



Standard Test Method for Analysis of Phytoplankton in Surface Water by the Sedgwick-Rafter Method¹

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1. Scope

1.1 This test method covers determining the density and taxonomic classification of phytoplankton. It is applicable both to relatively sparse or dense phytoplankton concentrations, provided the suspended-sediment concentration is low. The Sedgwick Rafter (S-R) method requires less costly apparatus than does the inverted microscope method but gives less accurate results. The inherent inaccuracy in the Sedgwick-Rafter method is due to the design of the counting chamber and cannot be circumvented by a different choice of optics. For this reason, the S-R method is limited to the use of objective lenses having a working distance of approximately 1.6 mm or more. With 10× oculars the maximum overall magnification is approximately 250×. High concentrations of suspended sediment can obscure the algal cells, and thus cause interference.

1.2 This test method is applicable to both freshwater and marine samples.

1.3 *This standard does not purport to address all of the safety problems, 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 specific precautionary information see Section 8.

2. Referenced Documents

2.1 ASTM Standards:²

D 1129 Terminology Relating to Water

D 1193 Specification for Reagent Water

D 3370 Practices for Sampling Water

D 4149 Classification for Sampling Phytoplankton in Surface Waters

2.2 Various taxonomic keys are required for identification of the algae. No single key is suitable for all species likely to be encountered. (See Greeson 1977; Weber 1973.)

¹ This test method is under the jurisdiction of ASTM Committee E47 on Biological Effects and Environmental Fate and is the direct responsibility of Subcommittee E47.01 on Aquatic Assessment and Toxicology.

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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.

3. Summary of Test Method

3.1 The microscope is calibrated to determine the field size on the superimposed ocular grid. A Sedgwick-Rafter chamber is filled with a preserved phytoplankton sample. After the algae settles to the bottom, the chamber is examined microscopically at 200 to 250× for the presence of algae. Those algal cells lying within the border of the ocular grid are identified and enumerated. The tally is used to calculate the algal density in cells per millilitre.

4. Significance and Use

4.1 Phytoplankton are basic to the food chain in all aquatic environments. In addition, they have long been considered to be important indicators of water-quality conditions. Phytoplankton data are also frequently used in the planning and design of water-treatment facilities and reservoirs.

5. Interferences

5.1 The presence of suspended sediment may obscure algal cells, making identification difficult. Colonial forms and the occurrence of algae in trichomes make the estimation of cell numbers difficult. Some preservation techniques may cause a loss of flagella, hampering identification.

6. Apparatus

6.1 *Microscope*, compound, with 10× oculars and 10×, 25×, 40×, and 90× objectives, substage condenser, and mechanical stage.

6.2 *Ocular Micrometer*, with Whipple grid.

6.3 *Sedgwick-Rafter Counting Cell*, 50 by 20 by 1 mm.

6.4 *Stage Micrometer*.

6.5 *Transfer Pipet*, 1-mL.

6.6 *Microscope Slides and Cover Glasses*, standard 76 by 25-mm noncorrosive slides. Cover glasses, round or square, clean and free of oil.

7. Reagents

7.1 *Purity of Reagent*—Reagent grade chemicals shall be used in all tests. Unless otherwise indicated, it is intended that all reagents shall conform to the specifications of the Committee on Analytical Reagents of the American Chemical Society,