

SOP for In Vitro Determination of Chlorophyll *a* Concentrations by Fluorescence

Isaac

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1 Scope and Application

- 1.1 This method provides a procedure for the fluorometric determination of chlorophyll *a* and its magnesium-free derivative, pheophytin *a* in marine and freshwater phytoplankton.
- 1.2 This method is modified from the US EPA Method 445.0 and APHA Standard Methods for the Examination of Water and Wastewater, 22nd Edition.

2 Summary of Method

- 2.1 Chlorophyll-containing phytoplankton in a measured volume of sample water are concentrated by filtering at low vacuum (13 cm Hg) through a glass fiber filter (Whatman GF/F). The pigments are extracted from the phytoplankton in 90% acetone and to ensure thorough extraction of chlorophyll *a*, are allowed to steep for at least 2hrs. The fluorescence of the sample is measured at the excitation wavelength of 485 nm and emission wavelengths 685 / 50 nm. Sample fluorescence is measured before and after acidification with 0.1M HCl to obtain a corrected chlorophyll *a* concentration.

3 Definitions

- 3.1 Stock Standard Solution (SSS) –
- 3.2 Laboratory Reagent Blank (LRB) –
- 3.3 Field duplicates –
- 3.4 Quality Control Sample (QCs) –

- 4 Interferences
- 5 Safety and Personnel Protective Equipment
- 6 Related Documents
- 7 Materials and Apparatus
- 8 Reagents and Standards
- 9 Collecting & Storing Samples
- 10 Procedure
- 11 Data Analysis and Calculations
- 12 QC/QA Criteria