A SELECTED BIBLIOGRAPHY OF CHLOROPHYLL
a DETERMINATION METHODOLOGY

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Introduction

The Marine Science Group at LBL is measuring chlorophyll a as part of the environmental assessment and measurement program of the Ocean Thermal Energy Conversion (OTEC) Program. Chlorophyll a is a green pigment which serves as a photocatalyst in photosynthesis. Marine environmental field studies typically utilize chlorophyll a concentrations as a measure of phytoplankton biomass. The ratio of total phytoplankton carbon (biomass) varies between 25 and 110 to 1 depending upon the class of phytoplankter and its state of nutrition (Fogg, 1975; Strickland, 1965; Subba Rao and Platt, 1969). As the knowledge of the chlorophyll a content is important to the understanding of the distribution of plant material in the ocean the methods used to gain this knowledge is also of importance.

Several accepted methods for the determination of chlorophyll a in seawater have been described: Holm-Hansen, 1965 & 1978; Strickland and Parsons, 1972; and Yentsch and Menzel, 1963. Tolstoy (1977) presents an excellent review of current methods of chlorophyll a determination as well as phytoplankton collection, sample storage, pigment extraction and their various sources of error. The spectrophotometric method described by Richards and Thompson (1952) has been widely accepted for analysis of waters rich in phytoplankton. An improved version of this method is outlined in Strickland and Parsons (1972). The fluorometric method, described by Yentsch and Menzel (1963), has the advantage of speed and greater sensitivity thereby requiring much smaller sample volumes although it is not as accurate or precise as the spectrophotometric method (Strickland and Parsons, 1972). The precision of the spectrophotometric method is ±5% at the 5 microgram level and, for the fluorometric method, better than ±8% of any value of chlorophyll a exceeding 0.5 microgram/liter (Strickland and Parsons, 1972).

Further discussion on fluorometric methodology, including light filter selection, is found in the Turner Designs paper. The physical/chemical characteristics, including the absorption spectra and excitation/emission spectra of chlorophyll a are discussed in Harris and Zscheile (1943), Parsons (1963), Richards (1952), Tolstoy (1977), Vernon and Seely (1966), White, Jones, Gibbs and Butler (1972) and Yentsch and Menzel (1963). Holm-Hansen and Riemann (1978) present some of the most recent improvements to chlorophyll a determination methodology including extraction solvent selection, sample storage and filter type selection.
A method for the continuous measurement of the in vivo chlorophyll a concentration is described by Lorenzen (1966). Studies on some of its problems and limitations are reported on by Cullen and Renser (1979), Kiefer (1973), Loftus and Seliger (1975), Maerker and Szekielda (1976) and Strickland (1968).

Phaeopigment, a degradation product of the different chlorophylls can interfere with the spectrophotometric or fluorometric determination of chlorophyll a because it absorbs light and fluoresces in the same region of the spectrum as chlorophyll a and if present, may cause errors in chlorophyll a values (Patterson and Parsons, 1963; Vernon, 1960). Therefore, when performing chlorophyll a determinations, measurements of phaeopigment must be included.

The following is a list of references compiled for researchers of chlorophyll a determination methodology. The references have been gathered from marine and freshwater research journals, texts and handbooks. The list is organized by aspects of analysis:

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Some articles overlap in coverage and appear in more than one category.

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References

Spectrophotometric Methods


ZSCHEILE, F.P., Jr. (1934) An improved method for the purification of chlorophylls a and b; quantitative measurement of their absorption spectra; evidence for the existence of a third component of chlorophyll. Botanical Gazette, 95, 529-562.

Fluorometric Methods


Filtration Methodology


Extraction Methodology


Pigments and Solvents Effects


SEELY, G. R., M.J. DUNCAN and W.E. VIDAVER (1972) Preparative and analytical extraction of pigments from brown algae with dimethyl sulphoxide. Marine Biology, 12, 184-188.


ZSCHIELE, F.P., Jr. (1934) An improved method for the purification of chlorophylls a and b; quantitative measurement of their absorption spectra; evidence for the existence of a third component of chlorophyll. Botanical Gazette, 95, 529-562.


Method Manuals and Surveys


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