# 5.18 Chlorophyll a of sampled water

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### (2) Objectives

We measured total chlorophyll a in seawater by using the fluorometric method.

### (3) Instruments and Methods

## (a) Reagents

Standard; chlorophyll a standard (SIGMA-ALDRICH Japan K.K.)

Extraction and dilution solutions; N,N-dimethylformamide

(Wako Pure chemical Industries, Ltd.)

Acidification reagent; 1.0 M HCl (Wako Pure chemical Industries, Ltd.)

## (b) Instruments

Spectorophotometer: UV-2400PC, manufactured by SHIMADZU CORPORATION

Fluorometer: 10-AU-005 manufactured by Turner Designs

Analytical condition was listed in table 5.18-1.

### (c) Method

Acidification method (Holm-Hansen et al., 1965)

### (4) Sampling

Following procedure is based on "Fluorometric determination of chlorophyll" (Holm-Hansen *et al.*, 1965). We collected samples from 10 - 12 depths between the surface and 500 m with bucket and Niskin bottles attached to the CTD-system.

Water samples were transferred to shading Nalgene bottles (ca.  $500 \text{ cm}^3$ ) from bucket and Niskin bottles. After sampling, water samples were vacuum-filtrated (<0.02MPa) through 25mm-diameter Whatman GF/F filter. Phytoplankton pigments retained on the filters were immediately extracted in a polypropylene tube with 7 ml of N,N-dimethylformamide. The tubes were stored at  $-20^{\circ}\text{C}$  under the dark condition to extract chlorophyll a for 24 hours or more.

#### (5) Standardization

The fluorometer was calibrated with a chlorophyll a standard in each cruise. The chlorophyll a standard concentration was determined by spectorophotometer. We prepared 9 dilutions from the chlorophyll a standard. Dilutions measuring with the fluorometer were taken before ( $F_o$ ) and after acidification ( $F_a$ ) with 2 drops 1.0 M HCl. We calculated linear calibration factor ( $K_x$ ) and the acidification coefficient ( $F_m$ ) from dilutions measurement data. The blank of DMF also measured with the fluorometer. The Blank value was subtracted from  $F_o$  and  $F_a$ .

### (6) Sample measurement

Following extraction, samples were removed from freezer in the dark room and the fluorometer was allowed to warm up and stabilize for 1 hour prior to measure. We measured Working Standard solution (ca. 20-30 ug/L) and DMF blank each 10 - 15 samples. Working Standard solution was measured for corrected  $K_x$  and  $F_m$ . All samples were measured on board.

## (7) Repeatability of sample measurement

During this cruise we measured Total chlorophyll *a* concentration in 2415 seawater samples at 211 casts. Replicate samples were taken at every CTD casts. The relative standard deviation of the replicate measurement was shown the table 5.18-2.

## (8) Preliminary Results

The result of total chlorophyll a was shown as the vertical distribution (Figure 5-18).

## (9) Data archive

All data will be submitted to JAMSTEC Data Management Office (DMO).

#### (10) Reference

- (1) Holm-Hansen, O., Lorenzen, C. J., Holmes, R.W., J. D. H. Strickland 1965. Fluorometric determination of chlorophyll. *J. Cons. Cons. Int. Explor. Mer.* 30, 3-15.
- (2) SHIMADZU CORPORATION 1996. UV-2400PC Instruction manual
- (3) TURNER Designs 1992. MODEL 10-AU-005 LABORATORY FIELD FLUOROMETER USER'S MANUAL

Table 5.18-1: Analytical conditions of "Acidification method" for chlorophyll *a* with Turner Designs fluorometer (10-AU-005)

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Acidification method		
Excitation filter (nm)	340-500	
Emission filter (nm)	>665	
Lamp	Daylight White	

Table 5.18-2: Repeatability of sample measurement

Leg	Leg1	Leg2
Number of replicate samples	97	114
R.S.D. (%)	4.8	3.2

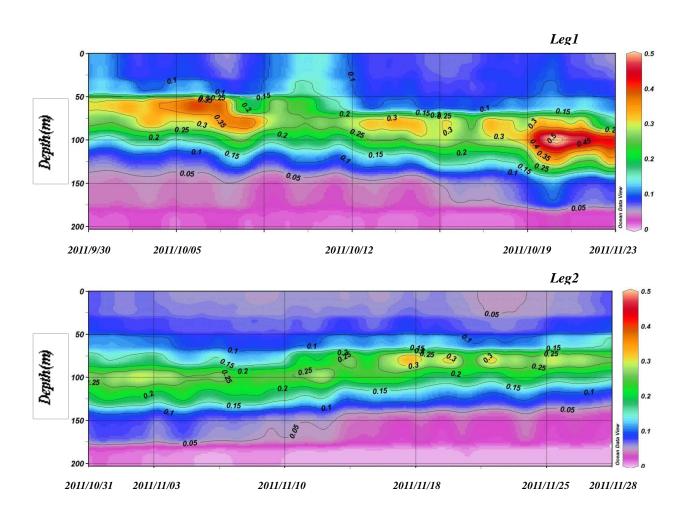


Figure 5.18-1: Vertical distribution of chlorophyll *a* concentration (µg/L) at Stn.8S in this cruise.