United States Environmental Protection Agency

Office of Water

www.epa.gov April 2022

Draft Method 1621

Screening Method for the Determination of Adsorbable Organic Fluorine (AOF) in Aqueous Matrices by Combustion Ion Chromatography (CIC)

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-22-002

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Notice

This document represents a draft of an AOF method currently under development by the EPA Office of Water, Engineering and Analysis Division (EAD). **This method is not approved 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 available. A revision of this draft method with a later publication date may be issued as public comments are received and multi-laboratory validation proceeds.

EAD expects to begin a multi-laboratory validation study of the procedure this year, 2022. 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 adjust 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, screening level method, subject to revision.

Acknowledgements

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

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 members of EPA's workgroup, and EPA's support contractor staff at General Dynamics Information Technology, including:

S. Bekah Burket	EPA Office of Water, Office of Science and Technology, Engineering and Analysis Division
Adrian Hanley	EPA Office of Water, Office of Science and Technology, Engineering and Analysis Division
Jody Shoemaker	EPA Office of Research and Development, Center for Environmental Solutions and Emergency Response
Jenifer Jones	EPA Office of Research and Development, Center for Environmental Solutions and Emergency Response
Thomas Speth	EPA Office of Research and Development, Center for Environmental Solutions and Emergency Response
Brian Pike	Pace Analytical® Services, LLC
Nick Nigro	Pace Analytical® Services, LLC
Mirna Alpizar	General Dynamics Information Technology
Harry McCarty	General Dynamics Information Technology
Brian Milewski	ASTM D19
William Lipps	Shimadzu Scientific Instruments, Inc., and member of ASTM D19
Tom Patten	Pace Analytical® Services, LLC and member of ASTM D19
Takuro Kato	Mitsubishi Chemical and member of ASTM D19
Jayesh Gandhi	Metrohm and member of ASTM D19

Disclaimer

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
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DRAFT Method 1621 – Screening Method for the Determination of Adsorbable Organic Fluorine (AOF) in Aqueous Matrices by Combustion Ion Chromatography (CIC)

1.0 Scope and Application

1.1 Method 1621 is for use in the Clean Water Act (CWA) as a screening method to estimate the concentration of adsorbable organic fluorine (AOF) in aqueous matrices by combustion ion chromatography (CIC).

1.2 The method measures organofluorine compounds from per- and polyfluoroalkyl substances (PFAS) and non-PFAS fluorinated compounds such as pesticides and pharmaceuticals that can be retained on at least 80 mg of granular activated carbon (GAC). The result is reported as the concentration of fluoride (F^-) in the sample.

1.3 Short-chain (less than 4 carbons) organofluorine compounds are poorly retained on GAC while long-chain (more than 8 carbons) hydrophobic organofluorine compounds readily adsorb to surfaces. These issues can cause low recoveries for these types of fluorinated compounds.

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 stratify in the container.

1.5 Relative to the Clean Water Act and the methods approved for compliance monitoring at 40 CFR Part 136, AOF is a "method-defined parameter" (MDP). A MDP is a parameter defined solely by the method used to determine the analyte. In the case of AOF, Draft Method 1621 estimates an aggregate concentration of any organofluorine compounds in the sample that are retained on the sorbent. Therefore, EPA has limited the extent to which this method may be modified without prior EPA review. At this time, the analyst may not use sorbents other than granular activated carbon, amounts of granular activated carbon less than 80 mg per sample, or sample containers made from different materials. EPA may include additional restrictions following completion of the multi-laboratory validation study.

1.6 The method development studies demonstrated that there were no negative effects on the adsorption of organofluorine onto carbon when the sample contains organic carbon concentrations up to 140 mg/L. The nitrate wash employed in the method is capable of removing up to 8 mg/L of inorganic fluorine that may be adsorbed in conjunction with organofluorines, reducing the positive bias from inorganic fluoride. The method is capable of adsorbing AOF in samples with chloride concentrations up to 500 mg/L without causing peak interference in the chromatogram. Due to the ubiquitous occurrence of PFAS, unless strict cleaning protocols are followed, the method can be subject to significant blank contamination. Nonetheless, the method can reliably screen for organofluorines at low part-per-billion levels.

1.7 For the reasons discussed in Sections 1.2 to 1.6, EPA has classified this procedure as a screening method that may be used to estimate the aggregate contributions of the organofluorine compounds present in the sample. As such, data users are advised that the numerical results generated by this method are not expected to be as accurate or precise as those from targeted methods for PFAS. In addition, given the large number of potential PFAS and other organofluorine compounds that may be present in environmental samples, EPA has adjusted some of the quality control and method performance testing approaches employed in this procedure to those more suited for a screening method.

1.8 The instrumental portion of this method is for use only by analysts experienced with CIC or under the close supervision of such qualified personnel. Each laboratory that uses this method must demonstrate the ability to generate acceptable results using the procedure in Section 9.2.

2.0 Summary of Method

2.1 Environmental aqueous samples are prepared and adsorbed using method-specific procedures. A 100-mL sample aliquot is passed through two GAC columns, each containing 40 mg of carbon.

2.2 The GAC columns are rinsed with sodium nitrate to remove inorganic fluoride, combusted at least 1000 °C in an oxygen or oxygen/argon stream, and the gaseous hydrogen fluoride is absorbed into reagent water.

2.3 The fluoride is separated by ion chromatography (IC) and identified by comparing sample fluoride retention time to retention times for calibration standards acquired under identical IC conditions and by using the external standard technique.

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 or plasticware, and other sample processing hardware may yield artifacts and elevated baselines causing misinterpretation of chromatograms. Specific selection of reagents and solvents may be required. All items must be routinely demonstrated to be free from interferences through the analysis of matrix blanks prepared with each batch.

4.2 Clean all equipment prior to, and after each use, to avoid fluoride cross-contamination. Typical cleaning solvents used include water, methanol, and methanolic ammonium hydroxide (1%). The residual AOF content of disposable plasticware and filters must be verified by batch/lot number.

4.2.1 Glass containers have been found to have high levels of fluoride contamination; therefore, it is highly recommended that the use of glass labware be avoided for this method whenever possible. If any glass labware is used in the preparation or storage of reagents, the glassware must be cleaned by rinsing with methanol, followed by rinsing with reagent water, followed by a second methanol rinse, and air drying.

4.2.2 **T**he combustion boats must be cleaned by soaking in methanol for at least one hour, scrubbing with cotton swabs, rinsing with reagent water, and baking in a kiln or furnace (Section 6.2.2), then stored in a desiccator or stored submerged in reagent water until use. Prior to use, baked combustion boats must be rinsed with methanol and dried. If the combustion boats are stored longer than three days, then they must be baked again prior to use.

4.2.3 All removable parts of the adsorption unit must be cleaned before sample analyses begin, between samples, and at the end of the analysis batch, using a cleaning solvent combination suggested by the manufacturer, followed by rinsing with reagent water.

4.2.4 All reusable plasticware must be washed with methanol followed by a minimum of five reagent water rinses.

4.2.5 It is highly recommended that the laboratory record and track which adsorption unit ports are used to process each sample. This may assist the laboratory in tracking possible sources of contamination for individual samples.

4.3 All materials used in the analysis must be demonstrated to be reasonably free from interferences by running method blanks (Section 9.3) at the beginning and with each sample batch (samples started through the extraction process on a given analytical batch to a maximum of 10 field samples).

4.3.1 The adsorption tubes can be a major source of fluoride contamination, both organic and inorganic. The type of capping material used in the columns can contribute as much as 30% of the background fluoride level. The level of fluoride background can also vary greatly between vendors. The analysis of method blanks provides important information regarding the presence of such contamination.

4.3.2 Each lot of GAC adsorption tubes must be tested by analyzing method blanks to ensure that contamination does not preclude AOF quantitation at the reporting limit. There may be background variation within a lot number; therefore, at least four columns from each new lot number must be tested prior to putting the lot number into use.

4.3.3 Baseline contamination of GAC observed during method development ranged from 0.1 to 6 µg F^-/L, based on a 100-mL sample size. Smaller sample volumes will produce higher method blank results.

4.3.4 Opened packages of GAC columns and combustion boats must be stored in a desiccator when not in use to prevent possible contamination from laboratory air.

4.4 Matrix interferences may be caused by contaminants that are co-adsorbed 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 co-adsorbed non-fluorine-containing contaminants can cause breakthrough of organofluorine analytes on the GAC.

4.5 Samples containing total suspended solids above 100 mg/L may clog the GAC columns and adsorption unit lines, which may prevent proper adsorption of the sample. These types of samples will need to be pretreated by following the instructions in Section 11.2.

4.6 Alcohols, aromatic substances and carboxylic acids may lead to negative interference. Organic carbon above 140 mg/L may lead to negative interference by inhibiting quantitative adsorption of halogen bound organic substances to the activated carbon. These effects may be assessed by dilution of the initial sample or spiking the sample with a standard solution at a known concentration.

4.7 Inorganic fluoride in the sample may lead to biased high results. The nitrate wash step in this method removes inorganic fluoride at a concentration up to 8 mg/L inorganic fluoride in the sample. Samples containing more than 8 mg/L of inorganic fluoride should be diluted prior to analysis. In addition, chloride elutes closely with fluoride in the ion chromatography portion of the analysis, which may cause an interference when chloride is present at concentrations greater than 500 mg/L.

4.8 Some organofluorines readily adsorb to surfaces. This method does not mitigate for surface adsorption because the use of solvents to rinse sample bottles, tubing, or other surfaces in the adsorption unit also would elute AOF from the GAC. Thus, results may be biased low for organofluorines that are prone to surface adsorption. Additionally, analytical results between different manufacturer's adsorption apparatus may not be comparable due to differences in the numbers of surfaces (e.g., tubing, syringes, etc.) with which the sample comes in contact during sample loading onto the GAC.

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 Perfluorooctanoic acid 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 **S**amples suspected to contain PFAS or other organofluorine compounds are handled using essentially the same techniques employed in handling infectious materials. Well-ventilated, controlled access laboratories are required. Assistance in evaluating the health hazards of particular laboratory conditions may be obtained 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 Protective equipment – Disposable plastic gloves, apron or lab coat, safety glasses or mask, and a glove box or fume hood. 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. Nitrile or polyethylene gloves are commonly used to reduce exposure of the hands.

5.3.2 Personal hygiene – Hands and forearms should be washed thoroughly after each manipulation and before breaks (coffee, lunch, and shift). Avoid the use of hand lotions.

5.3.3 Confinement – Isolated work areas posted with signs, segregated glassware and tools, and plastic absorbent paper on bench tops will aid in confining contamination.

5.3.4 Waste Handling – Good technique includes minimizing contaminated waste. Plastic bag liners should be used in waste cans.

5.4 Samples that may contain high concentrations of biohazards must be handled with PFAS- and fluoride-free 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 highly contaminated samples.

6.0 Equipment and Supplies

Note: *Brand names, suppliers, and part numbers listed below are for illustration purposes only and no endorsement is implied. Equivalent performance may be achieved using apparatus and materials other than those specified here (also see 40 CFR 136.6). 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 – Liquid samples– Sample bottle, high-density polyethylene (HDPE) or polypropylene, with linerless HDPE or polypropylene caps.

Note: *Do not use fluoropolymer-lined caps for sample containers.*

6.1.2 Compositing equipment – Automatic or manual compositing system incorporating containers cleaned per bottle cleaning procedure above (Section 4.2.4). 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 cleaning combustion boats

6.2.1 Laboratory sink with overhead fume hood

6.2.2 Kiln or muffle furnace – Capable of reaching 450 ºC within 2 hours and maintaining 450 - 500 ºC ± 10 ºC, with temperature controller and safety switch (Cress Manufacturing Co., Santa Fe Springs, CA, B31H, X31TS or equivalent; Thermo Scientific™ Cat# FB1315M or equivalent). For safety, the kiln or furnace should be vented outside the laboratory, or to a trapping system.

6.3 Equipment for sample and standard preparation

6.3.1 PFAS- and fluoride-free gloves, nitrile or polyethylene

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 Equipment for determining total suspended solids

6.3.4.1 Oven – Capable of maintaining a temperature of 103 - 105 ºC

6.3.4.2 Desiccator

6.3.4.3 Filtration equipment (e.g., vacuum filtration apparatus)

6.3.4.4 Filter, glass fiber, 47 mm, 1 µm (Millipore Sigma™ or equivalent)

6.3.5 Balances

6.3.5.1 Analytical – Capable of weighing 0.1 mg

6.3.5.2 Top loading – Capable of weighing 10 mg

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

6.3.7 Centrifuge tubes, polypropylene, with polypropylene caps for storing standards (Fisher Cat# 14-959-49A or equivalent)

6.3.8 Quartz wool, used as a pre-filter of samples with > 50 mg/L of suspended solids (Fisher Cat# AC451040500 or equivalent)

6.3.9 Graduated cylinder, polypropylene, 100 mL (Fisher Cat# 3664-0100 or equivalent) used to measure the reagent water used to prepare QC samples.

6.4 Syringes

6.4.1 Plastic, Luer-lock syringes for manually rinsing and drying of GAC (BD Cat No. 303134, or equivalent) if not able to be done automatically by the adsorption unit

6.4.2 Micro-syringes able to measure 10 – 1000 µL, for methanol-based solutions

6.4.3 Pipets with disposable polypropylene or polyethylene tips may be used for preparing calibration standards.

6.5 Adsorption Unit – Analytik Jena Automatic Preparation Unit (APU) Sim, or equivalent

The adsorption apparatus must be capable of loading 100 mL of aqueous sample at a flowrate no higher than 3 mL/min on up to three adsorption columns in series. The apparatus must also be capable of washing the columns with sodium nitrate. The apparatus must allow for either manual or automated rinsing of the columns with at least 3 mL of reagent water and drying with air.

Certain adsorption units may have fluoropolymer transfer lines that cannot be removed. In these cases, the preparation batch method blanks must be rotated among the ports to ascertain that there is no contamination in the system.

6.6 Glass columns containing 40 mg GAC (Metrohm Cat# SNG-ICT0008 or equivalent) and GAC column holder assemblies (Metrohm Cat# SNG-ICT0013, or equivalent). A minimum of two GAC columns in series are required per field and QC sample. Each new lot must be tested (Section 4.3.2) prior to using for sample analysis.

6.7 Steel rod to transfer GAC into combustion boats (Mandel Cat# NS-TX3SCR, or equivalent)

6.8 CIC Instrument – Automated furnace system integrated to an IC (Metrohm 930 Combustion IC PP system or other CIC equivalent system). Other instrument configurations may be possible.

6.8.1 Autosampler capable of introducing combustion boats with the GAC into the combustion tube

6.8.2 Ceramic combustion boats (Metrohm Cat# SNG-IC0030), or equivalent

6.8.3 Electric furnace capable of maintaining an inlet temperature of at least 900 °C and minimum outlet temperature of at least 1000 °C. The furnace unit must be capable of maintaining a constant flow oxygen gas (e.g., 300 - 400 mL/min) during pyrohydrolytic combustion per manufacturer's recommendations. If the system uses argon plus oxygen, then the instrument should be able to keep a constant flow of argon gas (e.g., ~ 200 mL/min).

6.8.4 Gas absorption unit with an absorption tube which can hold a minimum of 5 mL volume and can maintain a constant volume of reagent water. The adsorption unit should be able to rinse the tube and transfer lines with reagent water or other solution as per manufacturer's instructions before and after each sample combustion. The gas absorption unit should be interfaced to the IC and be capable of injecting an aliquot of the final absorption solution into the IC.

6.8.5 Combustion tube made of quartz, or a combination of quartz and ceramic, capable of withstanding temperatures up to 1100°C. The combustion tube may include quartz wool, or other suitable medium, to provide appropriate sample mixing to ensure complete combustion of the sample.

6.8.6 Water delivery system capable to introduce reagent water at a controlled rate to produce pyrohydrolytic combustion.

6.9 IC System

6.9.1 IC Column (Metrohm Metrosep A Supp 7, 5 µm, 4x150 mm column, Cat# 6.1006.620 or equivalent)

6.9.2 Guard column (Metrohm Metrosep A Supp 5, 5 µm, 4x50 mm, Cat# 6.1006.550 or equivalent)

6.9.3 Pumping system capable of delivering eluent at a flow rate between 0.1 to 3.0 mL/min with a precision of at least 2 %, or as recommended for this determination by the manufacturer.

6.9.4 Continuous eluent generation cartridges or devices are permitted.

6.9.5 Conductivity detector, temperature controlled to 0.01 °C, capable of at least 1 to 1000 µS/cm on a linear scale.

6.9.6 Interfaced data system to acquire, store, reduce, and output chromatographic data. The computer software should have the capability of processing data by recognizing an IC peak within the given retention time window. The software must allow integration of the

fluoride peak area response within specified time limits. The software must be able to construct linear regressions or quadratic calibration curves and calculate concentrations.

6.10 Desiccator - Used to store open packages of GAC columns (see Section 11.3.5) and combustion boats (Bel-Art™ Cat# F420310000 or equivalent). If storing combustion boats in a desiccator, the GAC columns and combustion boats must be stored separately (i.e., in a different desiccator) to prevent possible cross contamination.

7.0 Reagents and Standards

Unless otherwise indicated, all reagents shall conform to the specifications of the Committee on Analytical Reagents of the American Chemical Society (ACS), 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 determination.

7.1 Reagents

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

7.1.1 Methanol – (HPLC grade or better, 99.9% purity), verified by lot number before use, store at room temperature.

7.1.2 Methanolic ammonium hydroxide, *1%* – Add 3.3 mL ammonium hydroxide (28-30%) to 97 mL methanol, store at room temperature, replace after 1 month. Used for rinsing plasticware, glassware, and equipment (Section 4.2).

7.1.3 Potassium hydroxide (KOH), fluoride free, (Fisher Cat# P250-500 or equivalent) - Prepare a 0.5 M solution by dissolving 2.81 g KOH in 100 mL of reagent water (Section 7.1.8) and replace after 6 months. Used to adjust sample pH.

7.1.4 Sodium nitrate (Sigma-Aldrich Cat# 221341 or equivalent)

7.1.4.1 Sodium nitrate, *2M* – Dissolve 170 g of $NaNO_3$ (Section 7.1.4) in 1 L reagent water (Section 7.1.8), store at room temperature, replace after 1 year.

7.1.4.2 Sodium nitrate, *0.01M* – Dilute 5 mL of 2M $NaNO_3$ (Section 7.1.4.1) into 1 L reagent water (Section 7.1.8), store at room temperature in an amber container or protected from light and replace after 6 months.

7.1.5 Sodium thiosulfate (Alfa Aesar™ Cat# A1762936 or equivalent used to dechlorinate samples.

7.1.6 Gases

7.1.6.1 Oxygen – 99.999%

7.1.6.2 Argon – 99.999% (if used by the instrument)

7.1.7 Eluents – Various eluents may be used, based on manufacturer's recommendations, provided they give the proper resolution for the component peak. The background tends to

be lower when using hydroxide-based eluents (e.g., KOH, NaOH) in place of carbonate-based eluents. To lower the background when using carbonate-based eluents, an additional carbonate removal device may be installed.

Note: *The use of electrolytically generated hydroxide-based and carbonate-based eluents is permitted with this method and strongly encouraged. Part numbers are available from the instrument manufacturers. When using electrolytically prepared eluents only reagent water needs to be added to the system.*

7.1.7.1 Example Carbonate-based Eluent – Dissolve 0.168 g sodium bicarbonate [*1.0 mM*] (Fisher Cat# S233-500 or equivalent) and 0.6784 g sodium carbonate [*3.2 mM*] (Fisher Cat # S495-500 or equivalent) in 2 L reagent water (Section 7.1.8). Store at room temperature, shelf life 1 month.

7.1.7.2 Example Hydroxide-based Eluent – Dissolve 8.0 g (or 5.25 mL) of 50% w/w sodium hydroxide (Fisher Cat# SS410-4 or equivalent) in 950 mL of reagent water (Section 7.1.8). This makes a 100 mM solution. Store at room temperature, shelf life 2 months.

7.1.8 Reagent water – Laboratory purified water, ASTM Type I or Type II (Reference 13). Test prior to use for PFAS content by analyzing method blanks.

7.2 Diethyl-*p*-phenylenediamine (DPD) Test Kit (Fisher Cat# S72367 or equivalent) or chlorine test strips (Hach Cat# 2745050 or equivalent), for determination of residual chlorine.

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.

Note: *Standards for the PFAS compounds used in this method are available from Cambridge Isotope Laboratories, Wellington Laboratories, and other suppliers. Listing of specific suppliers does not constitute a recommendation or endorsement for use. All diluted solutions must be stored in HDPE or polypropylene containers that have been thoroughly rinsed with methanol.*

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 stoppered volumetric flask and fill to the mark with the required solvent. Once the compound is completely dissolved, transfer the solution to a clean container and cap.

When not being used, store standard solutions as recommended by the vendor. Place a mark on the container at the level of the solution for standards prepared in methanol so that solvent loss by evaporation can be detected. Replace the solution if solvent loss has occurred. The laboratory must maintain records of the certificates for all standards for traceability purposes.

7.3.1 Sodium Fluoride Stock Standard, 1000 mg F^-/L, 1 mL = 1 mg F^- (Reagents Specialty Chemicals and Solutions Cat# CF135100-120A or equivalent)

7.3.2 Fluoride Working Standard, 10 mg F^-/L – Dilute 0.5 mL of fluoride standard (Section 7.3.1) in 50 mL reagent water (Section 7.1.8).

7.3.3 Calibration standard solutions – At least 5 fluoride calibration concentrations, spanning the AOF concentration range of interest, are required to prepare the initial calibration curve (Section 10.1) if using a linear calibration model, while a minimum of 6 calibration standards are required if a second-order calibration model is used. The lowest level calibration standard must meet a signal-to-noise ratio of 10:1. Prepare the calibration standards by diluting the fluoride working standard (Section 7.3.1.1) in reagent water. A fluoride calibration range of approximately 1.0 µg F^-/L to 50 µg F^-/L (assuming a 100-mL sample volume) may be used as a starting point for determining the calibration range.

Low-level and mid-level calibration verification (CV) standards are analyzed, alternating throughout the analytical batch, at the beginning and at the end of the analytical batch, for the purpose of calibration verification.

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 10:1, as long as the required number of calibration points is met.*

7.3.4 PFAS Standards – Perfluorohexanesulfonic acid (PFHxS) Working Standard, 50 µg/mL (Wellington Laboratories Cat# L-PFHxS or equivalent)

AOF is calculated as a concentration of fluoride ion; therefore, the fluoride concentration for the PFAS standard used in the initial demonstration of capability and for spiking samples must be calculated for the compound as follows:

$$C_F = C_{PFAS} \times n_F \times \frac{MW_F}{MW_{PFAS}}$$

Where,

C_F	=	Concentration of fluoride ion in PFAS (or non-PFAS) compound in µg/mL
C_{PFAS}	=	Concentration of the PFAS (or non-PFAS) compound in µg/mL
MW_{PFAS}	=	Molecular weight of the PFAS (or non-PFAS) compound in $g \cdot mol^{-1}$
n_F	=	Number of fluorine atoms in the compound
MW_F	=	Atomic weight of fluorine, $g \cdot mol^{-1}$ (18.998)

7.4 Desiccant – W. A. Hammond Drierite™ Cat# 21005, or equivalent

8.0 Sample Collection, Preservation, Storage, and Holding Times

8.1 Collect samples in HDPE or polypropylene 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 automated sampling equipment. Collect approximately 100 mL of sample in an appropriately sized HDPE or polypropylene bottle. For this test, headspace is allowable.

Note: *Because this method consumes the entire volume of sample, aqueous samples must be collected at least in triplicate to allow sufficient volume for the determination of total suspended solids, inorganic fluoride, residual chlorine, and pH, as well as having sufficient volume for matrix spikes or reanalysis.*

Because the target analytes are known to bind to the interior surface of the sample container, subsampling should be avoided whenever possible. Therefore, if a sample volume smaller than 100 mL is to be used for analysis, it is highly suggested to collect the sample in an appropriately sized HDPE or polypropylene container.

8.2.2 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 cooler temperature is 0 - 6 ºC upon receipt. Once received by the laboratory, the samples must be stored at 0 - 6 ºC until sample preparation.

8.3 Holding times - Aqueous samples should be analyzed as soon as possible; however, samples may be held in the laboratory protected from light for up to 90 days when stored at 0 - 6 ºC (Reference 10).

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 organofluorine 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.

9.1.1 The laboratory must perform an initial demonstration of the capability (IDC) to demonstrate 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 changes in sample volumes and ion chromatography columns. 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 IDC 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.

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 narrative stating reason(s) for the modifications (see Section 1.7).

c) Results from all quality control (QC) tests comparing the modified method to this method, including:

i. Calibration (Section 10.0)
ii. Calibration verification (Section 13.1)
iii. Initial precision and recovery (Section 9.2.1)
iv. Analysis of blanks (Section 9.3)
v. Accuracy assessment (Section 9.7)

d) 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. Adsorption dates
iii. Analysis dates and times
iv. Analysis sequence/run chronology
v. Sample weight or volume (Section 11.0)
vi. Dilution data (Section 11.0)
vii. Instrument
viii. Ion chromatography column (dimensions, etc.)
ix. Operating conditions (temperatures, temperature program, flow rates)
x. Chromatograms and other recordings of raw data
xi. Quantitation reports, data system outputs, and other data to link the raw data to the results reported

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

Note: *The use of alternative ion chromatography columns or eluents may result in different retention times.*

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 9.3.

9.1.4 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 (OPR) standards, spiked with PFHxS standard at mid-level, and method blanks. These procedures are given in Section 13.0.

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 Adsorb and analyze four aliquots of reagent water, spiked with sufficient volume of PFHxS solution (Section 7.3.4) to achieve a concentration at or above the mid-point of the calibration curve. At least one method blank must be prepared with the IPR batch. If more than one method blank 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 must be included in this test.

9.2.1.2 Using results of the set of four analyses, compute the average percent recovery and the relative standard deviation (RSD) of the concentration.

9.2.1.3 Pending completion of the multi-laboratory validation study, the RSD should be < 20% and the average percent recovery should be 70 – 130%. If the RSD and average percent recovery meet the acceptance criteria, system performance is acceptable, and analysis of blanks and samples may begin. If, however, the RSD exceeds the precision limit or the average percent recovery falls outside the range for recovery, system performance is unacceptable. Correct the problem and repeat the test (Section 9.2).

9.2.2 Method detection limit (MDL) - Each laboratory must also establish an MDL for organofluorine using the MDL procedure at 40 CFR Part 136, Appendix B. The MDL determination should be performed using PFHxS as the spiked compound.

9.3 Method blanks – At least two method blanks must be analyzed with each sample batch (Section 4.3) to demonstrate freedom from contamination.

Because fluoride contamination in the GAC columns is a significant problem, maintaining a historical record of method blank data is highly recommended. Use the equation below to calculate the method blank AOF concentration if the data system software package does not perform the calculation automatically.

$$AOF = \left(C_{MB} \cdot \frac{V_{abs}}{V_{MB}}\right)$$

Where,

AOF = concentration of adsorbable organic fluorine in µg/L
C_{MB} = Method blank measured concentration (sum concentration of top and bottom GAC) reported without volume corrections
V_{abs} = total volume of absorption solution in mL of the sample prior to injection to IC
V_{MB} = volume in mL of method blank adsorbed

9.3.1 Analyze the first method blank aliquot before the analysis of the OPR (Section 14.5).

9.3.2 If AOF is found in the blank at a concentration greater than one-third the regulatory compliance limit, 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 lot number of GAC columns, or to contamination of the adsorption unit (e.g., a dirty column position), all affected samples must be re-adsorbed and reanalyzed, provided that enough sample volume is available.

If after sample reanalysis, the new blank still shows 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.

9.4 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 13.1 and 13.2), and for initial (Section 9.2.1) and ongoing (Section 13.3) 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.

9.5 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.

9.6 An OPR, processed through the entire sample preparation procedure in Section 11.3, is required with each adsorption batch. It is not possible to assess the recovery of every possible organofluorine compound that may be detected with this technique. Therefore, PFHxS was chosen as the OPR fortification analyte to represent organofluorine compounds because it is well retained on GAC in reagent water. It is recommended that OPR be spiked at a concentration at the mid-level of the calibration curve.

9.7 Matrix spike (MS) and matrix spike duplicate (MSD) are required at a frequency of one set per adsorption batch. PFHxS was chosen as the MS/MSD fortification analyte to represent organofluorine compounds because it is well retained on GAC in reagent water. It is recommended that MS/MSD be spiked at a concentration at or above the mid-level of the calibration curve. Calculate the percent recovery as follows:

$$\%R = \frac{\left(C_{MS} \cdot \frac{V_{abs}}{V_{MS}}\right) - C_n}{C_s} \times D \times 100$$

Where,

C_{MS} = MS measured concentration (sum concentration of top and bottom GAC) reported without blank subtraction and volume corrections
V_{abs} = total volume of absorption solution for the sample prior to IC injection, mL
V_{MS} = volume sample adsorbed, mL
C_n = native concentration of the unfortified sample
C_s = spiked concentration
D = dilution factor (if diluted before analysis)

Calculate the relative percent difference (RPD) as follows:

$$\text{RPD} = \frac{|\text{MS} - \text{MSD}|}{(\text{MS} + \text{MSD})/2} \times 100$$

Where,
MS = percent recovery of the MS
MSD = percent recovery of the MSD

10.0 Calibration and Standardization

10.1 Establishing instrument operating conditions

Establish IC operating conditions using the manufacturer's instructions. The table below shows the instrument settings used for the single-laboratory method validation. Other CIC systems may require different instrument settings; however, the method performance criteria must still be met.

IC parameters

Separator column:	Metrosep A Supp 7, 5 µm, 4 x 150 mm
Guard column:	Metrosep A Supp 5, 5 µm, 4 x 50 mm
Flow rate:	0.7 mL/min
Injection volume:	1 mL
Column temperature:	35 °C
Suppressor:	Metrohm
Detector cell temperature:	40 °C

Combustion parameters

Furnace inlet temperature:	1050 °C
Furnace outlet temperature:	1050 °C
Pyrolysis tube:	Quartz tube
Oxygen flow:	300 mL/min
Humidified argon flow:	200 mL/min
Water supply scale:	0.3 mL/min bled into front of combustion tube by dosing unit
Sample boat:	Ceramic
Adsorption solution:	Reagent water
Final absorption solution volume:	7.0 mL

10.2 Chromatographic conditions

10.2.1 The chromatographic conditions must be optimized for compound separation and sensitivity. Fluoride must be adequately resolved chromatographically from the water dip (if present). A resolution of at least 1.0 minute is necessary between the fluoride peak and the water dip for proper peak integration. Chloride concentrations over 500 mg/L, some constituents of the eluant, and other potential interferences may elute very close to the fluoride peak; therefore, the chromatographic conditions must be optimized to give a resolution of at least 1.0 minute between the fluoride and chloride and other interfering peaks. The same optimized operating conditions must be used for the analysis of all standards, blanks, IPR and OPR samples, and field samples.

The following table gives the chromatographic conditions used during the single-laboratory validation for this method using the specified instrument and column. Different instruments may require different operating conditions, especially when using different eluents. Any modifications to IC conditions must still produce conditions such that co-elution of the fluoride with the nearby chloride peak or other interfering peaks is minimized to reduce the probability of peak misidentification or positive bias.

Chromatographic Conditions

Isocratic Elution at 0.7 mL/min using 3.2 mM Sodium Bicarbonate and 1 mM Sodium Carbonate

Autosampler Combustion Program

Position	Wait time (s)	Speed (mm/s)
200 mm	15	5
End	540	5
Cool	300	

10.2.2 Establish the retention time of fluoride by combusting, in triplicate, the mid-level standard from the ICAL and analyze under the established IC conditions.

10.2.3 Calculate the width of the retention time window for fluoride at plus or minus three times the standard deviation of the retention times determined in Sec. 10.2.2.

10.2.4 Establish the center of the retention time window for fluoride by using the absolute retention time from the calibration verification standard at the beginning of the analytical batch. For samples analyzed during the same 24-hour period as an initial calibration, use the retention time of the mid-point standard of the initial calibration.

Note: *Procedures for establishing retention time windows from other sources may be employed if they are clearly documented and provide acceptable performance. Such performance may be evaluated using the results for the spiked QC samples described in this method, such as OPR samples and matrix spike samples.*

10.2.5 The retention time windows must be recentered when a new IC column is installed and after major maintenance.

10.3 Initial calibration

Prior to the analysis of samples, each IC system must be calibrated with a minimum of 5 standard concentrations (Section 7.3.3 and Table 2). The lowest concentration CAL standard should be at approximately three times the MDL. This method calibrates and quantifies the adsorbable organic fluorine concentration in a sample, using the external standard technique. Standards are injected onto a clean ceramic boat and go through combustion, but not through carbon adsorption. By combusting the calibration standards instead of doing a direct injection into the IC, the calibration compensates for any bias produced by loss of analyte that may happen during combustion of field samples. The calibration model should be either a linear regression (minimum 5 calibration points required) or a quadratic (minimum 6 calibration points required), no higher than 2nd order, and not forced through zero.

Note: *The laboratory may use more standards for the calibration curve as long as the criteria in Section 10.3.3 can be met.*

10.3.1 Initial calibration frequency

Each IC system must be calibrated after instrument and chromatographic conditions have been established before analysis of samples can take place. The initial calibration must be repeated whenever the laboratory takes corrective action that might change or affect the initial calibration criteria (e.g., replacing IC column, replacing suppressor), or if the CV acceptance criteria have not been met.

10.3.2 Initial calibration procedure

Prepare a minimum of 5 calibration standards (6 if using quadratic fit) using the working fluoride standard (see Section 7.3.2). Analyze each calibration standard by injecting 200 µL into a clean combustion boat. Pending completion of the multi-laboratory validation study, the calculated concentrations of the calibration standards should be within 80 – 120% of the concentration true value. The retention time (RT) for fluoride in the ICAL must fall within the window established in Section 10.2.4.

10.3.3 Instrument linearity

Assess the calibration linearity based on the relative standard error (RSE) using the equation below. The RSE must be ≤ 20%.

$$RSE = 100 \times \sqrt{\sum_{i=1}^{n} \frac{\left[\frac{x_i' - x_i}{x_i}\right]^2}{n - p}}$$

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)

10.3.4 Initial calibration corrective actions

If the instrument linearity or recovery 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.

10.4 Calibration Verification

10.4.1 The calibration must be verified by analyzing a calibration verification (CV) standard at the beginning and at the end of the analytical batch. CVs at low-level and mid-level calibration should be used throughout the analytical batch (e.g., opening CV at low-level, next CV at mid-level).

10.4.2 The RT for fluoride on the CV must fall within the window established in Section 10.2.4.

11.0 Sample Preparation and Adsorption

Samples containing high levels of suspended solids (e.g., visible particulates) can clog the GAC columns and prevent proper sample adsorption. Therefore, for aqueous samples from unfamiliar

sources that contain particulates, total suspended solids must be determined using the procedures in Section 11.1. Other analytical interferences are residual chlorine, sample pH, and inorganic fluoride. To check for these interferences in samples from unfamiliar sources, follow the sample pre-treatment steps in Section 11.2. One sample container must be selected to check for interferences in Section 11.1 and 11.2. Any necessary adjustments to the sample based on results from Section 11.2 must be done on the container reserved for AOF analysis (Section 11.3).

Note: *Do not use any fluoropolymer-containing 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 Total Suspended Solids (TSS) – Aqueous liquids or multi-phase samples consisting of mainly an aqueous phase. Choose one sample container to perform TSS analysis as described below. Separate this container and do not use the leftover sample for AOF sample processing in Section 11.3.

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

11.1.2 Filter 20.0 ± 0.02 mL of well-mixed sample through the filter.

Note: *The purpose of this analysis is simply to minimize the risk of clogging the GAC column from samples with TSS content on the order of 100 mg/L (see Section 11.3). Therefore, the volume of sample employed here was selected to identify samples high in TSS and still be representative of the bulk sample collected. The TSS result is not used in calculation of the AOF result itself.*

11.1.3 Dry the filter a minimum of 12 hours in the oven (Section 6.3.4.1) at a temperature between 103 -105 ºC and cool in a desiccator (Section 6.3.4.2).

11.1.4 Calculate total suspended solids (TSS) as follows:

$$TSS\ (mg/L) = \frac{(A - B) \times 1000}{C}$$

Where,
A = Weight of filter + sample residue, mg
B = Initial weight of filter, mg
C = Volume of sample filtered, mL

11.2 Sample Pretreatment

11.2.1 pH Verification

Before adsorption, sample pH must be verified to be ≥ 5. Using the same sample container used in Section 11.1, take an aliquot and check the sample pH with pH paper (Section 6.3.6 and Reference 12). If the pH of the sample is below 5, adjust the pH of the sample in the container to be used for AOF analysis (Section 11.3) with 0.5 M potassium hydroxide (Section 7.1.3). Lower molarities of KOH may be used; however, make sure that the final volume of solution used to adjust the pH does not exceed 1 mL to prevent diluting the sample.

11.2.2 Dechlorination

Test sample for the presence of chlorine using an aliquot from the sample container used in Section 11.1 and either type of testing product in Section 7.2. If chlorine is detected, dechlorinate the sample in the container that was designated for AOF analysis (Section 11.3) using sodium thiosulfate at 5 mg of thiosulfate per every 1 mg/L of chlorine, prior to sample adsorption.

11.2.3 Determination of Inorganic Fluoride

Determine the concentration of inorganic fluoride in the sample. This may be done by directly injecting a diluted aliquot of well-mixed sample from the container used in Section 11.1 into the IC using the same instrument parameters and procedure as for the calibration standards. A 1:10 sample dilution using reagent water (Section 7.1.8) is recommended to prevent damage to the column and minimize the matrix effects for the chromatographic peaks. Alternatively, other recognized procedures for determining fluoride in aqueous samples may be employed, including ion-selective electrode procedures (sample distillation is *not* required). Calculate the concentration of inorganic fluoride in mg/L.

Levels of inorganic fluoride > 8 mg F^-/L may bias the AOF results high. If inorganic fluoride concentration exceeds 8 mg F^-/L, transfer an aliquot of the sample from the container designated to be used for AOF analysis, to a new PFAS-free HDPE or polypropylene container, and dilute it with reagent water (final volume of sample must be 100 mL) to reduce the concentration of inorganic fluoride to just below 8 mg F^-/L. The lowest dilution possible must be targeted to prevent overdiluting the analytes of interest.

Note: *Transferring a sample aliquot between the parent container and a secondary container for the purpose of sample dilution may result in loss of analyte due to the propensity of organofluorines to adhere to surfaces.*

11.3 Sample Processing

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 levels of organofluorine; however, subsampling should be avoided whenever possible. Typical sample size is approximately 100 mL.

Note: *For samples containing > 100 mg/L of TSS (Section 11.1), a piece of quartz wool should be placed inside an empty column holder ahead of the GAC columns and used as a prefilter to prevent the GAC columns from clogging. The prefilter must be washed with the nitrate wash solution and combusted in the same manner as the GAC columns. The concentration from the prefilter must be added to the concentration of the GAC columns.*

If a sample is processed using a glass wool pre-filter, a method blank must be processed using the same prefilter material because the prefilter material will add to the background fluoride levels in the sample. The total fluoride of the blank (pre-filter + 2 GAC columns) must be subtracted from the sample results.

11.3.1 Prepare two method blanks and one OPR using reagent water in HDPE or polypropylene bottles. Select a volume of reagent water that is typical of the samples in the batch. Spike the OPR sample with PFHxS (Section 7.3.4.1) at the concentration of the mid-level calibration point.

11.3.2 Spike the MS and MSD samples directly in the original bottles at the mid-level calibration point using PFHxS (Section 7.3.3.1). Adjust the spiking level and the sample dilution to keep the expected results for these two aliquots within the calibration range of the instrument.

11.3.3 Weigh each sample and MS/MSD bottle (with the lid) to 0.1 g. The final volume of the aqueous sample analyzed is determined by weighing the full sample bottle and then the empty sample bottle (Section 11.3.9).

11.3.4 Add 0.5 mL of 2M sodium nitrate (Section 7.1.4.1) to each field sample, method blank, OPR, MS, MSD, and any other QC sample containers. Mix by inverting the containers 3 – 4 times.

11.3.5 Place two GAC columns in tandem in the column holder apparatus of the adsorption unit.

Note: *Opened packages of GAC columns must be kept tightly sealed and stored in a desiccator (Section 6.10) to minimize possible contamination from laboratory air.*

11.3.6 Load the sample onto the GAC columns at a flowrate of no more than 3 mL/min. The typical volume of sample is 100 mL, but because the sample containers allow collection of volumes up to 125 mL, the adsorption unit should be set up to consume the entire volume of sample if possible. Monitor for leaks throughout the column holder assembly.

11.3.7 After sample loading is complete, wash both GAC columns and the pre-filter material, if used, with 25 mL of 0.01M sodium nitrate (Section 7.1.4.2) to remove inorganic fluoride.

Note: *Higher volume of sodium nitrate may be used to further reduce positive interference by inorganic fluoride; however, this may decrease the recovery of organofluorides.*

11.3.8 Rinse both columns with at least 20 mL of reagent water to remove any residual nitrate. This rinse also helps to prolong the life of the pyrolysis tube by decreasing the potential for devitrification. Dry the columns using at least 3 mL of air using the syringe. Remove the GAC columns from the column holders. If samples are not to be combusted immediately, store columns in a desiccator at room temperature no longer than 5 days.

11.3.9 Determine the final volume of each field and QC sample used by weighing the empty sample bottle (with the lid) to 0.1g and subtracting the final weight from the initial weight (Sec. 11.3.3). In calculating the sample volume, assume a sample density of 1.0 g/mL.

11.3.10 Flush the sample transfer lines with 40 mL reagent water followed by 40 mL of a suitable solvent (e.g., methanol) and another 40 mL reagent water to ensure that all surfaces in contact with samples are flushed. Use solvents or solvent combinations recommended by the manufacturer to prevent harm to components of the adsorption unit.

12.0 Instrumental Analysis

12.1 Instrument operating conditions (Section 10.1 and 10.2) must have been set, and an initial calibration (Section 10.3) performed prior to analyses of any field samples. The same instrument operating conditions used for the initial calibration must be used to analyze field and QC samples.

12.2 Equilibrate the IC system by pumping eluent through until a stable baseline is obtained. Because fluoride from the air can accumulate in the combustion tube while the instrument sits idle for any period of time, at least two empty combustion boats must be combusted prior to analysis of any sample to clear fluoride from the system.

12.3 Perform a calibration verification check prior to sample analysis.

12.4 Transfer the carbon from each column into separate combustion boats using the column rod (do not combine the GAC from both columns on one combustion boat). When pushing the carbon into the combustion boats, push from the bottom of the column to ensure that the transfer rod does not come in contact with any particulates that may have collected on the capping material at the top of the carbon column. The percent breakthrough between the top and bottom GAC columns must be calculated (Section 14.3). Do not mix the columns (top column vs. bottom column) so that the percent breakthrough can be properly calculated. If quartz wool was used for prefiltration, it must be combusted in a separate combustion boat. Place the combustion boats in the autosampler of the CIC using clean forceps and combust following manufacturer's instructions.

Note: *The combustion boats must be cleaned by pre-baking them in a kiln or furnace for at least 1 hour prior to use. Clean combustion boats should be stored in a desiccator or submerged in reagent water to reduce buildup of organofluorine on the boats from laboratory air and should be used within 3 days of baking.*

12.5 The analytical sequence for a batch of samples analyzed during the same time period is as follows. Standards must be brought to room temperature prior to use.

1. Boat Blank Checks (at least 2)
2. Calibration Verification Standard
3. First Method Blank
4. OPR
5. Samples (10 or fewer)
6. MS
7. MSD
8. Second Method Blank
9. Calibration Verification Standard

If the results are acceptable, the results from the closing calibration verification analysis (#9 above) may be used as the opening analysis for the next analytical sequence, beginning with a new first method blank. The analytical sequence must not exceed 24 hours.

12.6 If the fluoride response exceeds the calibration range for any sample, the sample must be re-adsorbed using a lower sample volume, if there is sufficient sample volume available.

13.0 Performance Tests during Routine Operations

The following performance tests must be successfully completed as part of each routine instrumental analysis shift described in Section 12.5 above.

Note: All the acceptance limits listed in the sections below are subject to change after completion of the multi-laboratory validation study and are provided here as interim guidance.

13.1 Calibration verification (CV)

Prior to the analysis of any samples, analyze a low-level calibration standard.

13.1.1 The calibration is verified by analyzing a CV standard at the beginning of each analytical sequence and at the end of the analytical sequence. The percent recovery for both CV standards should be within 80 - 120%.

13.1.2 If the CV criterion is not met, recalibrate the instrument according to Section 10.3.

13.2 Fluoride in the field samples and batch QC must elute within ± 0.2 minutes of the retention time corresponding to the fluoride peak from the beginning CV.

13.3 Method Blanks – Analyze the first method blank after the analysis of the CV and prior to the analysis of samples. Analyze the second method blank after the MS/MSD and before the closing CV. The method blank should not exceed 5 µg/L, but preferably, method should be < 3 µg/L.

13.4 Ongoing precision and recovery (OPR)

13.4.1 After the CV and the method blank, analyze the OPR prior to analysis of samples from the same batch to ensure the analytical process is under control.

13.4.2 Compute the percent recovery of the analyte.

$$Recovery\ (\%) = \frac{Concentration\ found\ (\mu g/L)}{Concentration\ spiked\ (\mu g/L)}\ x\ 100$$

13.4.3 The OPR recovery should be within 70 – 130%. If the fluoride recovery meets the acceptance criteria, system performance is acceptable, and analysis of blanks and samples may proceed. If, however, the concentration falls outside of the given range, the adsorption processes are not being performed properly. In this event, correct the problem, re-prepare the sample batch, and repeat the ongoing precision and recovery test if sample volume allows.

13.5 Accuracy and Precision

13.5.1 Matrix Spikes – After the samples, prior to closing CV. The MS recovery should be within 50 – 150% recovery.

13.5.2 Duplication – Analyze an MSD after the MS. Calculate the relative percent difference of the recoveries using the equation in Section 9.7. The RPD should be < 20%.

14.0 Data Analysis and Calculations

14.1 Qualitative determination and peak identification

Organofluorine is positively identified in a blank, sample, or QC sample if the retention time (RT) falls within the window established in Sec. 10.2.4, centered on the RT from the opening CV.

14.2 Estimating sample concentration

As noted in Section 1.7, EPA has classified this procedure as a screening method that may be used to estimate the aggregate contributions of the organofluorine compounds in the sample. Because of the common occurrence of background levels of fluoride in the sorption media, the method subtracts the amount of fluoride observed in the method blank from the fluoride in the sample to estimate the AOF attributable to the sample itself. This type of background subtraction, while not permitted in many other EPA methods, is not unique to this procedure, and is a fundamental aspect of the method-defined parameter that is AOF. Moreover, the fluoride background levels in the GAC are likely to result in a method detection limit driven by the MDL_b rather than the spiked sample MDL_s value.

Calculate the AOF concentration as a sum of the top and bottom GAC columns (and prefilter(s) if used) concentrations adjusted by subtracting the sum of the top and bottom of the initial method blank, using the data software package for the CIC and the multipoint calibration.

Alternatively, the equation below can be used to calculate the AOF concentration if the data software package will not perform the calculation. Do not use the daily calibration verification data to quantitate AOF in samples. Adjust the final AOF concentrations to reflect the actual sample volume used.

$$AOF = (S \cdot \frac{V_S}{100} - BL \cdot \frac{V_{BL}}{100}) \cdot D$$

Where,

AOF = Concentration of adsorbable organic fluorine in µg/L
S = Concentration of the sample measurement
BL = Concentration of method blank measurement
V_S = Total amount of absorption solution in mL of the sample prior to injection to IC
V_{BL} = Total amount of absorption solution in mL of the method blank measurement prior to injection to IC
D = Dilution factor if sample was diluted before analysis

Note: *The blank subtraction process inherent in this method may yield negative concentrations for some samples. See Section 14.3 for instructions on how to report such results.*

Calculate the percent breakthrough for each of the column pairs, per sample, as follows:

$$\%\ Breakthrough = \frac{(M_2 - B_2) \times 100}{[(M_1 - B_1) + (M_2 - B_2)]}$$

Where,

M_1 = Mass measured for the first column, µg F^-
M_2 = Mass measured for the second column, µg F^-
B_1 = Mass measured for first column of the initial method blank, µg F^-
B_2 = Mass measured for second column of the initial method blank, µg F^-

Percent breakthrough must not exceed 50%. If % breakthrough exceeds the limit, the sample is exceeding the GAC capacity and must be reanalyzed at a lower volume, if sufficient sample volume is available.

14.3 Reporting of analytical results

The data reporting practices described here are focused on using a screening method for NPDES monitoring needs and may not be relevant to other uses of the method. Because of the inherent blank subtraction involved in this screening method, the calculations in Section 14.2 may yield negative values. Therefore, the reporting practices below differ from those in most other EPA methods.

14.3.1 Report results in µg F^-/L. Other units may be used if required in a permit or for a project. Report all QC data with the sample results.

14.3.2 Reporting level

14.3.2.1 Report adsorbable organofluorine as fluoride in each field sample or QC standard above the MDL determined in the laboratory to 2 significant figures. Report any value below the MDL and any negative value in a field sample as "< MB," where "MB" is the concentration of the associated method blank result subtracted in Section 14.2, or as required by the regulatory/control authority or permit.

14.3.2.2 Although the samples are blank corrected, the blank results must also be reported with the data. Report fluoride in a blank at or above the MDL to 2 significant figures. Report a result for each 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.

14.3.3 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.

15.0 Method Performance

Routine method performance is validated through analysis of aqueous reference samples, including MS/MSD samples. Ongoing method performance is monitored through QC samples analyzed alongside samples. The parameters monitored include percent recovery and blank concentrations.

16.0 Pollution Prevention

16.1 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.

16.2 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.

16.3 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).

17.0 Waste Management

17.1 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).

17.2 Samples at pH < 2 or 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.

17.3 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).

18.0 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. American Public Health Association, American Water Works Association, Water Environment Federation. SM 1090 Laboratory Occupational Health and Safety. In: Lipps W C, Braun-Howland E B, Baxter T E, eds. "Standard Methods for the Examination of Water and Wastewater." Approved by Standard Methods Committee 2010, published online August 2018; revised 23 April 2021. https://www.standardmethods.org/doi/10.2105/SMWW.2882.011

5. ASTM-2018. ASTM D3370-18, "Standard Practice for Sampling Water from Flowing Process Streams," ASTM International, West Conshohocken, PA, 2018, www.astm.org.

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. Willey, J., R. Anderson, A. Hanley, M. Mills, C. Hamilton, T. Thompson, and A. Leeson. 2021. Report on the Single-Laboratory Validation of PFAS by Isotope Dilution LC-MS/MS. Strategic Environmental Research and Development Program (SERDP) Project ER19-1409.

11. “Report on the Single-laboratory Validation of Clean Water Act Method 1621 for Adsorbable Organic Fluoride (AOF),” USEPA, Office of Science and Technology, Washington DC, EPA-820-R-22-003, April 2022.

12. ASTM-2021. ASTM D-8294-21, Standard Practice for Estimating pH to Verify Status of Aqueous Samples, ASTM International, West Conshohocken, PA, 2018, www.astm.org

13. ASTM-2018, ASTM D1193-06(2018), Standard Specification for Reagent Water, ASTM International, West Conshohocken, PA, 2018, www.astm.org

19.0 Tables, Diagrams, Flowcharts, and Validation Data

Table 1. Example Retention Time and MDL Values

Analyte	Retention Time (min)[a]	Organofluorine Fortified Conc. (µg/L)[b]	MDL (µg/L)
AOF	5.9	5.0	2.4

[a] Retention time based on Metrohm Metrosep A Supp 7 column and isocratic carbonate-bicarbonate elution. Other column and elution reagents will give different retention times.

[b] Fluoride concentration used to determine MDL_S. PFHxS used as source of organofluorine.

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 2. Example Calibration Standard Solutions

	Calibration Standards (µg F^-/ L)					
Analyte	CS1	CS2	CS3	CS4	CS5	CS6
AOF	1.0	2.0	5.0	10	25	50

Data for this table are derived from the single-laboratory validation study and are only provided as examples for this draft method.

Table 3. IPR and OPR QC Based on Single-Lab Data

	IPR		OPR
Compound	Recovery (%)	RSD (%)	Recovery (%)
PFHxS	85 – 115	7.5	75 - 110

Data for this table are derived from the single-laboratory validation study and are only provided as examples for this draft method.

Table 4. Summary of Quality Control

Method Reference	Requirement	Specification and Frequency
Section 10.3	Initial Calibration (ICAL)	Minimum 5 calibration standards for linear model and 6 calibration standards for non-linear models.
Sections 10.2.2, 10.2.3, 14.1	Retention Time (RT) window	After ICAL and at the beginning of analytical sequence
Section 10.4, 13.1	Calibration Verification (CV)	At the beginning and every 10 samples
Sections 9.1.3, 9.3, 13.3	Method Blank	Two per preparation batch
Section 13.4	Ongoing Precision and Recovery (OPR)	One per preparation batch
Sections 9.7, 11.3	Matrix Spike (MS/MSD)	One set per preparation batch

20.0 Figures

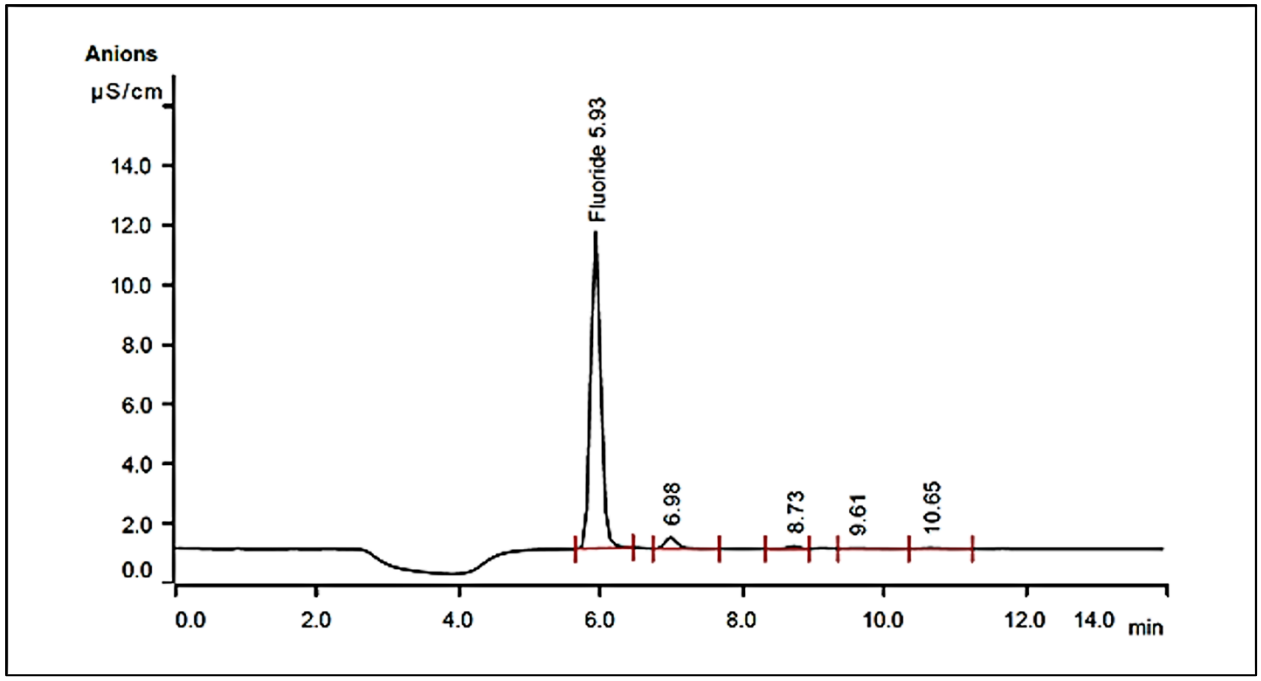

Figure 1. Sample Ion Chromatography

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

º C	degrees Celsius
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

cm	centimeter
g	gram
L	liter
M	molar
mg	milligram
min	minute
mL	milliliter
mm	millimeter
µg	microgram
w/w	percent weight by weight

21.2 Definitions and acronyms (in alphabetical order)

Adsorption batch – A set of up to 10 field samples adsorbed onto GAC during the same period of time (8-hour shift) using the same lot of GAC, reagents, and standards.

Analysis batch – A set of samples analyzed on the CIC during a 24-hour period that is bracketed by the appropriate method blanks and CVs.

AOF – Adsorbable organic fluoride, an aggregate measure of per- and polyfluoroalkyl substances (PFAS) and non-PFAS fluorinated organic compounds (such as pesticides and pharmaceuticals) that can be adsorbed from an aqueous sample on granular activated carbon and determined by combustion ion chromatography. Because of the common occurrence of background levels of fluoride in the sorption media, the method subtracts the amount of fluoride observed in the method blank from the fluoride in the sample to estimate the AOF attributable to the sample itself. AOF results are reported as the concentration of fluoride ion (F^-) in the sample.

Calibration standard (CS) – A solution prepared from a secondary standard and/or stock solutions and used to calibrate the response of the instrument.

Calibration verification standard (CV) – The mid-point calibration standard (CS-4) that is used to verify calibration. See Table 2.

CFR – Code of Federal Regulations

CIC – Combustion ion chromatography

CWA – Clean Water Act

GAC – Granular activated carbon

IC – Ion chromatograph or ion chromatography

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.

Matrix Spike (MS) and Matrix Spike Duplicate (MSD) – A set of field samples to which a known quantity of organofluorine compound is added in the laboratory prior to analysis. The MS/MSD are processed in the same way as the regular samples to determine the contribution of matrix interference and bias to the analytical method.

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-defined parameter (MDP) – A parameter defined solely by the method used to determine the analyte.

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

Must – This action, activity, or procedural step is required.

NIOSH – The National Institute of Occupational Safety and Health

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.

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 and have been found in animals, plants, as well as in the human body.

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.

Relative Standard Error (RSE) – The standard error of the mean divided by the mean and multiplied by 100.

RT – Retention time**;** the time it takes for an analyte to elute off the column

Should – This action, activity, or procedural step is suggested but not required.

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.