Standard Guide for Preparation of Biological Samples for Inorganic Chemical Analysis¹

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1. Scope*

- 1.1 This guide describes procedures for the preparation of test samples collected from such locations as streams, rivers, ponds, lakes, estuaries, oceans, and toxicity tests and is applicable to such organisms as plankton, mollusks, fish, and plants.
- 1.2 The procedures are applicable to the determination of volatile, semivolatile, and nonvolatile inorganic constituents of biological materials. Analyses may be carried out or reported on either a dry or wet basis.
- 1.3 The values stated in SI units are to be regarded as standard. No other units of measurement are included in this standard.
- 1.4 This standard does not purport to address all of the safety concerns, if any, associated with its use. It is the responsibility of the user of this standard to establish appropriate safety and health practices and determine the applicability of regulatory limitations prior to use. For a specific hazard statement, see 9.3.3.

2. Referenced Documents

2.1 ASTM Standards:²

D1129 Terminology Relating to Water

D1193 Specification for Reagent Water

3. Terminology

3.1 *Definitions*—For definitions of terms used in this guide, refer to Terminology D1129.

4. Summary of Guide

4.1 Samples are collected, where possible, with nonmetallic or TFE-fluorocarbon-coated sampling equipment to prevent

¹ This guide is under the jurisdiction of ASTM Committee D19 on Water and is the direct responsibility of Subcommittee D19.05 on Inorganic Constituents in Water.

contamination, stored in plastic containers, and kept either at 4°C or frozen until returned to an adequate facility for analysis.

- 4.2 Before analysis, samples are allowed to return to room temperature. Large foreign objects are mechanically removed from the samples based upon visual examination; smaller foreign objects are also removed mechanically, with the aid of a low-power microscope.
- 4.3 Wet samples of small organisms such as plankton, are mixed for preliminary homogenization, then allowed to settle, to remove most of the occluded water. Larger organisms, such as fish, should be patted dry, using paper towels.
- 4.4 Where less than a whole organism is to be analyzed, tissue excisions are made with nonmetallic tools such as plastic knives or TFE-fluorocarbon-coated scalpels.
- 4.5 Moisture determinations are made on separate samples from those analyzed for volatile or semivolatile constituents.
- 4.6 Analyses for volatile constituents are made using wet samples from which supernatant liquid or occluded water has been removed (see 4.3). The results may be calculated to the dry, original-sample basis, using the results of a moisture determination carried out on a separate sample.
- 4.7 Analyses for semivolatile constituents are made on wet samples or samples previously dried at a temperature (dependent on constituents of interest), or using a procedure, found to be adequate for the purpose, and specified in the corresponding analytical procedure.
- 4.8 Analyses for nonvolatile constituents are made on samples previously dried at a temperature (dependent on constituents of interest), or using a procedure found to be adequate for the purpose, and specified in the corresponding analytical procedure.
- 4.9 Digest the samples according to the procedures outlined in Section 9.
- 4.10 A flow diagram outlining typical procedures is shown in Fig. 1.

5. Significance and Use

5.1 The chemical analysis of biological material, collected from such locations as streams, rivers, lakes, and oceans can

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² For referenced ASTM standards, visit the ASTM website, www.astm.org, or contact ASTM Customer Service at service@astm.org. For *Annual Book of ASTM Standards* volume information, refer to the standard's Document Summary page on the ASTM website.

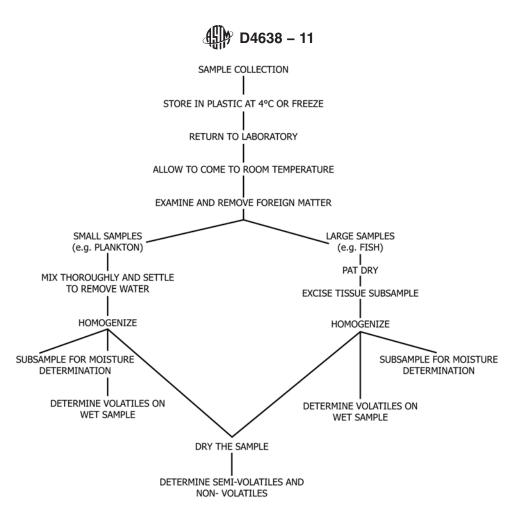


FIG. 1 Flow Diagram for the Preparation of Biological Samples for Inorganic Analysis

provide information of environmental significance. The chemical analysis of biological material used in toxicity tests may be useful to better interpret the toxicological results.

- 5.2 Many aquatic biological samples, either as a result of their size, or their method of collection, are inherently heterogeneous in that they may contain occluded water in varying and unpredictable amounts and may contain foreign objects or material (for example, sediment) not ordinarily intended for analysis, the inclusion of which would result in inaccurate analysis.
- 5.3 Standard methods for separating foreign objects, to facilitate homogenization, will minimize errors due to poor mixing and inclusion of extraneous material.
- 5.4 Standardized procedures for drying provide a means for reporting analytical values to a common dry weight basis, if desired. Analyses may also be carried out or reported on a wet weight basis.

6. Preliminary Treatment of Samples

- 6.1 Treat small heterogeneous samples, such as plankton, as follows:
 - 6.1.1 Allow for the sample to return to room temperature.
- 6.1.2 Remove foreign objects, such as leaves and twigs, mechanically, using nonmetallic instruments. Use a low-power microscope to facilitate removal of smaller foreign objects such as paint chips.

- 6.1.3 Transfer the sample to a beaker and thoroughly mix it with a glass stirring rod or equivalent, and allow it to settle so that most or all of the occluded water can be decanted.
- 6.1.4 If chemical analyses are to be carried out on a wet sample, and a large amount of material is available, remove a number of small portions (at least five) from random locations in the beaker, and composite them to obtain a representative sample of a size sufficient for chemical analysis and a separate moisture determination. Using a tissue disrupter, blender, or equivalent, homogenize the sample or composite (to ensure lack of contamination, carry a standard or blank, or both, through this procedure). Remove a subsample for moisture determination and proceed to Section 7. Retain the remainder and proceed to Section 9.
- 6.1.5 If chemical analyses are to be carried out on a dry sample, and a large amount of material is available, remove a number of small portions (at least five) from random locations in the beaker, and composite them to obtain a representative sample of a size sufficient for the analysis. Using a tissue disrupter, blender, or equivalent, homogenize the sample, or composite (to ensure lack of contamination, carry a standard or blank, or both, through this procedure), and proceed to Section 7.
 - 6.2 Treat large samples such as fish as follows:
 - 6.2.1 Allow the sample to return to room temperature.

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