

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

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July 29, 2016

## 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, 20<sup>th</sup> 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)

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### **3.2 Laboratory Reagent Blank (LRB)**

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### **3.3 Field duplicates**

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### **3.4 Quality Control Sample (QCs)**

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## **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**