle stream of nitrogen gas (6.13) to 0,5 ml and determining levels according to this document.

6.7 Reference substances, see Table 1.

Reference substances are analytical standards used for quantitative determination of the method analytes. Use only reference substances or solutions, where the content of linear isomers is at least 95 %. Make sure that the individual reference substances do not contain detectable concentrations of other target analytes to be determined by analysing alternate lots or second sources.

NOTE Solutions of reference substances are commercially available.

6.8 Internal standard substances, see Table 3.

Internal standard substances are labelled forms of the reference substances to be used in the analytical procedure to correct for recovery due to losses of analyte or changes in analytical conditions that could result in bias. Make sure that the internal standard substances do not contain detectable concentrations of the analytes to be determined by analysing new lots using this document.

NOTE Solutions of internal standard substances are commercially available.

6.9 Preparation of the solutions

Calculate the concentration of all reference substances and internal standard solutions with regard to the anion content.

Store the solutions at (5 ± 3) °C in the dark, protected against evaporation. Bring them to room temperature prior to use (i.e. before dilution or spiking or injection).

6.9.1 Individual stock solutions of the reference substances

Stock solutions of the individual reference substances (6.7) in methanol (6.6) or acetonitrile (6.3) should be of mass concentration to enable dilution to the desired range, e.g. 50 µg/ml each.

6.9.2 Individual stock solutions of internal standard substances

Stock solutions of the individual internal standard substances (6.8) in methanol (6.6) or acetonitrile (6.3) should be of mass concentration to enable dilution to the desired range, e.g. 50 µg/ml each.

6.9.3 Native stock solution (reference substances)

Prepare a solution of the reference substances with a mass concentration of, for example, 0,1 µg/ml each.

Fill, for example, 1 ml of each solution of the individual reference substances, for example 50 µg/ml (6.9.1), into a 500 ml volumetric flask (7.7) and make the solution up to the mark with methanol (6.6).

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6.9.4 Labelled stock solution (internal standard substances)

Prepare a solution of the labelled internal standard substances with a mass concentration of, for example, 0,1 µg/ml each.

Fill, for example, 1 ml of each solution of the individual internal standard substances, e.g. 50 µg/ml (6.9.2), into a 500 ml volumetric flask (7.7) and make the solution up to the mark with methanol (6.6).

6.9.5 Spiking solution (reference substances)

Prepare a solution of the reference substances with a mass concentration of, for example, 10 ng/ml each.

Fill, for example, 1 ml of the native stock solution e.g. 0,1 µg/ml (6.9.3) into a 10 ml volumetric flask (7.7) and make the solution up to the mark with methanol (6.6).

This solution is used for recovery samples (see 12.1).

6.9.6 Spiking solution (internal standard substances)

Prepare a solution of the labelled internal standard substances with a mass concentration of, for example, 10 ng/ml each.

Fill, for example, 1 ml of the labelled stock solution e.g. 0,1 µg/ml (6.9.4) into a 10 ml volumetric flask (7.7) and make the solution up to the mark with methanol (6.6).

This solution is used for water samples (see 9.1.2) and spiking recovery samples (see 12.1).

6.9.7 Reference solution

Prepare the reference solutions by setting up dilutions of the stock solutions (6.9.3, 6.9.4). Add the same amount of internal standards to each reference solution.

Prepare the reference solution, for example a solution with a mass concentration of the substances to be determined and of the internal standard substances e.g. 1 ng/ml each.

Fill, for example, 0,1 ml of native stock solution (reference substances) e.g. 0,1 µg/ml (6.9.3) and 0,1 ml of labelled stock solution (internal standard substances) e.g. 0,1 µg/ml (6.9.4) into a 10 ml volumetric flask (7.7) and make the solution up to the mark with methanol (6.6).

6.10 Acetate buffer, for solid-phase extraction, 0,025 mol/l, pH 4.

Mix, for example, 0,5 ml of acetic acid (6.2) with e.g. 349,5 ml of water (6.1). Dissolve e.g. 0,116 g of ammonium acetate (6.5) in e.g. 60 ml of water (6.1). Mix e.g. 200 ml of the diluted acetic acid with e.g. 50 ml of the ammonium acetate solution.

6.11 Ammonia/methanol solution, for solid-phase extraction, with a mass fraction of approximately 0,1 %.

Mix, for example, 0,4 ml of ammonia solution (6.4) with e.g. 99,6 ml of methanol (6.6), with a volume of (NH_3) of e.g. 0,1 %.

6.12 Solid-phase extraction material, weak anion exchanger on a copolymer-based. Suitable materials are available commercially (see Annex A).

6.13 Nitrogen, N_2, purity >99,9 %.

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7 Apparatus

Equipment of which any part may come into contact with the water sample or the extract shall be free from interfering compounds.

The blank determination shall be conducted before the sampling. Clean labware and apparatus for solid-phase extraction by washing with water (6.1) and methanol (6.6) if background contamination is detected in labware and apparatus.

Equipment in contact with sample or reference solutions should be made of polypropylene or polyethylene. It was not tested except for compounds listed in Annex H for sampling whether the use of glassware may lead to adsorption of some of the analytes within the scope of the method.

7.1 Sample bottles

Narrow-neck flat-bottomed polypropylene or polyethylene bottles, normal volume 50 ml, 100 ml, 250 ml, 500 ml and 1 000 ml, with conical shoulders and screw caps.

NOTE Glass bottles can be used for compounds listed in Annex H. Glass bottles can be used for sampling provided that storage conditions of samples have been validated in each case. See Annex H for examples.

The entire sample shall be extracted and the appropriate-sized sample bottle should be used to collect the sample.

Sample bottles shall be checked for possible background contamination before use. The bottles and screw caps are washed with methanol (6.6) and dried before use in order to minimize contamination, if background contamination is detected in sample bottles.

7.2 Pipettes, with polypropylene tips.

7.3 Solid-phase extraction medias (cartridges or disks), made of inert non-leaching plastic, e.g. polypropylene or polyethylene frits.

The cartridges shall be packed with 50 mg to 1 000 mg of solid-phase extraction material (6.12) as sorbent. In general, 150 mg to 250 mg of sorbent (see Annex A) in a single cartridge is sufficient for up to 500 ml of water.

NOTE 1 The 500 mg of sorbent (see Annex A) in a single cartridge is sufficient for 1 000 ml of sea water.

NOTE 2 The stationary phase can be modified if analytes are not recovered quantitatively (for example neutral substances such as FOSA, *N*-MeFOSA and *N*-EtFOSA) using solid-phase extraction material (6.12) for example strongly hydrophilic reversed -phase copolymer or silica-based.

7.4 Reservoir column, normal volume 60 ml, with adaptor for cartridges (7.3), made of polypropylene or polyethylene.

7.5 Vacuum or pressure assembly, for the extraction step.

7.6 Sample tubes, made of, for example, polypropylene or polyethylene, for collecting and concentrating the eluate, e.g. test tube, nominal volume of 15 ml.

7.7 Volumetric flasks, with inert stoppers, made of polypropylene or polyethylene not containing fluoropolymer materials, normal volume 10 ml, 50 ml, 100 ml and 500 ml.

7.8 Graduated cylinder, normal volume 50 ml, 100 ml and 500 ml.

7.9 Evaporation assembly, using a nitrogen (6.13) stream passing through a stainless-steel needle.

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7.10 Vials, made of polypropylene or polyethylene not containing fluoropolymer materials, capacity e.g. 1,5 ml, depending on the auto-sampler, with e.g. polyethylene snap-on caps.

7.11 Liquid chromatograph, temperature-controlled and with all necessary accessories, including gases, LC columns (see Annex B), injector and tandem mass spectrometer (7.12).

7.12 Tandem mass spectrometer, with an ion source capable of generating ions for the analytes of interests [e.g. electrospray ionization (ESI)] and capable of determining the *m/z* values of selected precursor ions and product ions of the target substances listed in Table 2 and Table 3.

7.13 Analytical balance, capable of weighting to the nearest 0,1 g.

7.14 Centrifuge, capable of 3 000 rpm.

7.15 pH indicator paper.

8 Sampling

Take, preserve and handle samples as specified in ISO 5667-1 and ISO 5667-3.

For sampling, use thoroughly cleaned sample bottles (7.1). Fill the bottle with the water to be sampled.

Store samples in a refrigerator at (5 ± 3) °C and analyse within four weeks.

PFAS compounds with 11 or more carbon atoms may fall out of solution during storage. 8:2 FTUCA in sea water samples is not stable for four weeks. Sample storage conditions should be checked to confirm maximum sample storage times. A storage study should be conducted during the method validation stage for all analytes routinely determined. The entire sample shall be extracted. If the entire sample is analysed and the sample bottle is rinsed with solvent, the longer chain compounds should be quantitatively recovered.

9 Procedure

9.1 Solid-phase extraction

9.1.1 General

In general, in this procedure, samples are analysed without pre-treatment. Before starting the analysis, the sample and internal standard substances (6.9.6) shall have time to equilibrate to room temperature before analysis.

NOTE Annex F and Annex G provide examples of sample preparation for rapid methods without sample extraction by solid-phase extraction in 9.1, but these procedures do not have sufficient data for method validation.

9.1.2 Sample preparation

Weigh the sample bottle with its original cap and water sample, to the nearest 1 g or mark the line on the sample bottle (7.1) with the sample volume.

The entire sample shall be extracted. The water sample which is collected into the sample bottle (7.1) shall not be separated into new sample bottles to avoid losses of target analytes due to sorption to the sample bottle (7.1).

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The pH value of the sample shall be adjusted to the pH value of 3 with acetic acid (6.2) or ammonia solution (6.4) by pH indicator paper (7.15), if necessary.

NOTE Low recoveries of internal standard substances (6.9.6) can be improved by adjusting the pH value to 3, especially for short chain PFAS such as PFBA in a sea water sample.

Add the spiking solution containing the internal standard substances (6.9.6) to the water sample in the sample bottle (adding e.g. 100 µl of each, actual amount can be adjusted depending on the sample matrix) and mix thoroughly by shaking.

If the solid-phase extraction cartridge becomes clogged due to large amounts of suspended particulate in the sample, it may be possible to carry out the operation in Annex D or to divide the sample between two cartridges and pool the extracts. There may be a risk of increased blank level, which shall be checked for.

9.1.3 Conditioning of the solid-phase extraction material

The following procedure describes that used for commercially available 6 ml copolymer cartridges packed with 150 mg of sorbent sandwiched between two polyethylene frits.

Wash the cartridge in the following sequence with 4 ml of ammonia/methanol solution (6.11), 4 ml of methanol (6.6) and lastly 4 ml of water (6.1) prior to use. Make sure that the sorbent packing in the cartridge does not run dry. Retain the water in the cartridge (with the water level just above the packing) to keep the sorbent activated.

NOTE The solvent and water volumes used for conditioning depend on the amount the solid phase material used (for examples see Annex A).

9.1.4 Sample extraction

Start the extraction immediately after conditioning the sorbent packing. Make sure that no air bubbles are trapped in the sorbent bed when changing from conditioning to extraction. Do not let the sorbent material in the cartridge go dry and ensure it is immersed in water at all times.

Let sample (see 9.1.2) run through the cartridge, conditioned as specified in 9.1.3, at a rate of one drop per second (3 ml/min to 6 ml/min). Regulate the flow rate by changing the vacuum or the pressure (7.5), respectively.

Collect the sample, using a reservoir column (7.4) connected to the cartridge (7.3) with an adaptor.

Extract the entire sample in the sample bottle (7.1), to avoid losses resulting from adsorption to the wall of sample bottle (7.1).

Rinse the wall of sample bottle (7.1) and reservoir column (7.4) with a volume of methanol (6.6) which corresponds to at least 0,5 % of original sample volume. This aliquot of methanol is collected and used as elution solvent for sample extraction (see 9.1.5).

NOTE In the case of the longer chain PFAS such as PFUnDA, PFDoDA, PFTrDA, PFTeDA, PFHxDA and PFOcDA, loss can result from adsorption to the sample bottle (7.1) and reservoir column (7.4). Losses due to sorption to the sample bottle can be reduced by rinsing the sample bottle with methanol.

Measure the volume (in millilitres) of the water used in the extraction by reweighting the empty sample bottle with its original cap and calculate the net mass of sample, to the nearest 1 g, from the difference in weight (see 9.1.2). Assuming a density of 1 g/ml, the value of the net mass (in grams) is equivalent to the volume (in millilitres) of the water used in the extraction. Alternatively, add water (6.1) to the empty sample bottle up to the mark (see 9.1.2), and measure the water volume using a graduated cylinder (7.8). This volume is equivalent to the volume (in millilitres) of the original water sample.

9.1.5 Elution

Add 4 ml of water (6.1) and 4 ml of acetate buffer solution (6.10) to the cartridge and discard the eluate.

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