Table 5.6

Sample Containers, Preservation Techniques and Hold Times

Table 5.6.1
Sample Containers, Preservation Techniques and Hold Times
(Water/Wastewater Samples)

Parameter	Container [1]	Preservation [2,3]	Maximum Holding Time [4]
Nutrients/Wet Chemistry:			
Alkalinity [17]	P, G	Cool, ≤6 °C	14 days
Ammonia[17]	P, G	Cool, ≤6 °C, H_2SO_4 to pH < 2	28 days
Chloride	P, G	None required	28 days
Chlorophylls[21, 22]	P, G	Dark, cool, 6°C Filtered, dark, freeze, -20°C	48 hours chilled until filtration, and analyze immediately or 48 hours chilled until filtration and 28 days (frozen) after filtration
Color[17]	P, G	Cool, ≤6 °C	48 hours
Fluoride	P	None required	28 days
Hydrogen (pH)	P, G	None required	Analyze immediately
Kjeldahl & organic nitrogen[17]	P, G	Cool, ≤6 °C, H_2SO_4 to pH < 2	28 days
Molecular	P	Cool < 10°C	48 hours chilled, (samples with test code PMA-HF183 analyze immediately)
Nitrate[17]	P, G	Cool, ≤6 °C	48 hours
Nitrate-nitrite[17]	P, G	Cool, ≤6 °C, H_2SO_4 to pH < 2	28 days
Nitrite[17]	P, G	Cool, ≤6 °C	48 hours
Oil & grease	G	Cool, ≤6 °C, H_2SO_4 to pH < 2	28 days
Organic carbon[17]	P, G	Cool, ≤6 °C, H_2SO_4 to pH < 2	28 days
Inorganic Carbon[17]	P	Cool, ≤6 °C	14 days
Orthophosphate[17]	P, G	Filter immediately, Cool, ≤6 °C	48 hours
Phosphorus, total[17]	P, G	Cool, ≤6 °C, H_2SO_4 to pH < 2	28 days
Residue, total[17]	P, G	Cool, ≤6 °C	7 days
Residue, filterable[17]	P, G	Cool, ≤6 °C	7 days
Residue, Nonfilterable (TSS) [17]	P, G	Cool, ≤6 °C	7 days
Silica[17]	P	Cool, ≤6 °C	28 days
Specific conductance[17]	P, G	Cool, ≤6 °C	28 days
Sulfate[17]	P, G	Cool, ≤6 °C	28 days
Sulfide[17]	P, G	Cool, ≤6 °C. add Zn Acetate + NaOH to pH > 9	7 days
Temperature	P, G	None required	Analyze immediately
Turbidity[17]	P, G	Cool, ≤6 °C	48 hours

Algal Assay			
Algal Growth Potential, Limiting Nutrients	P, G	Dark, cool, ≤6 °C	72 hours to processing, 30 days from processing to set up
Aquatic Toxicity			
Toxicity, acute and chronic[18]	P, G	Cool, ≤6 °C	36 hours
Bacterial			
Coliform, total, fecal, and *E. coli*[19, 20]	PA, G	Cool, <10 °C, 0.008% $Na_2S_2O_3$[5]	8 hours
Fecal streptococci[19]	PA, G	Cool, <10 °C, 0.008% $Na_2S_2O_3$[5]	8 hours
Enterococci[19]	PA, G	Cool, <10 °C, 0.008% $Na_2S_2O_3$[5]	8 hours
Biological			
Invertebrates	P	Formalin or Cool < 6°C	Not applicable or 24 hours chilled until preserved with formalin or identified
Algae	P	Cool < 6°C	Analyze immediately or preserve with Lugol's
Metals : [7]			
Metals (except chromium VI and mercury)	P, G	HNO_3 to pH < 2	6 months
Mercury (trace)	G	HCl [16] to pH < 2	90 days
Mercury, routine	P, G	HNO_3 to pH < 2	28 days

Table 5.6.1
Sample Containers, Preservation Techniques and Hold Times
(Water/Wastewater Samples)
(continued)

Parameter	Container [1]	Preservation [2,3]	Maximum Holding Time [4]
Organics: [8]			
Purgeable Halocarbons[17]	G, Teflon-lined septum	Cool, ≤6 °C Cool, ≤6 °C, 0.008 % $Na_2S_2O_3$[5], HCl to pH 2 [9]	7 days 14 days
Purgeable aromatic[17]	G, Teflon-lined septum	Cool, ≤6 °C Cool, ≤6 °C, 0.008 % $Na_2S_2O_3$ [5], HCl to pH 2 [9]	7 days 14 days
Acrolein & acrylonitrile[17]	G, Teflon-lined septum	Cool, ≤6 °C, 0.008 % $Na_2S_2O_3$ [5], adjust pH to 4-5 [10]	14 days
Phenols [11, 17]	G, Teflon-lined cap	Cool, ≤6 °C, 0.008 % $Na_2S_2O_3$[5], store in dark	7 days until extraction 40 days after extraction
Benzidines [11, 17]	G, Teflon-lined cap	Cool, ≤6 °C, 0.008 % $Na_2S_2O_3$[5], store in dark	7 days until extraction [12, 13]
Phthalate esters [11, 17]	G, Teflon-lined cap	Cool, ≤6 °C, 0.008 % $Na_2S_2O_3$[5], store in dark	7 days until extraction 40 days after extraction
Nitrosamines [11, 14, 17]	G, Teflon-lined cap	Cool, ≤6 °C, 0.008 % $Na_2S_2O_3$[5], store in dark	7 days until extraction 40 days after extraction
PCBs [11, 17]	G, Teflon-lined cap	Cool, ≤6 °C, store in dark	7 days until extraction 40 days after extraction
Nitroaromatics & Isophorone[17]	G, Teflon-lined cap	Cool, ≤6 °C, 0.008 % $Na_2S_2O_3$[5], store in dark	7 days until extraction 40 days after extraction
Polynuclear Aromatic Hydrocarbons (PAHs) [17]	G, Teflon-lined cap	Cool, ≤6 °C, store in dark	7 days until extraction 40 days after extraction
Haloethers [11, 17]	G, Teflon-lined cap	Cool, ≤6 °C, store in dark	7 days until extraction 40 days after extraction
Chlorinated Hydrocarbons[11, 17]	G, Teflon-lined cap	Cool, ≤6 °C, store in dark	7 days until extraction 40 days after extraction
Acid Herbicides, Personal Care products & Urea Herbicides[17]	G, Teflon-lined cap	Cool, ≤6 °C, store in dark	7 days until extraction 40 days after extraction
Sucralose[17]	G, Teflon-lined cap	Cool, ≤6 °C, store in dark	21 days until extraction 40 days after extraction
Carbamates[17]	G, Teflon-lined cap	Cool, ≤6 °C, Monochloroacetic acid to pH 3, store in dark	7 days until extraction 21 days after extraction
Diquat and Paraquat[17]	P	Cool, ≤6 °C, Heptafluorobutyric acid to pH 3, store in dark	7 days until extraction 21 days after extraction
ERLN Agents[17]	G, Teflon-lined cap	Cool, ≤6 °C, store in dark	7 days until extraction
Pesticides[17]	G, Teflon-lined cap	Cool, ≤6 °C, store in dark	7 days until extraction 40 days after extraction
1,4-Dioxane[17]	G, Teflon-lined cap	Cool, ≤6 °C, Na_2SO_3, $NaHSO_4$, pH ≤4	28 days until extraction 28 days after extraction

Perfluorinated Alkyl Substances*[17]	P, Polypropylene liner in cap	Cool, ≤6 °C, store in dark	28 days until extraction 30 days after extraction

*Samples for *Perfluorinated Alkyl Substance analysis should not come in contact with any fluoropolymer such as Teflon or its analogues.*

Key to Notes for Table 5.6.1

1. High density polyethylene (P), glass (G), sterilized plastic (PA) such as polypropylene or other autoclavable plastic.

2. Sample preservation should be performed immediately upon sample collection. For composite chemical samples, each aliquot should be preserved at the time of collection. When use of an automated sampler makes it impossible to preserve each aliquot, then chemical samples may be preserved by maintaining at 4^{oC} until compositing and sample splitting is completed.

3. When any sample is to be shipped by common carrier or sent through the United States Mail, it must comply with the Department of Transportation Hazardous Materials Regulations (49 CFR Part 172). The person offering such material for transportation is responsible for ensuring such compliance. For the preservation requirements of this table, the Office of Hazardous Materials Transportation Bureau, Department of Transportation has determined that the Hazardous Materials Regulations do not apply to the following materials: Hydrochloric acid (HCl) in water solutions at concentrations of 0.04% by weight or less (pH about 1.96 or greater); Nitric acid (HNO3) in water solutions at concentration of 0.15% by weight or less (pH about 1.62 or greater); Sulfuric acid (H2SO4) in water solutions at concentrations of 0.35% by weight of less (pH about 1.15 or greater); Monochloroacetic acid buffer 1M in water solutions (pH 3) and Sodium hydroxide (NaOH) in water solutions at concentrations of 0.008% by weight or less (pH about 12.30 or less).

4. Samples should be analyzed as soon as possible after collection. The times listed are the maximum times that samples may be held before analysis and still be considered valid. Samples may be held for longer periods only if the permittee, or monitoring laboratory, has data on file to show that the specific types of samples under study are stable for the longer time, and has received a variance from the Regional Administrator under Part 136.3(e). Some samples may not be stable for the maximum holding time given in the table. A permittee or monitoring laboratory is obligated to hold the sample for a shorter time if knowledge exists which shows this is necessary to maintain sample stability. See Part 136.3(e) for details.

5. ASTM D7365–09a specifies treatment options for samples containing oxidants (e.g., chlorine). Also, Section 9060A of Standard Methods for the Examination of Water and Wastewater (20th and 21st editions) addresses dechlorination procedures for microbiological analyses.

6. Sampling, preservation and mitigating interferences in water samples for analysis of cyanide are described in ASTM D7365–09a. There may be interferences that are not mitigated by the analytical test methods or D7365–09a. Any technique for removal or suppression of interference may be employed, provided the laboratory demonstrates that it more accurately measures cyanide through quality control measures described in the analytical test method. Any removal or suppression technique not described in D7365–09a or the analytical test method must be documented along with supporting data.

7. For dissolved metals, samples should be filtered immediately on-site before adding preservative.

8. Guidance applies to sample to be analyzed by GC, LC, or GC/MS for specific compounds.

9. Sample receiving no pH adjustment must be analyzed within seven days of sampling.

10. The pH adjustment is not required if acrolein will not be measured. Samples for acrolein receiving no pH adjustment must be analyzed within 3 days of sampling.

11. When the extractable analytes of concern fall within a single chemical category, the specified preservative and maximum holding times should be observed for optimum safeguard of sample integrity. When the analytes of concern fall within two or more chemical categories, the sample may be preserved by cooling to ≤6 °C, reducing residual chlorine with 0.008% sodium thiosulfate, storing in the dark and adjustment the pH to 6-9; samples preserved in this manner may be held for seven days before extraction and for forty days after extraction. Exceptions to this optional preservation and holding time procedure are noted in footnote 5 (re the requirement for thiosulfate reduction of residual chlorine), and footnotes 12, 13 (re the analysis of benzidine).

12. If 1,2-diphenylhydrazine is likely to be present, adjust the pH of the sample to 4.0±0.2 to prevent rearrangement to benzidine.

13. Extracts may be stored up to 7 days before analysis if storage is conducted under an inert (oxidant-free) atmosphere.

14. For the analysis of diphenylnitrosamine, add 0.008% $Na_2S_2O_3$ and adjust pH to 7-10 with NaOH within 24 hours of sampling.

16. HCl that has been redistilled two to four times in a quartz sub-boiling still is highly recommended. Trace Metals Grade HCl satisfies this recommendation.

17. Aqueous samples must be preserved at ≤6 °C, and should not be frozen unless data demonstrating that sample freezing does not adversely impact sample integrity is maintained on file and accepted as valid by the regulatory authority. Also, for purposes of NPDES monitoring, the specification of "≤ °C" is used in place of the "4 °C" and "<4 °C" sample temperature requirements listed in some methods. It is not necessary to measure the sample temperature to three significant figures (1/100th of 1 degree); rather, three significant figures are specified so that rounding down to 6 °C may not be used to meet the ≤6 °C requirement. The preservation temperature does not apply to samples that are analyzed immediately (less than 15 minutes).

18. Place sufficient ice with the samples in the shipping container to ensure that ice is still present when the samples arrive at the laboratory. However, even if ice is present when the samples arrive, immediately measure the temperature of the samples and confirm that the preservation temperature maximum has not been exceeded. In the isolated cases where it can be documented that this holding temperature cannot be met, the permittee can be given the option of on-site testing or can request a variance. The request for a variance should include supportive data which show that the toxicity of the effluent samples is not reduced because of the increased holding temperature. Aqueous samples must not be frozen. Hand-delivered samples used on the day of collection do not need to be cooled to 0 to 6 °C prior to test initiation.

19. Sample analysis should begin as soon as possible after receipt; sample incubation must be started no later than 8 hours from time of collection.

20. For fecal coliform samples for sewage sludge (biosolids) only, the holding time is extended to 24 hours for the following sample types using either EPA Method 1680 (LTB-EC) or 1681 (A-1): Class A composted, Class B aerobically digested, and Class B anaerobically digested.

21. Collect samples in opaque bottles and process under reduced light. A secondary device, such as a Van Dorn/Niskin or bucket, may be used to collect the sample and then expeditiously transfer into an opaque bottle.

22. Samples must be filtered within 48 hours of collection. Add magnesium carbonate to the filter while the last of the sample passes through the filter.

Table 5.6.2
Sample Containers, Preservation Techniques and Hold Times
(Residuals, Soil and Sediment Samples [1])

Parameter Group	Methods	Container	Preservation	Max Holding Time
Volatile Organics	Purge-and trap, GC and GC-MS	Glass, 40 mL vial[4] or 4 oz wide-mouth jar with Teflon/silicone septum [2] 5g or 25g EnCore[4] Sampler	3 3	14 days[4] 7 days[4]
Semivolatile Organics (50 grams sample)	GC, HPLC and GC-MS	Glass, 8 oz wide-mouth with Teflon lined cap	3	14 days extraction 40 days analysis
Perfluorinated Alkyl Substances*	HPLC-MS MS	P, Polypropylene liner in cap, wide-mouth	3	28 days extraction 30 days analysis
Total Metals (except mercury and chromium VI)	ICP-AES & ICP-MS	Glass or plastic, 8 oz wide-mouth (200 grams sample)	3	6 months
Mercury	Manual Cold Vapor AA	Glass or plastic, 8 oz wide-mouth (200 grams sample)	3	28 days

**Perfluorinated Alkyl Substances should not come in contact with any fluoropolymer such as Teflon or its analogues.*

Key to Notes and Preservation:

1. Adapted from tables 3-1 and 4-1 in Test Methods for Evaluating Solid Waste, SW-846, EPA, Third Edition, 1986, and First Update in 1987. The term residuals includes:
   a. Concentrated waste samples
   b. Sludge of domestic or industrial origin

2. Sample should not be homogenized (mixed) prior to filling container. Container should be filled by packing as much sample into it leaving minimal headspace. Field sample cannot be composited for analysis. If TCLP is requested, collect sample in 8 oz. wide mouth jar with Teflon-lined cap.

3. Soils, sediments and sludges should be kept cool at 4^{O}C from collection time until analysis. No preservation is required for concentrated waste samples.

4. Soils, sediments and sludges preserved with sodium bisulfate should be kept cool at 4^{O}C from collection time or, if unpreserved, frozen to less than -10^{O}C within 48 hours after collection.

Table 5.6.3
Sample Containers, Preservation Techniques and Hold Times
(Biological Tissue [1])

Parameter Group	Methods	Container	Preservation	Max Holding Time
Semivolatile Organics (50 grams sample)	GC, HPLC and GC-MS	P, G, 4 oz wide-mouth (20 grams sample)	Freeze < -18°C	365 days extraction 40 days analysis
Perfluorinated Alkyl Substances (PFAS)[2]	HPLC-MS MS	HDPE or PP, 4 oz wide-mouth (20 grams sample)	Freeze < -18°C	365 days extraction 30 days analysis
Total Metals (except mercury and chromium VI)	ICP-AES & ICP-MS	P, G, 4 oz wide-mouth (20 grams sample)	Freeze < -18°C	365 days
Mercury[3]	Thermal decomposition and Cold-Vapor AA	P, G, 4 oz wide-mouth (20 grams sample)	Freeze < -18°C	365 days

[1] This preservation applies to fish and other soft tissue from any animal, including organs.

[2] Samples for *PFAS* analysis should not come in contact with any fluoropolymer such as Teflon or its analogues.

[3] Tissue samples of hair and feathers do not require preservation. They can be stored at room temperature in a glass or plastic container in a dark location.