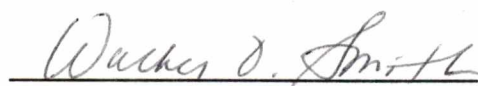


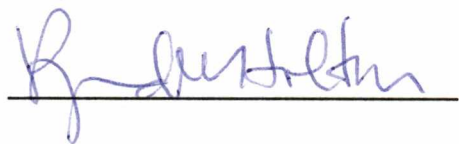
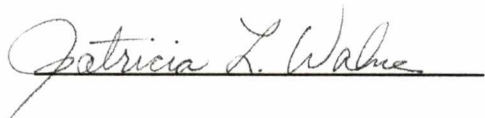
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I am submitting herewith a thesis written by Ying Hong entitled "Studies on Transparent Exopolymer Particles (TEP) Produced by *Phaeocystis* sp. In the Ross Sea." I have examined the final copy of this thesis for form and content and recommend that it be accepted in partial fulfillment of the requirements for the degree of Master of Science, with a major in Botany.

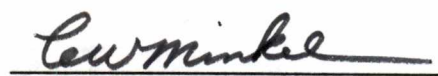


Walker O. Smith, Major Professor

We have read this thesis
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The Graduate School

STUDIES ON TRANSPARENT EXOPOLYMER PARTICLES (TEP) PRODUCED
BY *Phaeocystis* sp. (PRYMNESIOPHYCEAE) IN THE ROSS SEA,
ANTARCTICA

A Thesis
Presented for the
Master of Science
Degree
The University of Tennessee, Knoxville

Ying Hong

May 1996

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ABSTRACT

Transparent exopolymer particles (TEP) produced by *Phaeocystis* sp. (Prymnesiophyceae) were studied quantitatively in cultures and in the Ross Sea, Antarctica for the first time. Concentration of TEP produced by *Phaeocystis* were generally higher than those produced by diatoms, and were functions of growth rate and photosynthetic response, which were tightly coupled with irradiance and salinity. These two factors, therefore, influenced the production of TEP in culture. In the Ross Sea TEP concentrations were primarily a function of chlorophyll, and were minimal at chlorophyll levels below $3 \mu\text{g l}^{-1}$, but increased rapidly above this concentration. This two-state system indicated TEP formation might involve a coagulation mechanism, a possibility supported by a correlation between TEP concentrations and wind stress. Distributions of TEP were studied during a *Phaeocystis* bloom, and the different vertical distribution patterns between TEP and chlorophyll result in a maxima in the TEP: chlorophyll ratio in the bottom of the water column at most the stations investigated. TEP concentrations were measured repeatedly at the same station but at different times during the cruise, and higher TEP concentrations appeared in a deep layer in later surveys. This evidence supports the hypothesis that mucous flocs instead of *Phaeocystis* sp. colonies contribute significantly to carbon vertical flux because TEP is tightly related with flocs. A conceptual model of TEP related to the *Phaeocystis* sp. blooms and a carbon vertical cycle is suggested.

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NOMENCLATURE

α^B	Initial slope of P-I curves indicates the efficiency of photosynthesis in weak light
β^B	Slope at high irradiances indicates the strength of photoinhibition
C_{787}	Absorption of the blank at 787 nm
Chl	Chlorophyll a concentration
DOC	Dissolved organic carbon
E_{787}	Absorption of sample at 787 nm
F_o	Fluorescence before acidification
F_a	fluorescence after acidification
F_x	Calibration factor
I	Irradiance
K	Growth rate
M	Weight of the standard of xanthin
N	Number of hours of incubation
N_1	Number of cells at time one
N_2	Number of cells at time two
P	Photosynthetic rate
P^B	Photosynthetic rate per unit chlorophyll
P_m^B	Light-saturated Chl-specific rate of photosynthesis
P_s^B	Maximum photosynthetic rate in the absence of photoinhibition
P_{max}	Maximum photosynthetic rate
R	Total activity (dpm) of bicarbonate

R_b	Dark bottle count (dpm) corrected for quenching
R_s	Sample counts (dpm) corrected for quenching
S	Salinity
TEP	Transparent exopolymer particles
V	Volume filtered of the samples
W	Weight of the total carbon dioxide

CHAPTER I

INTRODUCTION

Phaeocystis is a prymnesiophycean alga, important in polar waters. Its blooms have been reported in the Greenland Sea (Smith et al., 1991), the Weddell Sea (Buck and Garrison, 1983), and the Ross Sea (El-Sayed et al., 1983; SooHoo et al., 1987). This organism is characterized by two different morphological stages: unicellular and colonial. The former with free-living cells of 3 to 8 μm in length, is either flagellated or nonmotile. The latter are composed of cells without flagella, embedded in a mucilaginous matrix composed mainly of polysaccharide (Weisse and Scheffel-Moser, 1990). Colonial *Phaeocystis* blooms are important components of polar carbon cycles because they are rich in mucilaginous polysaccharide, which can support an active "microbial loop" (Guillard and Hellebust, 1971; Lancelot et al., 1987; Smith et al., 1991; Davidson and Marchant, 1992). It has been estimated that more than 50% of *Phaeocystis* biomass consists of mucilaginous polysaccharide (Lancelot et al., 1987). When large colonies dominate the bloom, mucilaginous polysaccharide comprises as much as 85% of the total particulate matter (Rousseau et al., 1990). This phytoplankton also has been found to exude dissolved organic substances at high rates under some conditions (Guillard and Hellebust, 1971; Eberlein et al., 1985). Guillard and Hellebust (1971) found that *P. pouchetii* in its colonial phase excreted 15-64% of its photossimilated carbon, and release rate equal to 2-18% of recently fixed carbon have also been reported (Eberlein et al.,

1985; Verity et al., 1988). The differing reports on the percentage of photosynthetic carbon liberated may be due to different growth conditions. For example, lower light intensity was considered as a possible factor causing a decrease of dissolved organic carbohydrate (DOC) liberated (Guillard et al., 1971; Verity et al., 1988). In addition, nutrient-stressed cells were also hypothesized to excrete more organic carbon than nutrient-replete cells (Verity et al., 1988). Because of the massive concentrations of organic carbon formed during *Phaeocystis* blooms, models of carbon flux of polar systems clearly must include the multiple effects of this species.

Vertical fluxes of *Phaeocystis* blooms could be mediated by grazing (Smith et al., 1991; Granéli et al., 1993; Hansen et al., 1993; Passow and Williamson, 1994). Although colonies are available to specific grazers (Granéli et al., 1993; Hansen et al., 1994), most *Phaeocystis* sp. blooms are not directly grazed by zooplankton and thus escape this trophic transformation (Alldredge and Silver, 1988; Hansen et al., 1993; Crocker and Passow, 1995). Ungrazed *Phaeocystis* colonies, the mucoid sheath, and the dissolved organic substances they produce can be degraded by bacteria. The efficiency of this microbial pathway, however, is uncertain (Lancelot et al., 1987; Rousseau et al., 1990; Davidson and Marchant, 1992). Some studies have shown that heterotrophic activity does not completely utilize the DOC pool produced during *Phaeocystis* blooms in polar waters (Eberlein et al., 1985; Davidson and Marchant, 1992; Passow and Williamson, 1994; Carlson et al., unpubl.). Davidson and Marchant

(1992) reported that the high concentrations of dissolved organic substances accumulated during a bloom of *Phaeocystis pouchetii* were apparently not significantly used by bacteria, because the numbers and activity were low at that time, a finding similar to that found in the Ross Sea (Carlson et al., unpubl.). Therefore, the question arises as to how does the process transform DOC into particles and contribute to the carbon cycle during *Phaeocystis* blooms in polar regions.

A number of studies have suggested that sedimentation of marine snow associated with algal blooms contributes significantly to vertical fluxes of carbon (Alldredge and Silver, 1988; Wassmann et al., 1990; Alldredge et al., 1993; Riebesell, 1993 ; Kiørboe and Hansen, 1993; Passow and Alldredge, 1994; Passow and Williamson, 1994; Wassmann 1994; Logan et al., 1995). Wassmann et al. (1990) observed massive sedimentation of *Phaeocystis* in the Barents Sea and attributed the flux at the end of the bloom to the formation of rapidly sinking marine snow. It has been commonly assumed that nature of *Phaeocystis* colonies and their extracellular polysaccharide mucus would be sticky, and that this sticky nature would enhance the formation of aggregates. However, recent evidence suggests that senescent colonies do not have higher stickiness coefficients than actively growing ones (Passow and Wassmann, 1994). However, mucilage of *Phaeocystis* and particulate polysaccharide from sediment traps collected during *Phaeocystis* blooms were suggested to promote the formation of large mucous flocs (Passow and Williamson, 1994). The

extracellular carbohydrate material of aged *Phaeocystis* gives rise to adhesive webs and sticky capsular secretions which are easily colonized by bacteria (Passow and Alldredge, 1994). These materials enhance the formation of macroaggregates (marine snow), and deplete particulate and dissolved organic matter from surface water (Riebesell, 1993; Passow and Alldredge, 1994; Passow and Williamson, 1994; Wassmann, 1994). Therefore, the role of extracellular polysaccharide in the process of aggregation during *Phaeocystis* blooms awaits further study in the laboratory and field to understand its importance in the cycling and fluxes of carbon in polar oceans.

Recently, a new class of particles, called transparent exopolymer particles (TEP), has been found to be important in the formation of aggregates associated with diatom blooms. The numbers of TEP increased by an order of magnitude, and the percentage of diatoms attached to TEP increased from 3% to more than 10% during a diatom bloom (Passow and Alldredge, 1994). These discrete particles are produced from dissolved carbohydrate polymers exuded by phytoplankton. They may play an important role in the vertical flux of carbon during *Phaeocystis* blooms, since *Phaeocystis* produces more carbohydrate than most other phytoplankton (Guillard and Hellebust, 1971; Eberlein et al., 1985; Lancelot and Mathot, 1985; Verity et al., 1987; Weisse Scheffel-Moeser, 1990; Davidson and Marchant, 1992; Passow and Williamson, 1994; Obernosterer and Herndl, 1995). However, studies on the TEP produced by *Phaeocystis* are limited. Because of the trophic role of *Phaeocystis* and its importance in polar

waters, as well as the potential role of TEP in aggregate formation and subsequent vertical flux, it is important to study the abundance, and distribution of TEP production during *Phaeocystis* blooms in order to understand its significance.

Studies on TEP produced by phytoplankton

TEP has been observed in natural waters and in algal cultures (Alldredge et al., 1993). They are the same types of particles observed by Emery et al. (1984) and Gordon (1970), and can be made visible by staining with alcian blue (copper phthalocyanine with 4 methene-tetraethyl-cis-o-thiouronium-chloride side chains). After staining, TEP can be enumerated microscopically. However, this method is labor intensive, especially since stained particles do not have high enough contrast to use image analysis systems to enumerate the particles. As a result a simple quantitative method was developed to determine the concentrations of TEP spectrophotometrically (Passow and Alldredge, in press), and the quantity of TEP in seawater has been found to vary by four orders of magnitude using this method.

TEP originates from the dissolved polysaccharide released by phytoplankton (Passow and Alldredge, 1994). In seawater these polymers align and coalesce via cation bridging, forming particles ranging from submicrons to several hundred microns in size. On the filters these particles are discrete, highly deformable discs or strings, containing no or few visible inclusions, whereas others are sheets or ropes of stainable carbohydrates imbedded with

inclusions and detritus. Because TEP exist as discrete particles, they provide a means by which dissolved organic material can be transformed to particulate matter, comprise the matrix of marine snow, and serve as a substrate and microhabitat for attached bacteria (Aldredge et al., 1993; Wassmann, 1994; Logan et al., 1995). It is assumed that the properties of TEP generated by different species vary (Kjørboe and Hansen, 1993; Crocker and Passow, 1995; Logan et al., 1995). They differ considerably in size, shape and stickiness between species. During a bloom dominated by dinoflagellates in Monterey Bay, TEP were relatively small (8 to 10 μm), whereas during a diatom bloom most TEP were more than 100 μm in length (Aldredge et al., 1993). TEP produced by some species do not stick to cells or cause flocculation; on the other hand, some algae produce TEP with high stickiness coefficients (up to four orders of magnitude higher than those of the living cells; Crocker and Passow, 1995). This type of TEP can stick to cells and cause phytoplankton to aggregate (Crocker and Passow, 1995; Logan et al., 1995). TEP are also produced at different growing stages (Eberlein et al., 1990). Decho (1990) reported two different classes of mucus secreted by algae. One type is released during their exponential growth, whereas another is liberated during stationary growth. The latter are looser and may precipitate, giving rise to the particulate mucus similar to that described for *Phaeocystis*. Senescent *Phaeocystis* colonies disintegrate and the fragments of the discrete colonies stained blue, and as such were considered to be TEP (Passow and Aldredge, 1994). Although production of

TEP may link several processes in the ocean, there is very little information available to describe the interrelationships among the particulate classes. In order to understand the role of *Phaeocystis* in the vertical transport of the carbon in the Ross Sea, the following points need to be understood: 1) TEP concentrations, distributions and dynamics during a *Phaeocystis* bloom; 2) the ability of *Phaeocystis* to produce these particles; and 3) the factors that affect the production of TEP in a *Phaeocystis* bloom.

The influence of salinity and irradiance on polar phytoplankton

Some environmental factors influence photosynthetic performance (Verity et al., 1988; Peperzak, 1993; Cota et al., 1994; Kirst and Wiencke, 1995). Under long-term hyperosmotic conditions, algae accumulate organic material (Stefels and Van Boekel, 1993). Light dependent excretion has also been observed in a temperate strain of *Phaeocystis* (Guillard and Hellebust, 1971; Verity et al., 1988). Because TEP are part of the organic matter photosynthetically fixed by phytoplankton, their production could be impacted by salinity and irradiance. These two parameters are also factors that can affect the growth rate of *Phaeocystis*, but their influence is inadequately studied. To obtain an accurate estimate of the impacts of these factors on TEP production, the influence of light and salinity on the growth of *Phaeocystis* must be described.

Irradiance has been considered as one of the limiting factors on the growth rate of phytoplankton in polar waters (Smith et al., 1991; Cota et al., 1994; Figueiras et al., 1994). Previous investigations on the relationship of

irradiance and *Phaeocystis* were focused mostly on the short-term process of photosynthetic production. It has been observed that *Phaeocystis* can incorporate inorganic carbon at relatively low irradiances, while at high irradiances photoinhibition is less severe than in diatoms (Lancelot and Mathot, 1987) or even absent (Verity et al., 1988). It also has been shown that the proportion of carbohydrates produced by colonial cells increases with irradiance (Guillard and Hellebust, 1971; Lancelot and Mathot, 1985; Veldhuis et al., 1986; Veldhuis, 1987), and *Phaeocystis* seems able to adapt to wide ranges of irradiance (Davidson and Marchant, 1992). In addition, the rate of photosynthetic production versus irradiance is different from the measured growth rate as a function of daily irradiance, since the two processes occur on fundamentally different time scales (Peperzak, 1993). Therefore, I conducted long- and short-term experiments to test the effects of irradiance on the growth of *Phaeocystis* and on TEP production.

In addition to irradiance, salinity varies seasonally in the Southern Ocean. For the most part ice algae grow in 34 ppt salinity, which is the salinity of the underlying water column. However, during brine drainage salinity may exceed 50 ppt. On the other hand, salinities as low as 20 ppt may be encountered at the ice edge during melting. Hence salinity potentially may influence growth of algae in polar waters. The influence of salinity on algal blooms has been suggested by some studies (Grant and Horner, 1976; Bates and Cota, 1986; Poplin et al., 1986; Arrigo and Sullivan, 1992). However, investigations on the ranges of salinity in

which ice algae can grow are paradoxical. Grant and Horner (1976) and Arrigo and Sullivan (1992) observed that ice algal growth occurred over a broad range of salinity (5-60 ppt), while other results showed that growth was restricted to a narrow range of salinities (19.5 to 31.5 ppt: Bates and Cota, 1986). According to these results, the long-term growth response seems to show a broader salinity range than does the short-term photosynthetic response. For example, Arrigo and Sullivan (1992) found a wider salinity tolerance in ice microalgae than did Bates and Cota (1986). The relatively long time period of acclimation (two days rather than 30 min.) was offered as one explanation, although it also may have been due to variable species composition. Poplin et al. (1986) have studied the relationship between species composition and environmental factors by discriminant analysis, and they showed that the response to salinity was species-dependent. The influence of salinity on the growth of *Phaeocystis* has not been investigated, and therefore its role is far from clear. One purpose of this study is to estimate the impact of salinity on *Phaeocystis* growth and the production of TEP .

Finally, it is necessary to study the response of *Phaeocystis* to simultaneous changes in salinity and irradiance because the algae may be exposed to specific light-salinity combinations under certain temperatures. The possible synergistic effect between irradiance and salinity will be studied so that the interaction between the two can be evaluated.

Objectives

Although a number of studies have suggested that TEP have important roles in marine systems, the distribution, abundance and characteristics of this new class of particles and the relationship to *Phaeocystis* in polar waters remain largely unknown. *Phaeocystis* blooms are common features of many polar waters and may play an important role in the vertical transport of carbon. In order to understand this carbon flux, the TEP produced by this species must be quantitatively assessed. This study combines the results from the laboratory experiments and field investigations. Specifically, the distribution and concentration of TEP in the Ross Sea were investigated during a *Phaeocystis* bloom, and its correlation with environmental parameters was determined. The capability of *Phaeocystis* sp. to produce TEP at different irradiances and salinities was studied in the laboratory to corroborate the field investigation. Therefore, the objectives of this study were as follows:

1. To examine the abundance of TEP produced by *Phaeocystis* sp. during austral spring in the Ross Sea, Antarctica;
2. To determine the capacity of *Phaeocystis* to produce TEP at different irradiances and salinities;
3. To examine the factors that influence the TEP abundance during *Phaeocystis* blooms;
4. To examine the correlation between TEP concentrations and *Phaeocystis* biomass during blooms; and

5. To investigate whether the vertical distribution of TEP is related to that of *Phaeocystis* sp.

CHAPTER II

MATERIALS AND METHODS

Phaeocystis sp Cultures

A culture of *Phaeocystis* sp. (Prymnesiophyceae) was obtained from the Center for Culture of Marine Phytoplankton, Bigelow Laboratory for Ocean Sciences. This strain was collected from McMurdo Station, Antarctica and maintained in f/2 medium. Before the experiments, the cultures were incubated at 4°C under continuous light. Light was provided from cool white fluorescent tubes and light levels ranged from 10 $\mu\text{E m}^{-2} \text{s}^{-1}$ to 100 $\mu\text{E m}^{-2} \text{s}^{-1}$. One liter batch cultures were allowed to adapt to the experimental salinity for two days prior to use.

Sample Collection

Field studies were conducted in the Ross Sea, Antarctica, between November 11 and December 8, 1994 on the *R.V Nathaniel B. Palmer*. This study area, located between 73°30' and 76°30'S and 168°30'E and 175°19'W, included a total of 21 stations along three transects at which TEP concentrations were measured. The transects can be defined by stations at which TEP samples were collected (Figure 1):

Transect 1: Stations 7,9,18,19,60,70,77;

Transect 2: Stations 22,37,77,70,31,47,25,23; and

Transect 3: Stations 22,47,45,39

At each station water samples were collected from depths ranging from 0

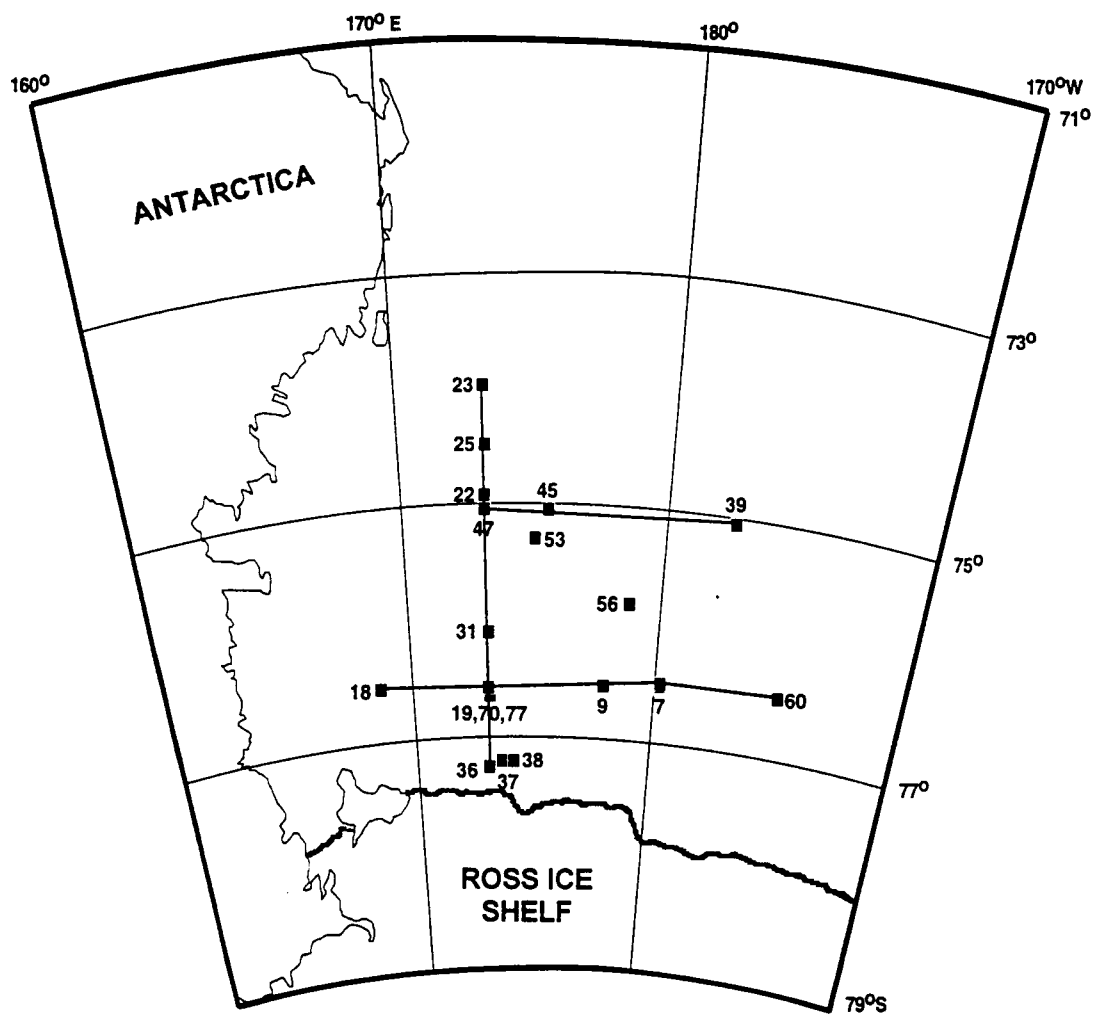


Fig. 1 Map showing location of stations sampled during field study

to 150 m using a rosette system (General Oceanics) fitted with 24 10-liter Niskin bottles. Continuous profiles of salinity, temperature and irradiance were obtained by the use of a Seabird CTD system and a BioSpherical instruments quantum meter attached to the rosette. Nutrient concentrations (ammonium, nitrate, nitrite, phosphate and silicate) were quantified on samples taken from these casts using standard autoanalyzer techniques (Friederich and Whitledge, 1972).

Concentrations of chlorophyll a were determined fluorometrically by filtering seawater samples (100-300 ml) through Gelman GF/F 25 mm glass fiber filters (nominal pore size of $0.7\mu\text{m}$) and extracting in 90% acetone. Samples were sonicated for 15 minutes on ice, extracted for at least 10 minutes more, and the resultant fluorescence was measured before and after acidification (3 drops of 5% HCl) on a Turner Designs Model 10 fluorometer calibrated with a purified chlorophyll a (Sigma) dissolved in 90% acetone.

Measurement of growth rates

It is easy to use *in vivo* fluorescence of cells to measure algal growth, but the result of this measurement has no absolute units. In this study, parallel experiments were conducted to measure the increase in cell numbers, chlorophyll a concentrations and *in vivo* fluorescence of cells through time, so that the relationships among these three variables could be established by regression analysis.

To estimate growth rates, *Phaeocystis* sp. colonies were cultured at 4°C and 12:12 LD for 14 days. At two-day intervals triplicate 10 ml samples were

collected for the measurement of *in vivo* fluorescence of cells. The same samples were filtered through GF/F filters and the chlorophyll a of the filters was extracted into 90% acetone and placed in a refrigerator for 24 hr. The fluorescence of chlorophyll a was measured and calculated according to equation 1:

$$\text{Chl a } (\mu\text{g l}^{-1}) = K (F_o - F_a) V_E (\text{scale } V_F) \quad (1)$$

where K equaled 153.7 (determined from standard chlorophyll solution for that particular fluorometer), F_o is the fluorescence before acidification, F_a is the fluorescence after acidification, V_E is the volume of 90% acetone used, and V_F is the sample volume filtered.

Cell numbers were determined by Utermohl technique (Smayda, 1978). Prior to sampling, the cultures were shaken vigorously, and 5 ml of the cultures were placed in 20 ml vials and fixed with 0.1 ml formalin (35%) solution. For the purpose of disrupting the colonies, the solution was acidified with 0.2 ml HCl and shaken on a vibromix, after which cell numbers were quantified in a Petroff-Hausser bacteria counting chamber. The growth rate was calculated using the formula (Smayda, 1978) :

$$K = \text{Log}(N_1/N_2)/(3.322/t) \quad (2)$$

where K is the growth rate (division per day) and N is number of cells. Subscripts denote numbers of cells at two time periods and denote the day of sampling. The relationship between *in vivo* and chlorophyll a fluorescence, as well *in vivo* fluorescence and cell numbers, was determined by linear regression.

Studying the Photosynthetic Response of *Phaeocystis*

Individual photosynthesis vs irradiance experiments were conducted using *Phaeocystis* cultures. *Phaeocystis* was incubated in a small volume photosynthetron (Lewis and Smith, 1983), which is a light chamber with at least 25 irradiance levels and cooler system. Additional replicate experiments with 28 and 36 ppt salinities were performed to test for potential synergistic effects between light and salinity. All treatments were incubated for two hours at 4°C after being preconditioned to each salinity for two days.

After the pretreatment, cultures were thoroughly but gently mixed, and 40 ml were removed and placed in a beaker which was kept on ice and in the dark. *Phaeocystis* samples then were injected with 40 μ l of 1.03 mCi ml⁻¹ ¹⁴C-bicarbonate. After mixing again, 32 aliquots of 1 ml algal suspensions were removed and placed into separate 20-ml glass scintillation vials. These are the treatment subsamples. An additional four aliquots of 1 ml of samples were treated identically. Three were used for time-zero controls (R) and the other for dark ¹⁴C-uptake correction. All treatment subsamples (including the dark correction) were incubated in a photosynthetron at irradiances ranging from 0 to 400 μ E m⁻² s⁻¹ for 2 h at 4°C. The incubations were stopped by adding 100 μ l 6N HCl, and the samples were dried at 60°C. Ten ml of scintillation cocktail (Aquasol) were added to dried samples after they were rehydrated in 1 ml deionized water, and the radioactivities of samples were counted by liquid

scintillation techniques.

The photosynthetic rate (P) was estimated by equation 3:

$$P \text{ (mg C/m}^3\text{ h}^{-1}\text{)} = (1.05W(R_s - R_b)) / (RN) \quad (3)$$

where R is total activity (dpm) of bicarbonate added, N is the number of hours of incubation, 1.05 is an isotope discrimination factor, R_s is the sample counts (dpm) corrected for quenching, R_b is the dark bottle count (dpm) corrected for quenching and W is the weight of the total carbon dioxide present in mg C m^{-3} at time zero, as determined by equation 4:

$$W = 12,000 T_o \quad (4)$$

where $T_o = 0.067S$, and S is salinity (ppt) of seawater or culture medium. The photosynthetic parameters were fitted to the equations of Platt et al. (1982):

$$P^B = P_s^b (1 - e^{-a})e^{-b} \quad (5)$$

where P^B is the photosynthetic rate per unit chlorophyll, a is $\alpha^B I / P_s^B$, and b is $\beta^B I / P_s^B$. The initial slope α^B is an indicator of the efficiency of photosynthesis in weak light (the quantum yield), P_s^b is the maximum photosynthetic rate in the absence of photoinhibition, and β^B is the slope at high irradiances which describes the strength of photoinhibition. The superscript B indicates that all parameters are normalized to biomass.

Measurement of TEP Concentration

Experiments were conducted either with natural seawater or with batch cultures of *Phaeocystis* at different growth stages and at various salinity and irradiance conditions. TEP concentrations were determined

spectrophotometrically based on the methods of Passow and Alldredge (1995). From 5-10 ml of cultures and 50 - 100 ml of sea water (field samples) were filtered through a 100 μm nylon net, and then the supernatant was passed through 0.4 μm Nuclepore filters under low ($<1/3$ atm) vacuum pressure. TEP were stained on damp filters with an aqueous solution of 0.02% alcian blue (Sigma) dissolved in 0.06% acetic acid. The staining solution was prefiltered through a 0.2 μm filter before use, as the stain aggregates within itself. The filters were fully covered with stain (500 μl) for about 2 seconds, and then were rinsed twice with distilled water. Field samples were frozen in liquid nitrogen until they could be analyzed in the laboratory. The concentration of TEP was quantified by analyzing each filter after soaking for 2-3 h in 5 ml of 80% H_2SO_4 (the filters in beakers were agitated gently 2-3 times per hour). The absorption maximum of the solution lies at 787 nm and absorption was measured in a 1-cm cuvette against 80% H_2SO_4 as a reference. TEP concentrations were expressed as xanthan equivalents ($\mu\text{g l}^{-1}$) and were estimated from equation 6:

$$\text{TEP} = (E_{787} - C_{787}) V_f^{-1} F_x \quad (6)$$

where E_{787} is the absorption of the sample, C_{787} is the absorption of the blank, which was prepared with sea water and from culture medium. V_f is the volume filtered in liters, and F_x is the calibration factor which is obtained from the standard preparation procedure (see below).

A standard was prepared by mixing about 15 mg of gum xanthin into 200 ml distilled water for 30 minutes. The solution was then ground with a tissue

grinder and the dry weight of the standard solution was determined by filtering 3-5 ml aliquots onto pre-weighed 0.2 μ m Nuclepore filters. Alcian blue stainable particles were measured colorimetrically by filtering 3-5 ml of standard solution as described above. The calibration factor (F_x) was calculated according to equation 7:

$$F_x = M[(Est_{787} - C_{787}) V_{st}^{-1}]^{-1} \quad (7)$$

where M is the weight of the standard (μ g l⁻¹ gum xanthin), Est_{787} is average absorption, C_{787} is the absorption of the blank, and V_{st} is the volume filtered of the standard in liters.

The Influence of Salinity and Irradiance on the Growth of *Phaeocystis* and on TEP Production

To estimate growth rates and TEP concentrations at different salinities and irradiances, *Phaeocystis* cells were cultured at three irradiances (100, 50 and 15 μ E m⁻² s⁻¹), with each including three salinity treatments (28, 32 and 36 ppt). Attenuation was provided by appropriate layers of neutral density screen. At two day intervals, triplicate 10 ml samples were collected to measure *in vivo* fluorescence. The same samples were used to measure TEP concentrations spectrophotometrically, and the TEP concentrations were expressed on a per μ g of chlorophyll a basis.

The influence of irradiance and salinity on TEP production was also determined by short-term experiments. After *Phaeocystis* cells were cultured to stationary stage in 32 ppt salinity, *in vivo* fluorescence was measured to estimate

the chlorophyll a concentration. At the same time, 32 aliquots of 1 ml algal suspensions from culture were removed and placed into separate 20-ml glass scintillation vials. All subsamples including the dark corrections were incubated in a photosynthetron at irradiances ranging from 0 to 400 $\mu\text{E m}^{-2} \text{ s}^{-1}$ for 4 h at 4°C. Additional replicate experiments with samples grown at 36 ppt salinity were also conducted. TEP concentrations were determined spectrophotometrically, and TEP concentration per μg chlorophyll a in each treatment was calculated.

Statistical Analysis

In order to determine whether salinity and irradiance affect production of TEP independently or synergistically, a 2-way analysis of variance was performed by the SAS program. The probability level for a type I error was set *a priori* at 0.05. If a significant difference was tested among treatments, contrast analysis was used to compare the significant difference between each group. In order to determine whether there were trends in the observed TEP concentrations that could be related to trends in the environmental parameters, correlation analysis and a stepwise multiple regression for the dependent variable TEP was performed by using SPSS 6.1.

CHAPTER III

RESULTS

In this investigation, it was hypothesized that *Phaeocystis* colonies have a high potential to produce TEP at stationary stage, and that factors influencing growth rate will influence TEP production. In order to test this hypothesis, the change of TEP concentrations through time was measured and the influences of salinity and irradiance on the growth rate and TEP production were tested simultaneously.

Influences of Salinity and Irradiance on *Phaeocystis* Growth Rate

In vivo fluorescence of cells was used to measure algal biomass and growth rate to examine *Phaeocystis* growth rate. Linear relationships between *in vivo* fluorescence, cell number, and chlorophyll a concentrations were found (Figures 2 and 3; $R^2 = 0.93$ between cell numbers and *in vivo* fluorescence, and $r^2 = 0.99$ between chlorophyll a and *in vivo* fluorescence). The growth rates (K) of *Phaeocystis* measured after three days were used in two-way ANOVA analysis to estimate the influences of salinity and irradiance (Table 1 and Appendix A). The results of this analysis are shown in Table 2. Both salinity and irradiance impacted the growth rate significantly (P values were less than 0.01), and the interaction between these two factors was significant (P=0.022). Contrast analysis showed that there was a clear difference between the 15 and 100 $\mu\text{E m}^{-2} \text{s}^{-1}$ irradiance group, as well as between the 50 and 100 $\mu\text{E m}^{-2} \text{s}^{-1}$ group. However, the difference between 15 and 50 $\mu\text{E m}^{-2} \text{s}^{-1}$ group was not

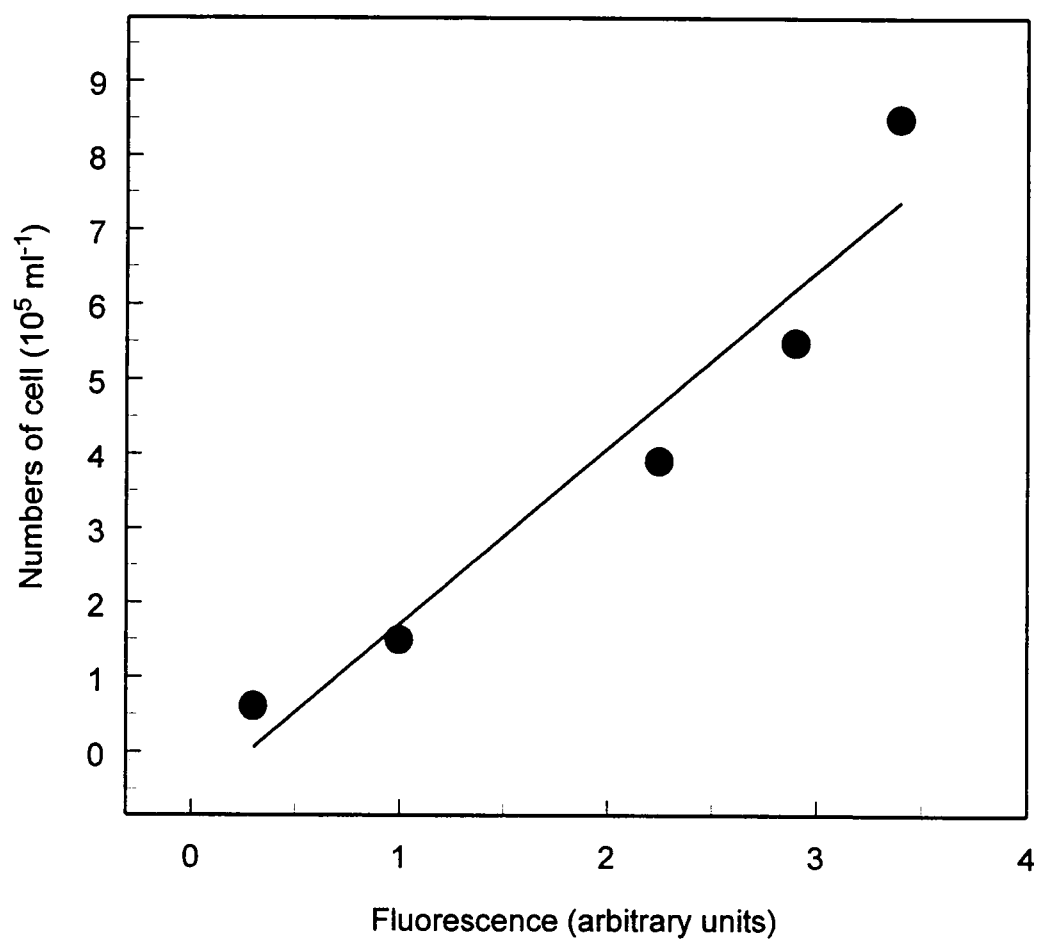


Figure 2. Relationship between *in vivo* fluorescence and cell numbers
($R^2=0.93$ and cell numbers= $2.361 \times \text{fluorescence} - 0.6528$)

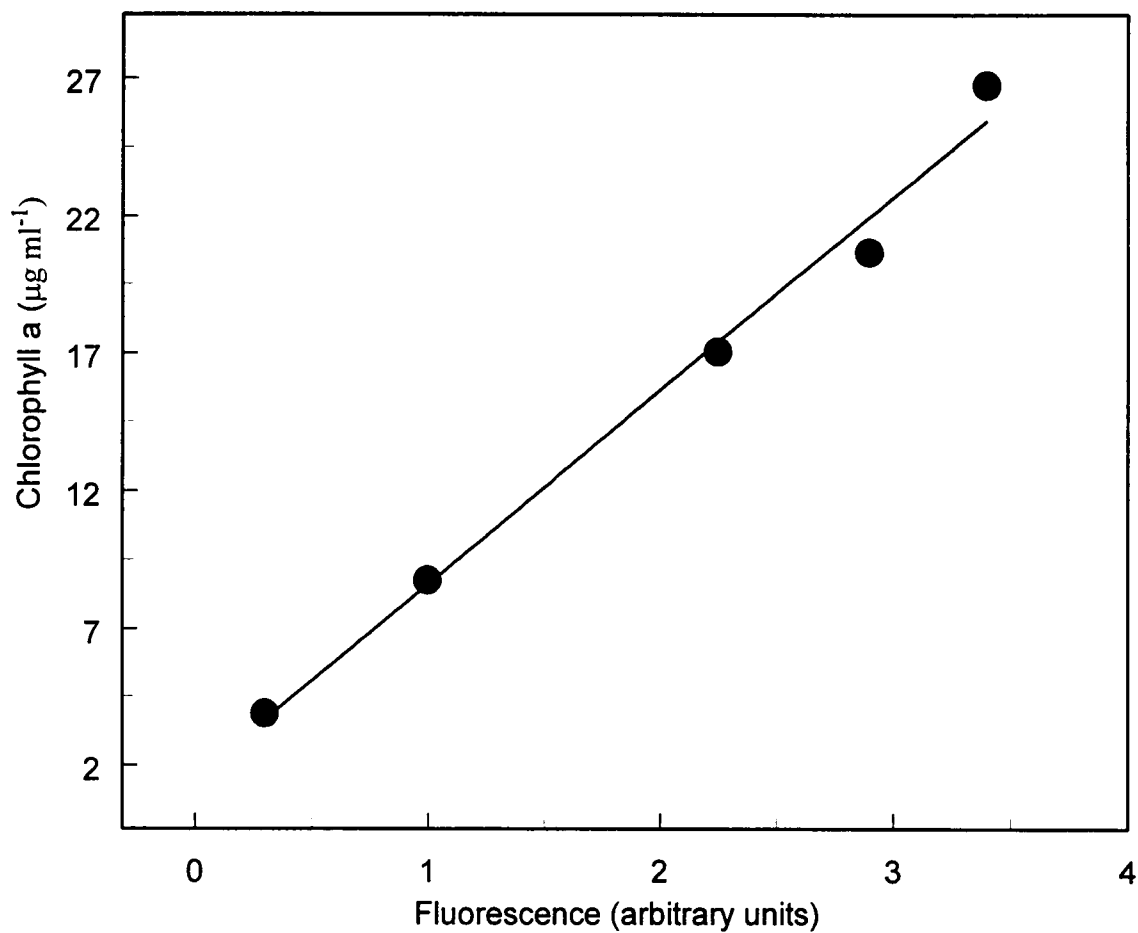


Figure 3. Relationship between *in vivo* fluorescence and Chlorophyll a (Chlorophyll a = $7.033 \times \text{fluorescence} + 1.561$, and $R^2 = 0.99$)

Table 1. Growth rates (K) of *Phaeocystis* sp. cultured at different salinities and irradiances

Time (hours)	Irradiance ($\mu\text{E m}^{-2} \text{s}^{-1}$)	Salinity (ppt)	K (division d ⁻¹)
60	15	28	1.10
60	15	32	1.10
60	15	36	1.65
60	50	28	1.50
60	50	32	1.63
60	50	36	1.63
60	100	28	0.75
60	100	32	1.07
60	100	36	1.47

Table 2. ANOVA and contrast analysis of growth rates (K) of *Phaeocystis* sp. cultured at different salinities and irradiances

Source	p value >F
Irradiance	<0.001
Salinity	<0.001
Irradiance*salinity	0.222
28 ppt vs 32 ppt	0.32
28 ppt vs 36 ppt	0.018
32 ppt vs 36 ppt	0.13
15% vs 50%	0.058
15% vs 100%	0.20
50% vs 100%	0.004

* F: Test hypothesis(H_0) that there is no different in k among the treatments. Large p value is more consistent with H_0 , and the difference in K among the treatments will be significant as $p < 0.05$.

significant. It was found that the magnitude of the irradiance effect on growth was influenced by salinity (Figs. 4-6). For example, the growth curve in three irradiance groups were almost the same when salinity was 36 ppt. (Fig. 6). Among the three salinities tested, the growth rate of cultures grown at 28 ppt differed significantly from those grown at 36 ppt based on the result of contrast analysis. It is also true that the differences in growth curve among the three salinities groups depended on irradiance (Fig. 7-9), with high irradiance ($100 \mu\text{E m}^{-2} \text{ s}^{-1}$) leading to a greater difference in growth rate than that at lower irradiances (15 and $50 \mu\text{E m}^{-2} \text{ s}^{-1}$).

The influence of irradiance on *Phaeocystis* growth was further confirmed by the short-term photosynthetic response (Appendix C). When *Phaeocystis* was incubated at a series of irradiances and three salinities, photosynthesis was characterized by a hyperbolic response (Fig. 10-11). Two parameters (P_{max}^B and α^B) were derived from the data and used to conduct an analysis of variance (Table 3 and Appendix B). The results showed that salinity influenced the photosynthetic performance significantly.

TEP Produced by *Phaeocystis* Cultures

Concentrations of TEP in cultures began to increase when cell numbers stopped increasing (i.e., when the cultures reached stationary phase), and concentrations of TEP increased through time (Fig. 12). The mean TEP abundance was $450 \text{ ng xanthan } (\mu\text{g Chl a})^{-1}$, and maximum and minimum values were $2,338 \text{ ng xanthan } (\mu\text{g Chl a}^{-1})$ and $67 \text{ ng xanthan } (\mu\text{g Chl a}^{-1})$ respectively.

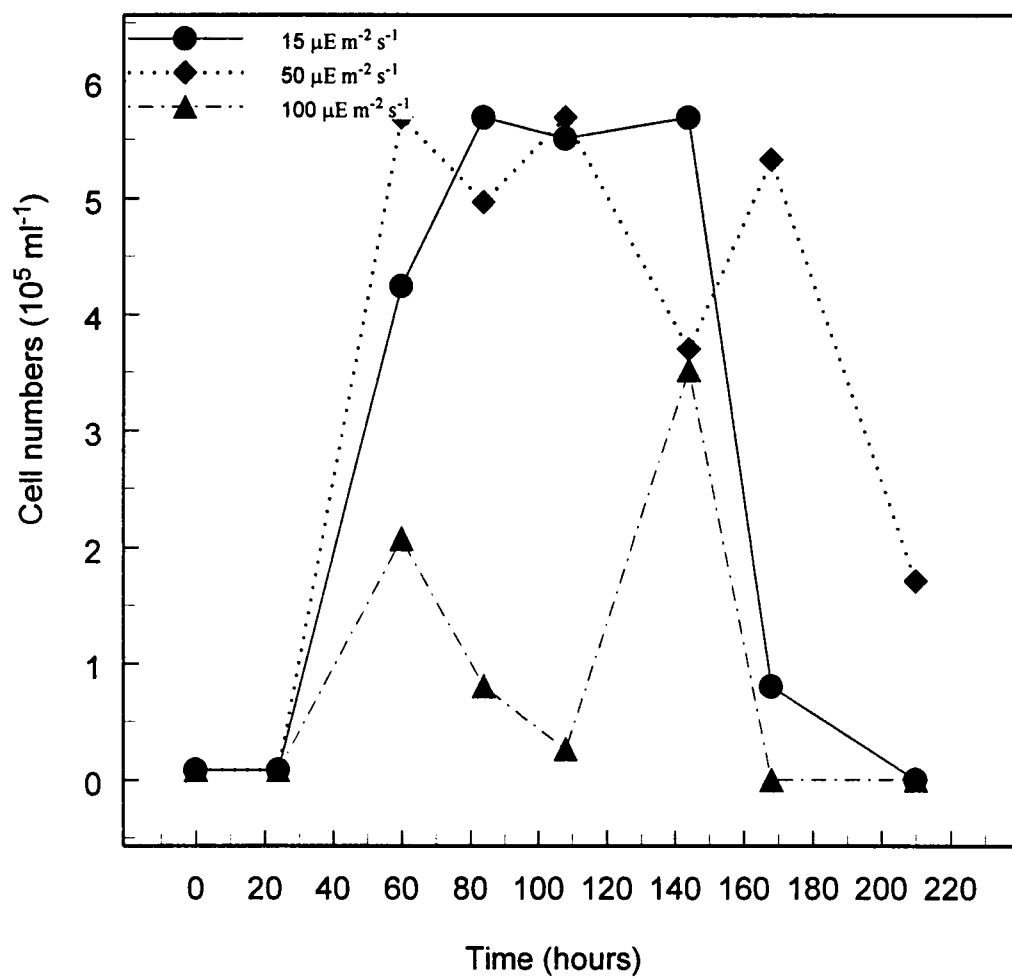


Figure 4. Growth of *Phaeocystis* sp. cultured at 28 ppt salinity and three irradiances

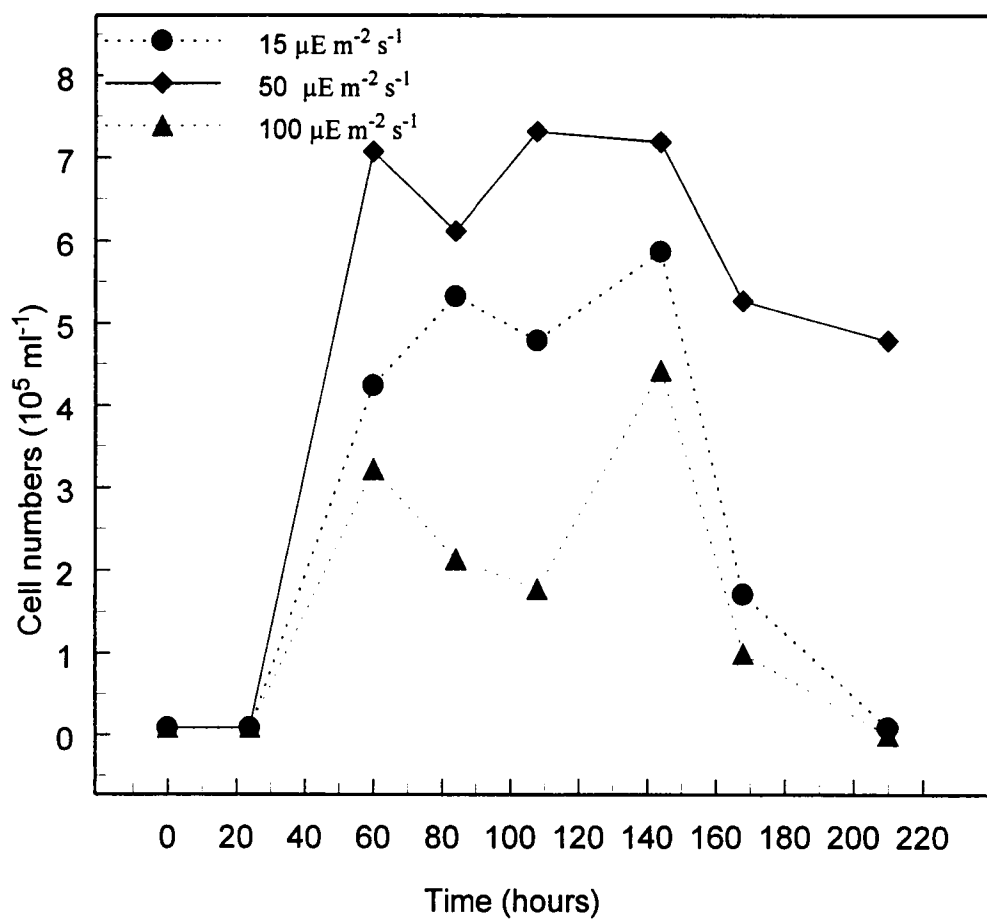


Figure 5. Growth of *Phaeocystis* sp. cultured at 32ppt salinity and three irradiances

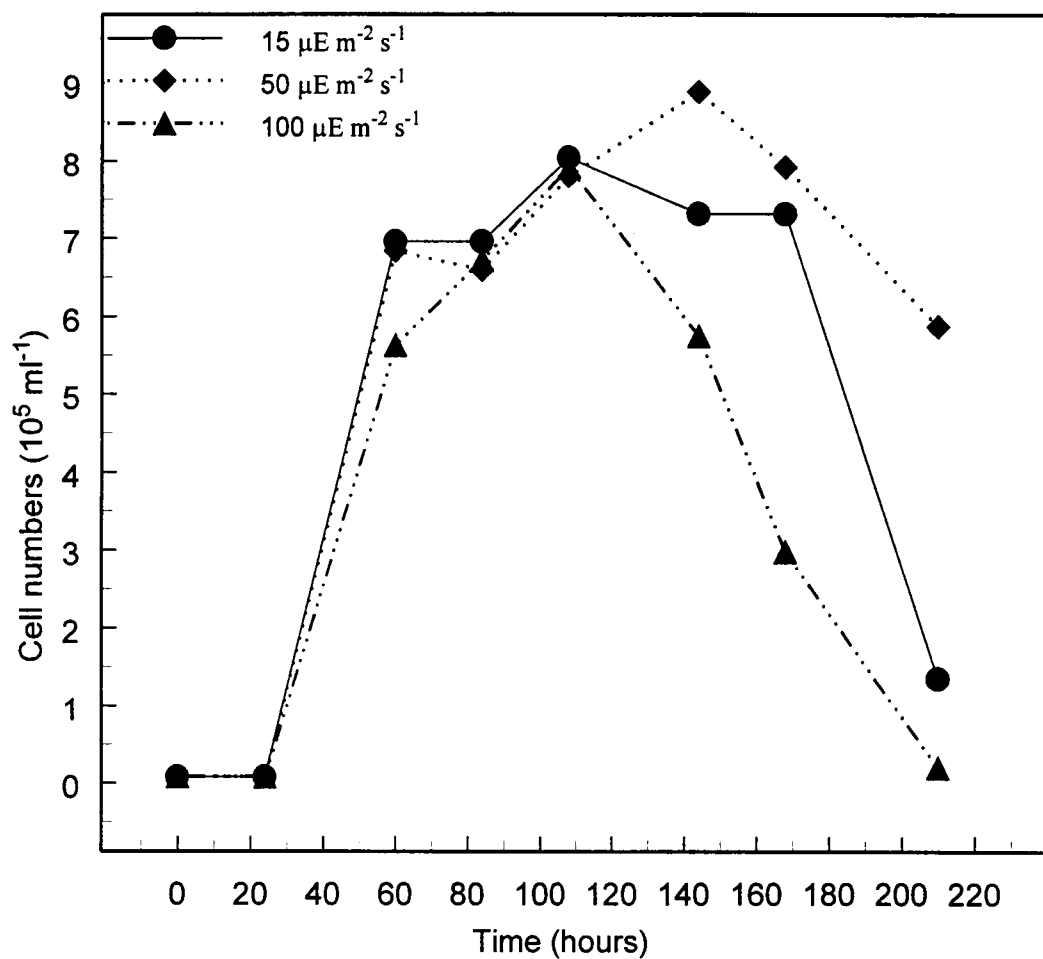


Figure 6. Growth of *Phaeocystis* sp. cultured at 36 ppt salinity and three irradiances

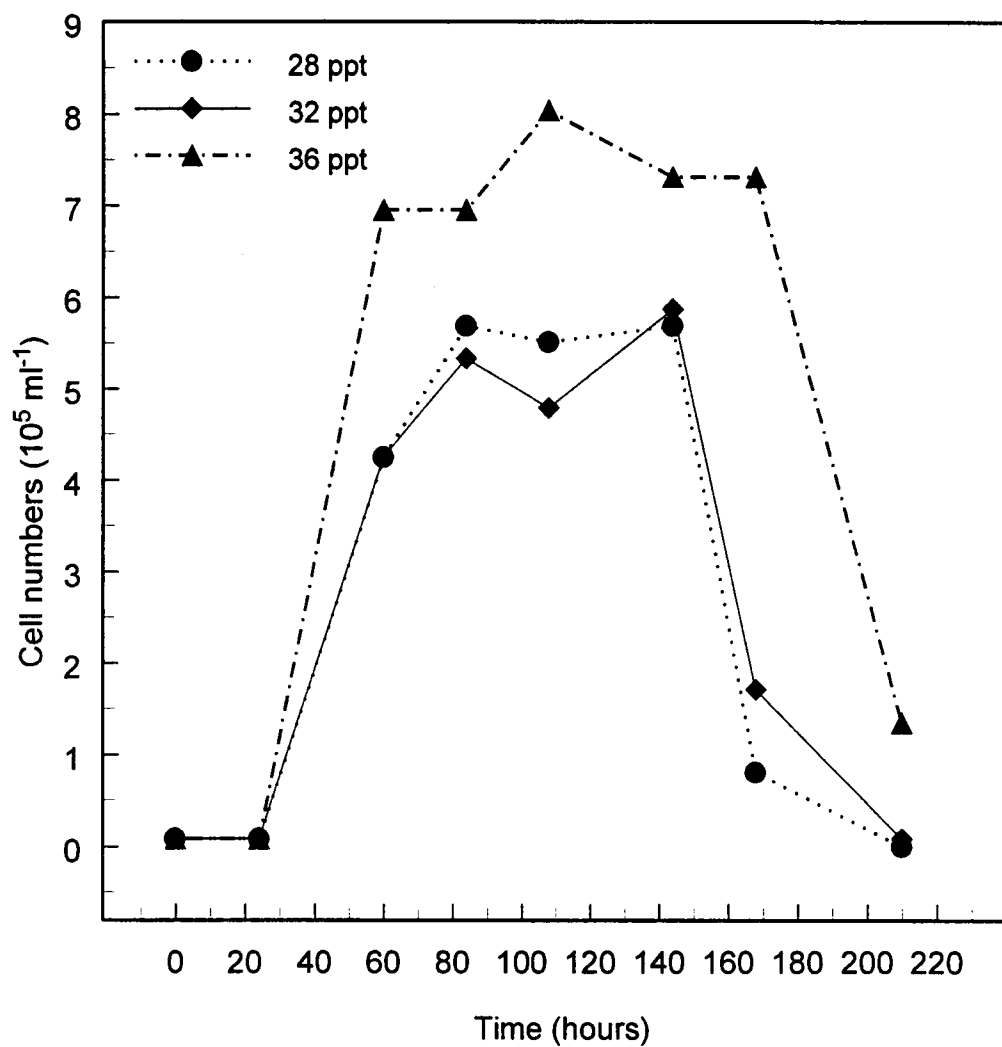


Figure 7. Growth of *Phaeocystis* sp. cultured at $15 \mu\text{E m}^{-2} \text{ s}^{-1}$ irradiance and three salinities

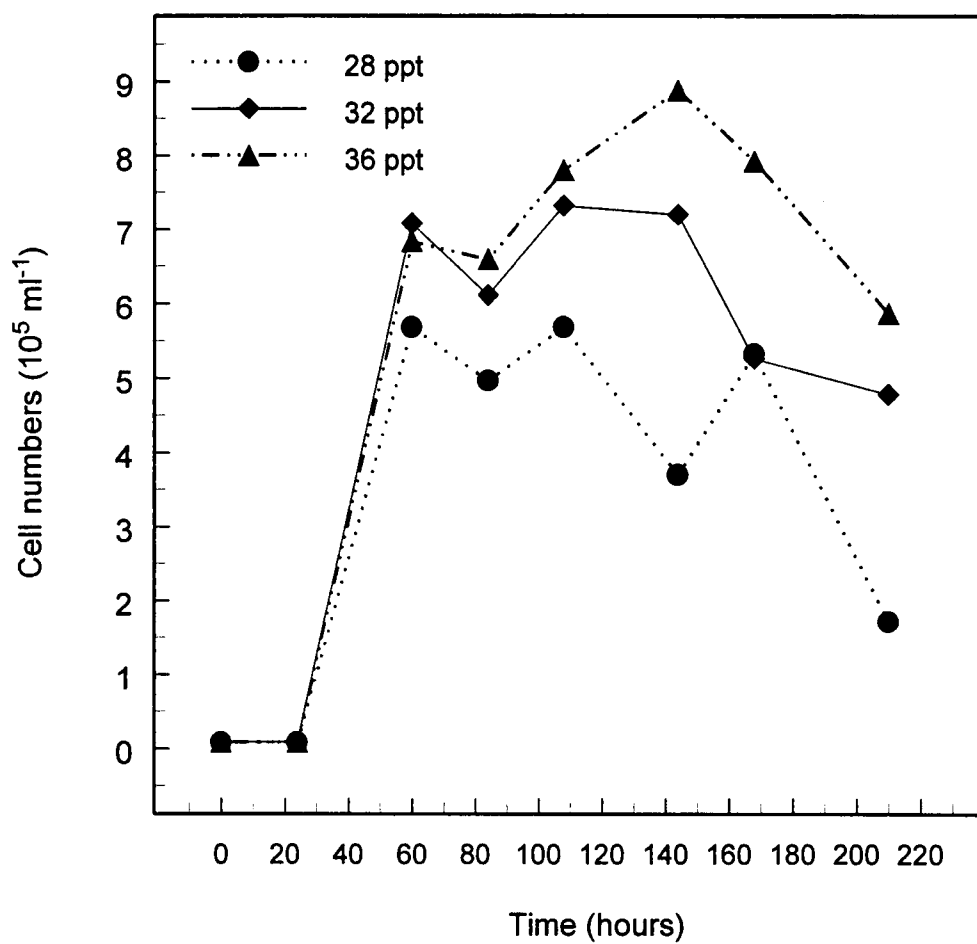


Figure 8. Growth of *Phaeocystis* sp. cultured at $50 \mu\text{E m}^{-2} \text{ s}^{-1}$ irradiance and three salinities

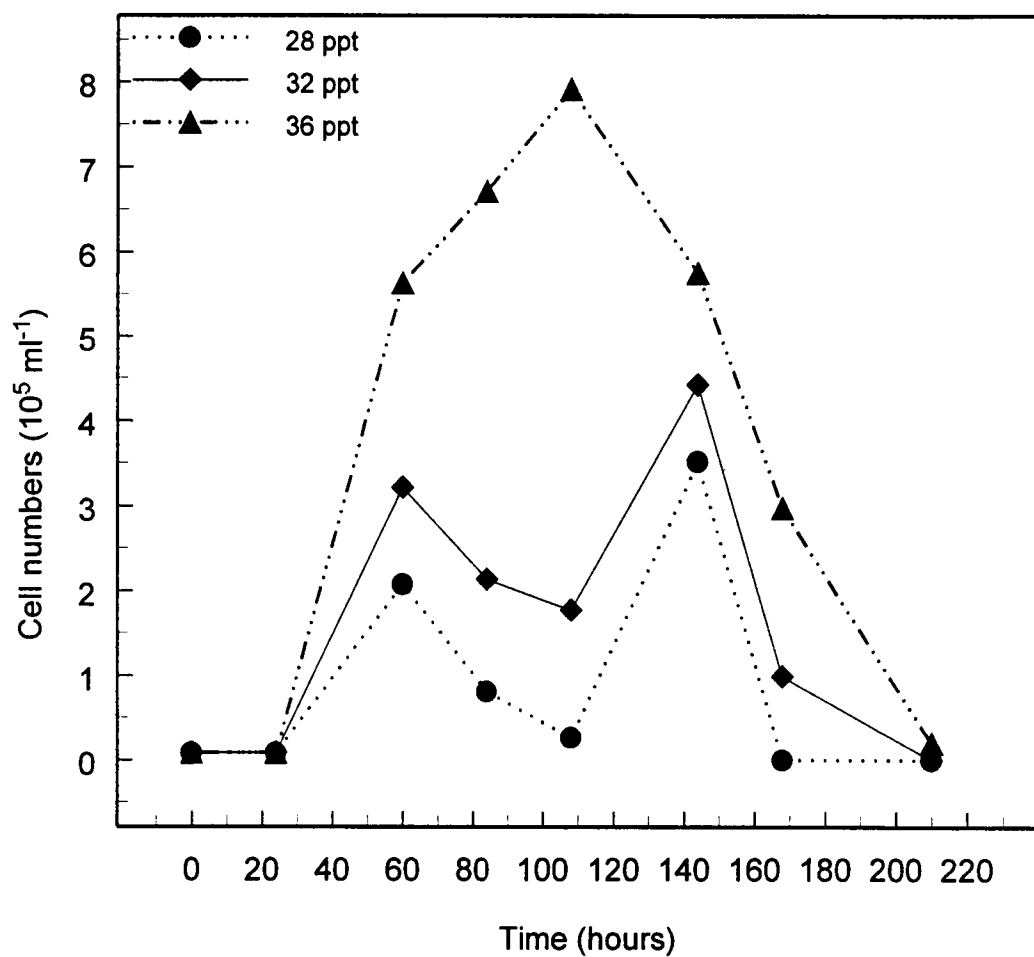


Figure 9. Growth of *Phaeocystis* sp. cultured at $100 \mu\text{E m}^{-2} \text{ s}^{-1}$ irradiance and three salinities

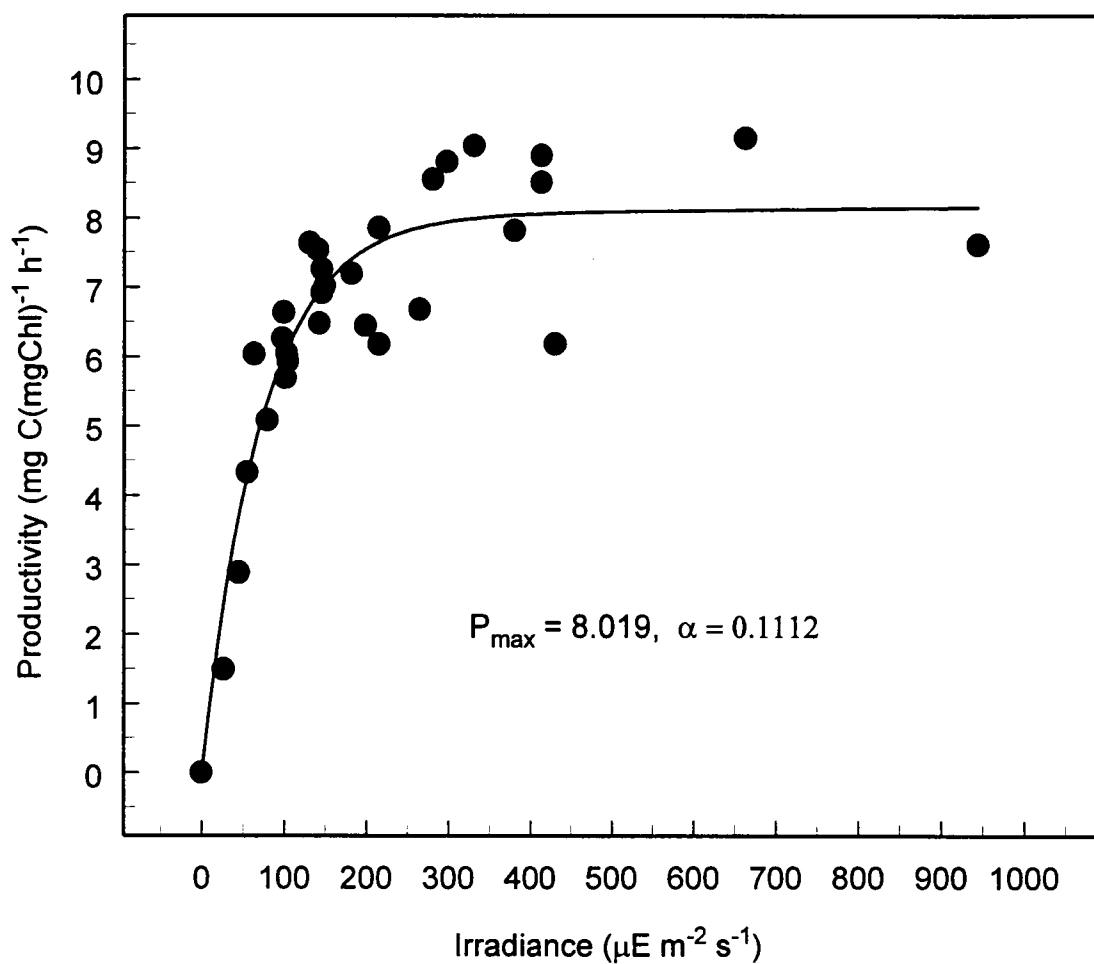


Figure 10. Photosynthesis vs. irradiance curves for *Phaeocystis* sp. cultured at 4°C for two hours. α is 0.01 mg (mg Chl)⁻¹ h⁻¹ (μmol m⁻² s⁻¹)⁻¹ and $P_{\max}^B$ is 0.91 mg C (mg Chl)⁻¹ h⁻¹

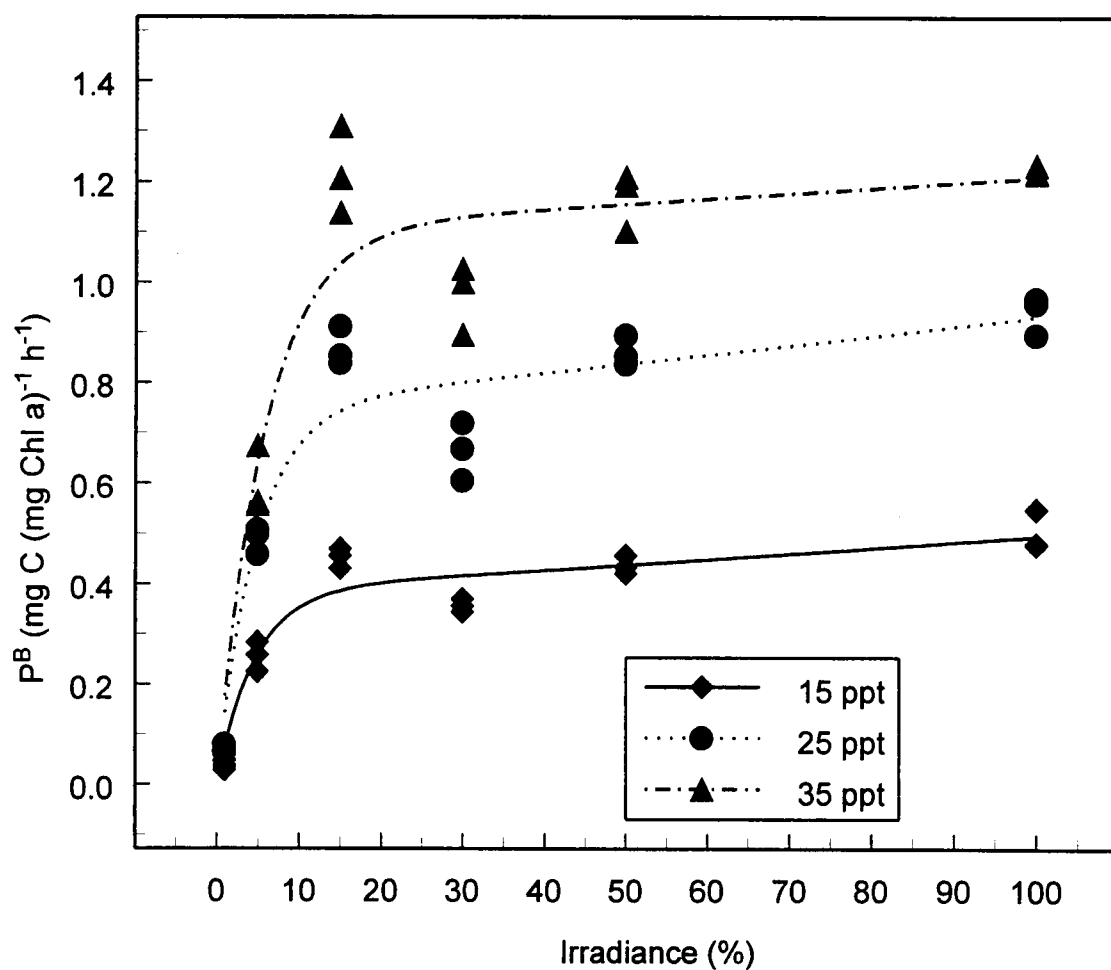


Figure 11. Photosynthesis vs. irradiance curve of *Phaeocystis* sp. cultured at 15, 25 and 35 ppt salinities. $P^B_{\max}=0.39, 0.75$ and $1.10 \text{ mg C } \alpha=0.08, 0.16$ and $0.19 \text{ mg C (mg Chl)}^{-1} \text{ h}^{-1} (\mu\text{mol m}^{-2} \text{ s}^{-1})^{-1}$

Table 3. Photosynthetic rate ($P_{\max}$) and photosynthetic efficiency (α^B) of *Phaeocystis* sp. cultured at different salinities.

Salinity (ppt)	Sample #	$P_{\max}$ (mg C (mg Chl) ⁻¹ h ⁻¹)	α^B (mg C (mg Chl) ⁻¹)(μ mol m ⁻² s ⁻¹) ⁻¹
15	1	0.389	0.389
15	2	0.370	0.370
15	3	0.408	0.408
25	1	0.694	0.694
25	2	0.791	0.791
25	3	0.769	0.769
35	1	1.075	1.075
35	2	1.118	1.118
35	3	1.150	1.150

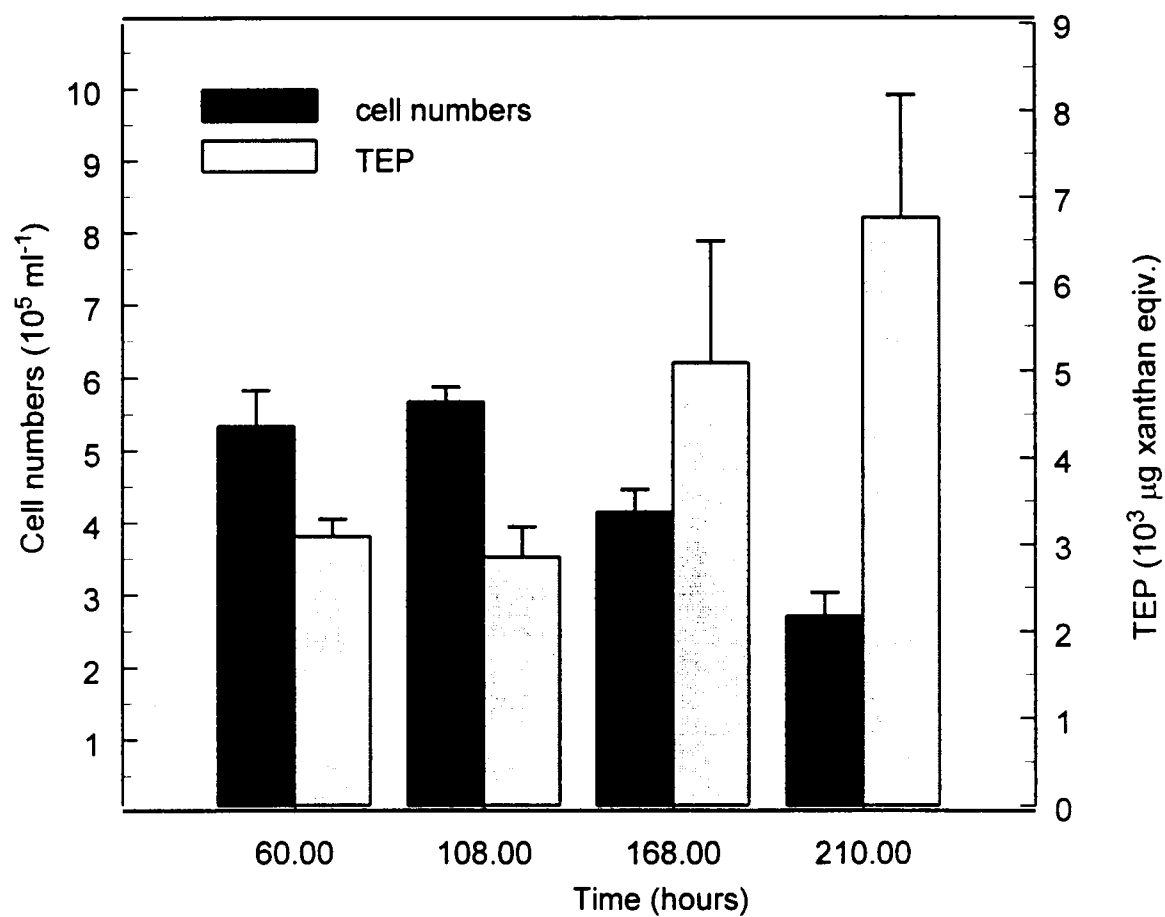


Figure 12. The changes of TEP concentrations and cell numbers through time

TEP concentrations in 90% samples were within the ranges of 100 to 1000 ng xanthan ($\mu\text{g Chl a}^{-1}$; Appendix E).

Influence of Salinity and Irradiance on TEP Production

It was observed that TEP production varied as a function of culture conditions such as salinity and irradiance (Table 4). The results of an analysis of variance (Table 5) showed that irradiance and salinity impacted TEP concentrations significantly ($P < 0.001$, $P = 0.01$). Contrast analysis indicated that TEP production in the 15 and 50 $\mu\text{E m}^{-2} \text{s}^{-1}$ groups significantly differed from the 100 $\mu\text{E m}^{-2} \text{s}^{-1}$ group ($P = 0.01$ and 0.004). These differences are shown in Figs. 13-15. The response of TEP to variable irradiance in most samples during short incubations exhibited similar trends as photosynthesis; that is, TEP concentrations generally increased as irradiance increased (Fig. 16 and Appendix D). The contrast analysis did not show a significant difference in TEP production among the three salinity groups. However, the mean TEP concentration in the three salinity groups was 644, 981 and 1110 ng xanthan ($\mu\text{g Chl a}^{-1}$), respectively, and the change of TEP concentration through time at 28 ppt salinity differed from that of the other two groups (Figs. 17-19). Accordingly, at least in one treatment, low salinity influenced the TEP production. This impact might be masked by the interaction between salinity and irradiance when the salinity groups, each of which included three irradiances, were compared. For example, there was less influence of irradiance on the TEP production observed in the 32 ppt and 36 ppt groups than in the 28 ppt groups.

Table 4. TEP concentrations in *Phaeocystis* sp. cultures grown at different salinities and irradiances

Time (hours)	Irradiance ($\mu\text{E m}^{-2} \text{s}^{-1}$)	Salinity (ppt)	TEP concentration (ng xanthan ($\mu\text{g Chl a}$) $^{-1}$)
210	15	28	6859
210	15	32	696
210	15	36	516
210	50	28	830
210	50	32	383
210	50	36	432
210	100	28	1850
210	100	32	752
210	100	36	1362

Table 5. ANOVA and contrast analysis of TEP produced by *Phaeocystis* sp. cultured at different salinities and irradiances

Source	F value	p value>F
Irradiance	21.2	<0.001
Salinity	6.8	0.01
Irradiance*salinity	3.2	0.05
28 ppt vs 32 ppt	0.15	0.70
28 ppt vs 36 ppt	1.89	0.19
32 ppt vs 36 ppt	0.97	0.34
15 ppt vs 50 ppt	0.27	0.61
15 ppt vs 100 ppt	8.62	0.01
50 ppt vs 100 ppt	11.93	0.004

* F: Test hypothesis(H_0) that there is no difference in K among the treatments. Large p values are more consistent with H_0 , and the difference in K among the treatment will be significant as $p < 0.05$.

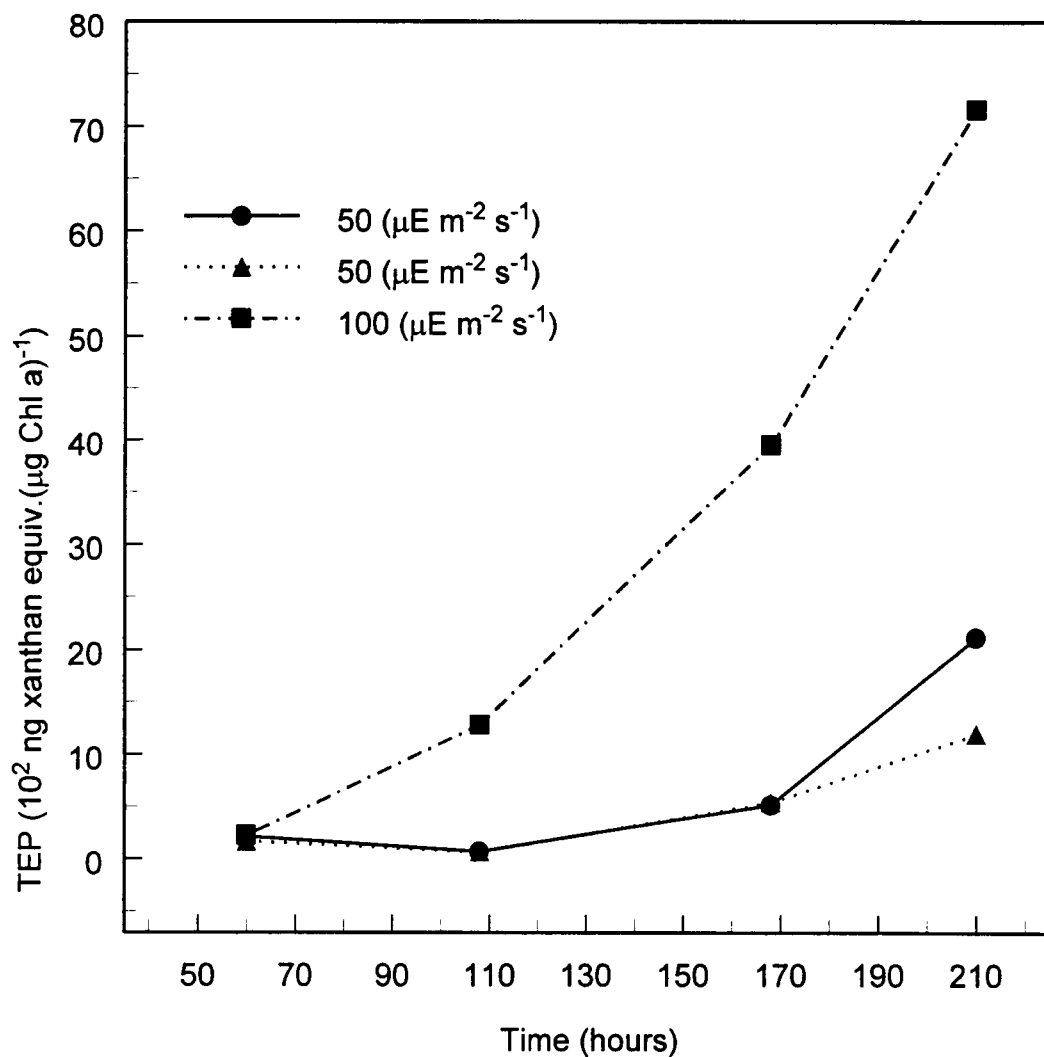


Figure 13. The changes of TEP concentrations through time for *Phaeocystis* cultured at 28 ppt salinity and three irradiances

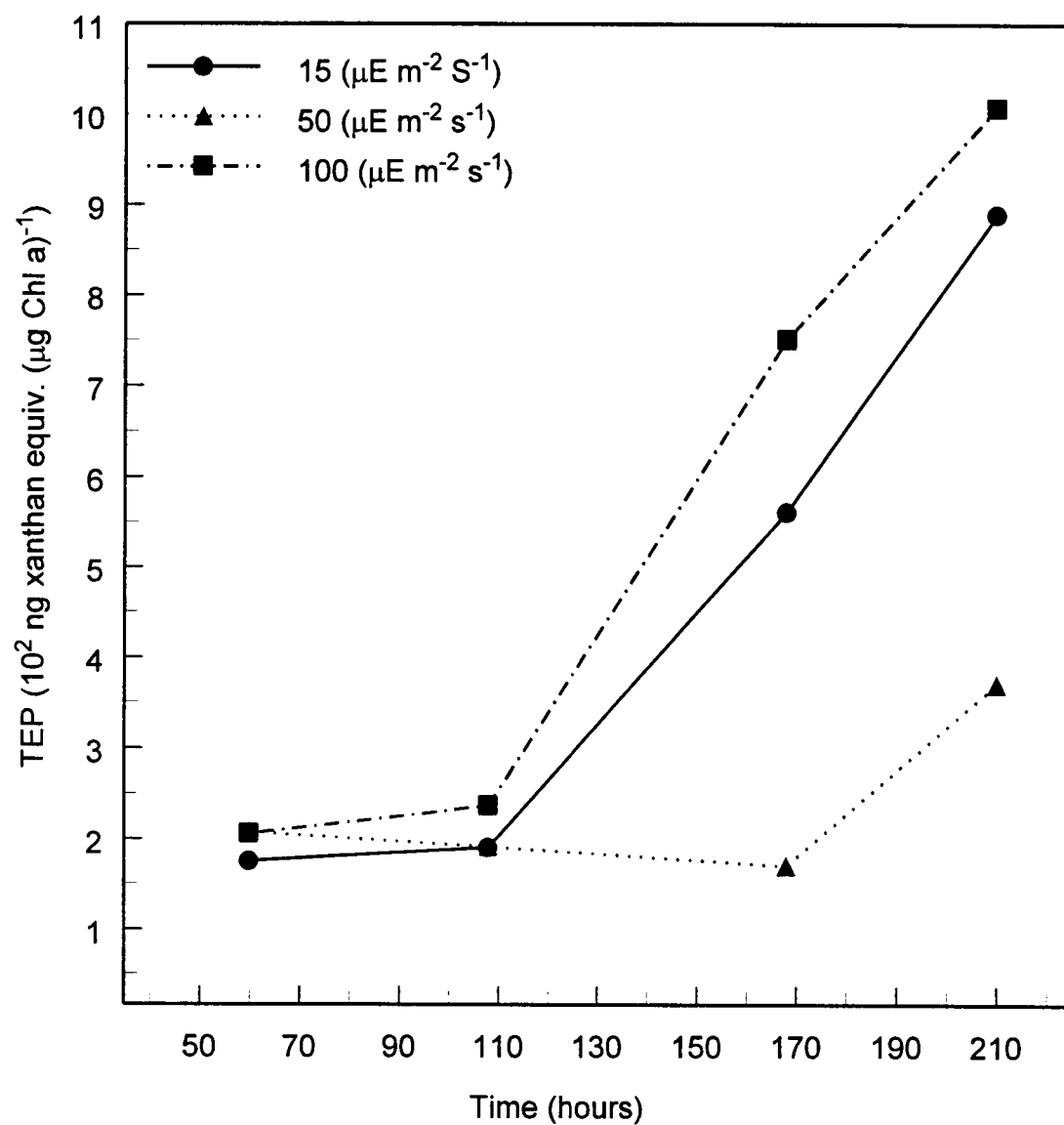


Figure 14. The changes of TEP concentrations through time for *Phaeocystis* cultured at 32 ppt salinity and three irradiances

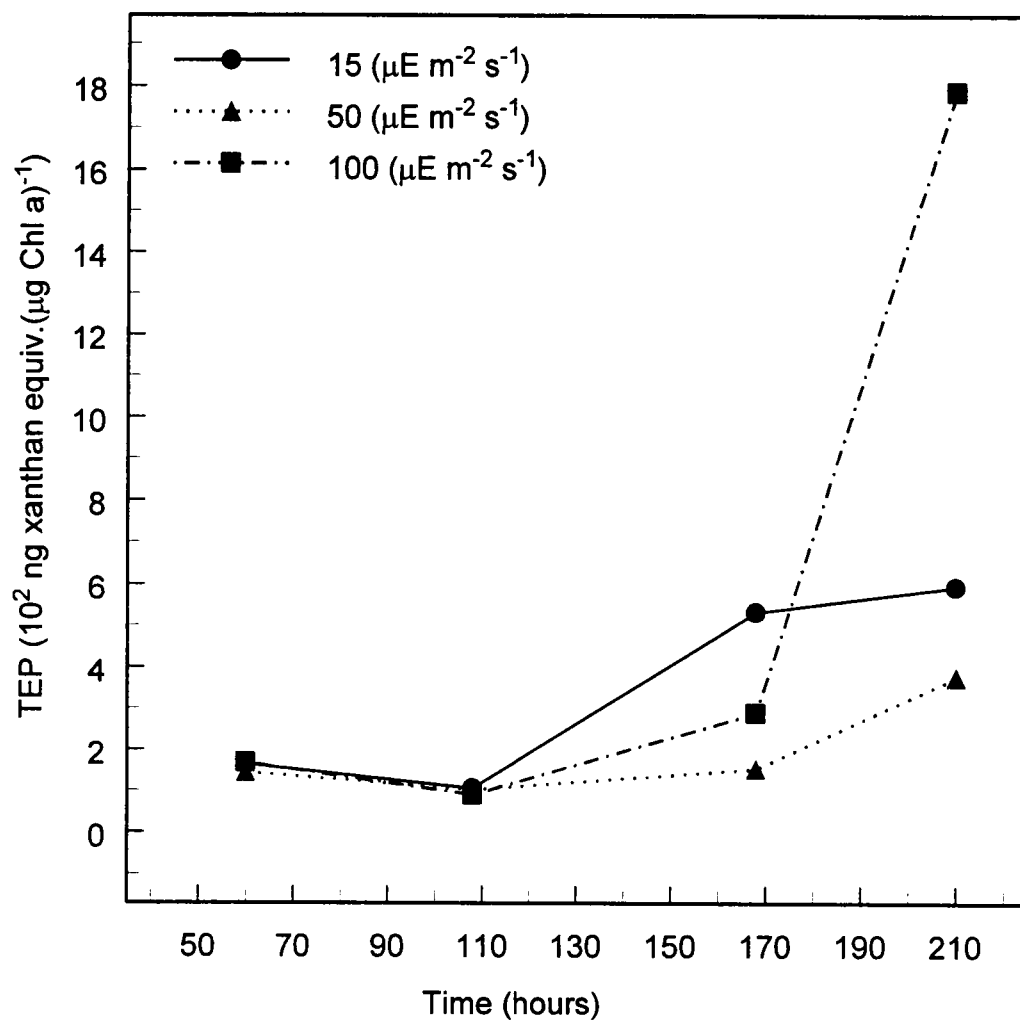


Figure 15. The changes of TEP concentrations through time for *Phaeocystis* cultured at 36 ppt salinity and three irradiances

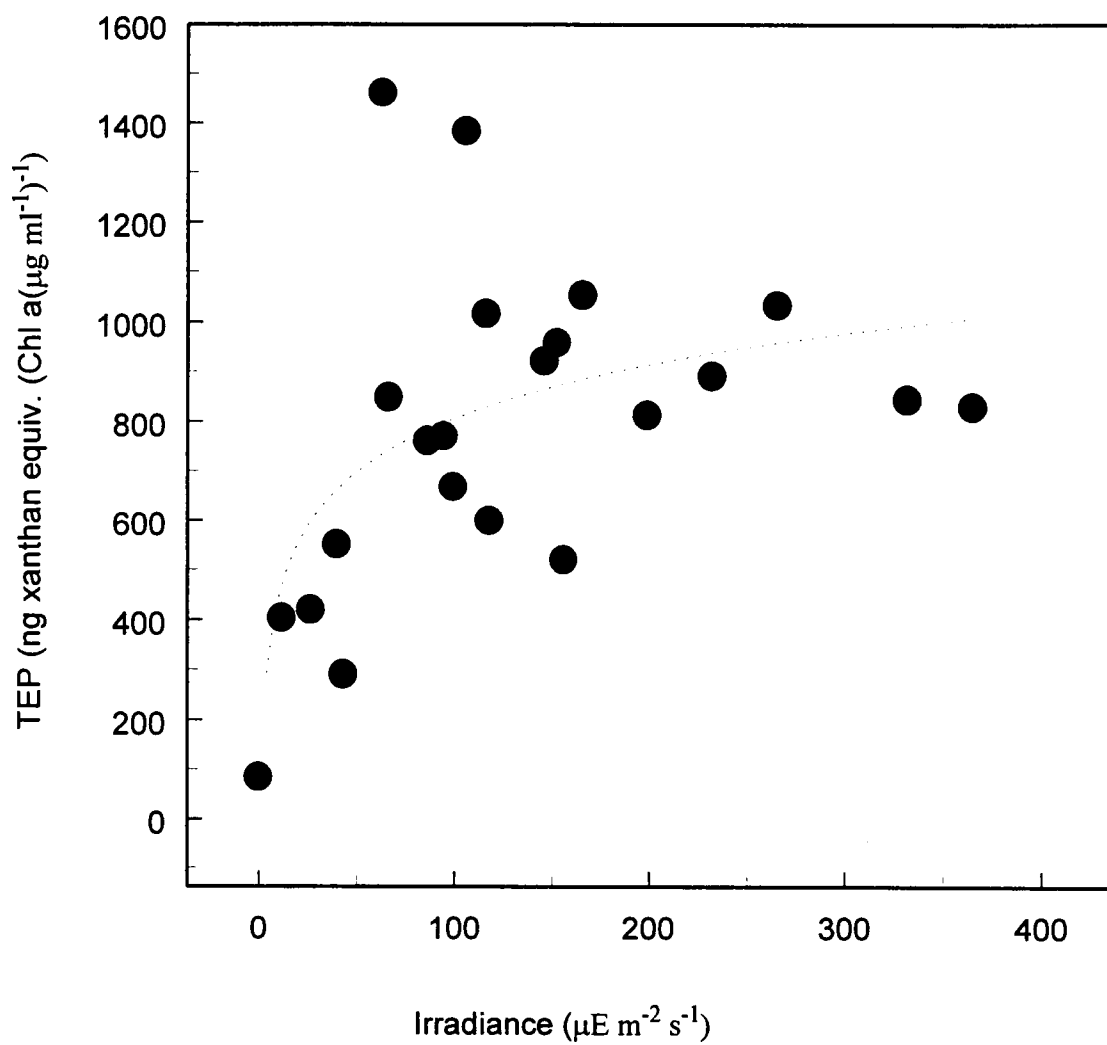


Figure 16. TEP concentrations of *Phaeocystis* sp. cultured for 4 hours at 2°C

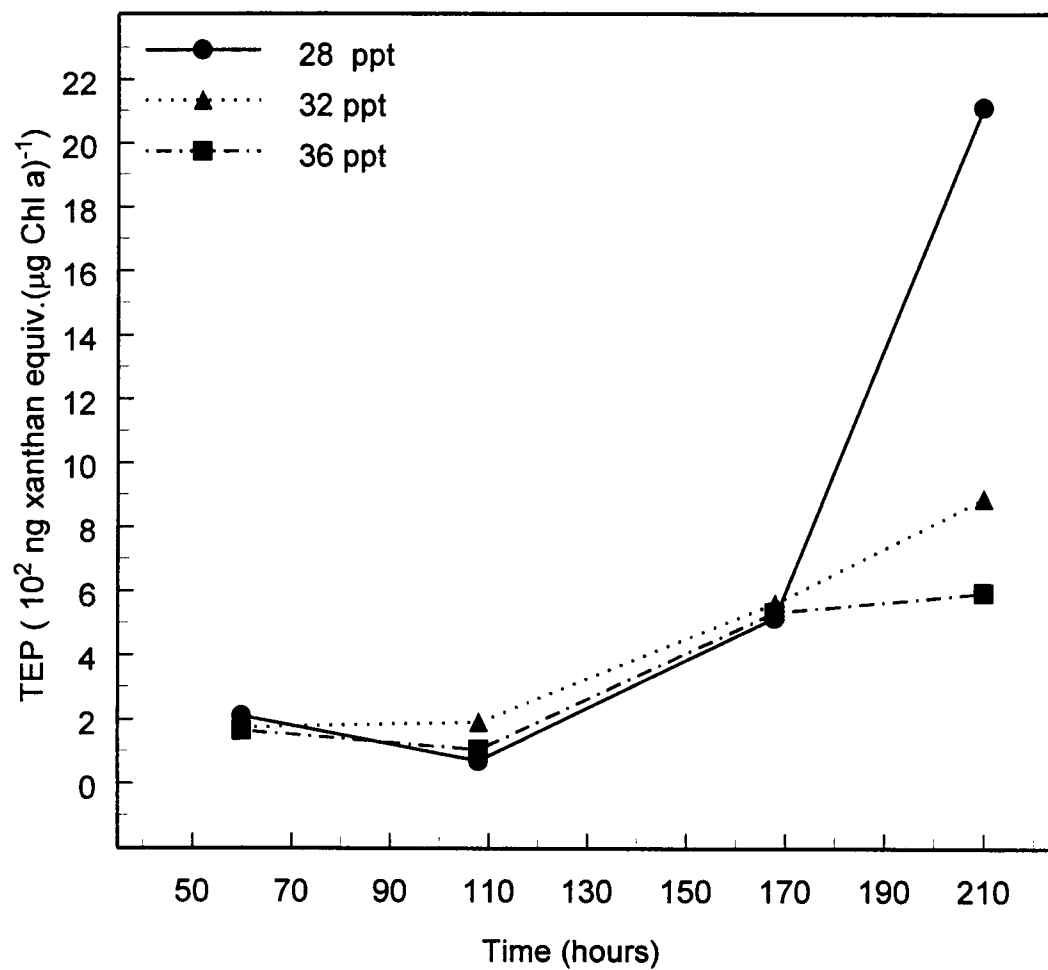


Figure 17. The changes of TEP concentrations through time for *Phaeocystis* cultured at $15 \mu\text{E m}^{-2} \text{ s}^{-1}$ irradiance and three salinities

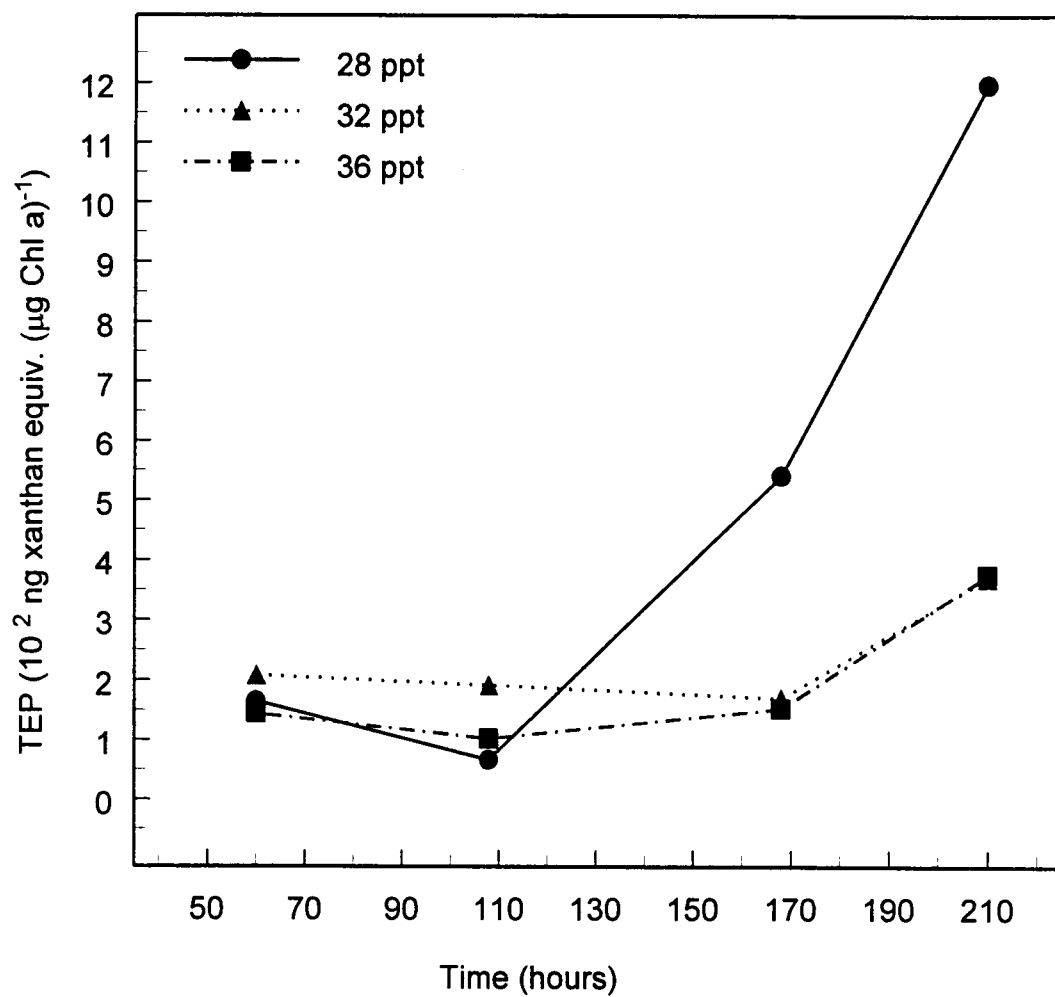


Figure 18. The changes of TEP concentrations through time for *Phaeocystis* cultured at $50 \mu\text{E m}^{-2} \text{ s}^{-1}$ irradiance and three salinities

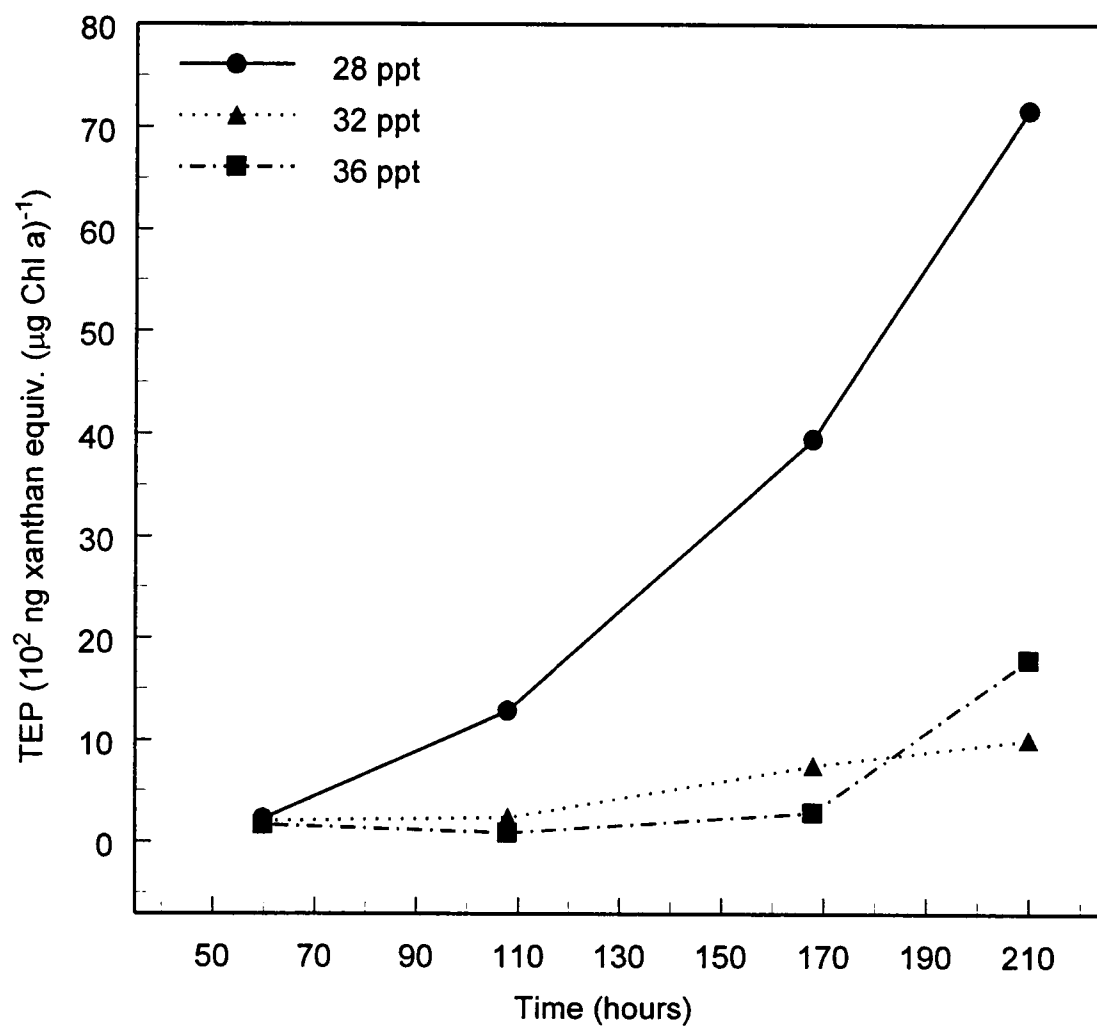


Figure 19. The changes of TEP concentrations through time for *Phaeocystis* sp. cultured at $100 \mu\text{E m}^{-2} \text{s}^{-1}$ irradiance and three salinities

The results described above have supported my hypothesis that *Phaeocystis* can produce TEP during its stationary stage, and the amount of TEP produced depended on the time and the factors that influenced *Phaeocystis* growth. These studies indicate that both light and salinity impact the TEP production, but irradiance has more of an influence than salinity.

TEP Abundance in the Ross Sea

The hydrography of the Ross Sea mixed layer during the field study is summarized in Table 6. In general, temperatures were low and ranged from -1.9°C to 0°C, while nitrate concentrations were high (mean of which was 26.1 μM). The salinity ranged from 34.2 ppt to 34.5 ppt, and the mean was 34.4 ppt. The maximum chlorophyll concentration was 7.8 $\mu\text{g l}^{-1}$ with a mean of 4.0 $\mu\text{g l}^{-1}$. TEP abundances in the Ross Sea at the time of survey varied by more than three orders of magnitude (0 -2800 $\mu\text{g l}^{-1}$ xanthan equiv.)(Appendix F). The average TEP concentration was 308 $\mu\text{g l}^{-1}$ xanthan. In most samples the TEP concentration ranged from 100 to 1000 $\mu\text{g l}^{-1}$ xanthan equiv., and were lower than 500 $\mu\text{g l}^{-1}$ xanthan in 80% of the samples. Its abundance in the water column increased during the development of the algal bloom, as concentrations in TEP at the site dominated by *Phaeocystis* (Fig.20) increased 2 to 5-fold from November 18 to December 5. It was also found that *Phaeocystis* produced a similar amount of TEP as diatoms, as evidenced by the similarity between TEP concentrations at stations that were dominated by *Phaeocystis* and *Pseudonitzschia*, respectively (Fig 21).

Table 6. Summary of various environmental parameters for the study areas

Parameter	Values		
	Maxium	Minimum	Average
Temperature (°C)	0	-1.9	-1
Salinity (ppt)	34.5	34.2	34.34
Wind speed (m s ⁻¹)	25	2	14
Nitrate (μM)	30.8	18.9	26.1
Ammonium (μM)	1.35	0.01	0.19
Phosphate (μM)	2.13	1.57	1.88
Silicate (μM)	78.5	69.2	75.1
Chlorophyll (μg l ⁻¹)	7.78	0.24	4.0

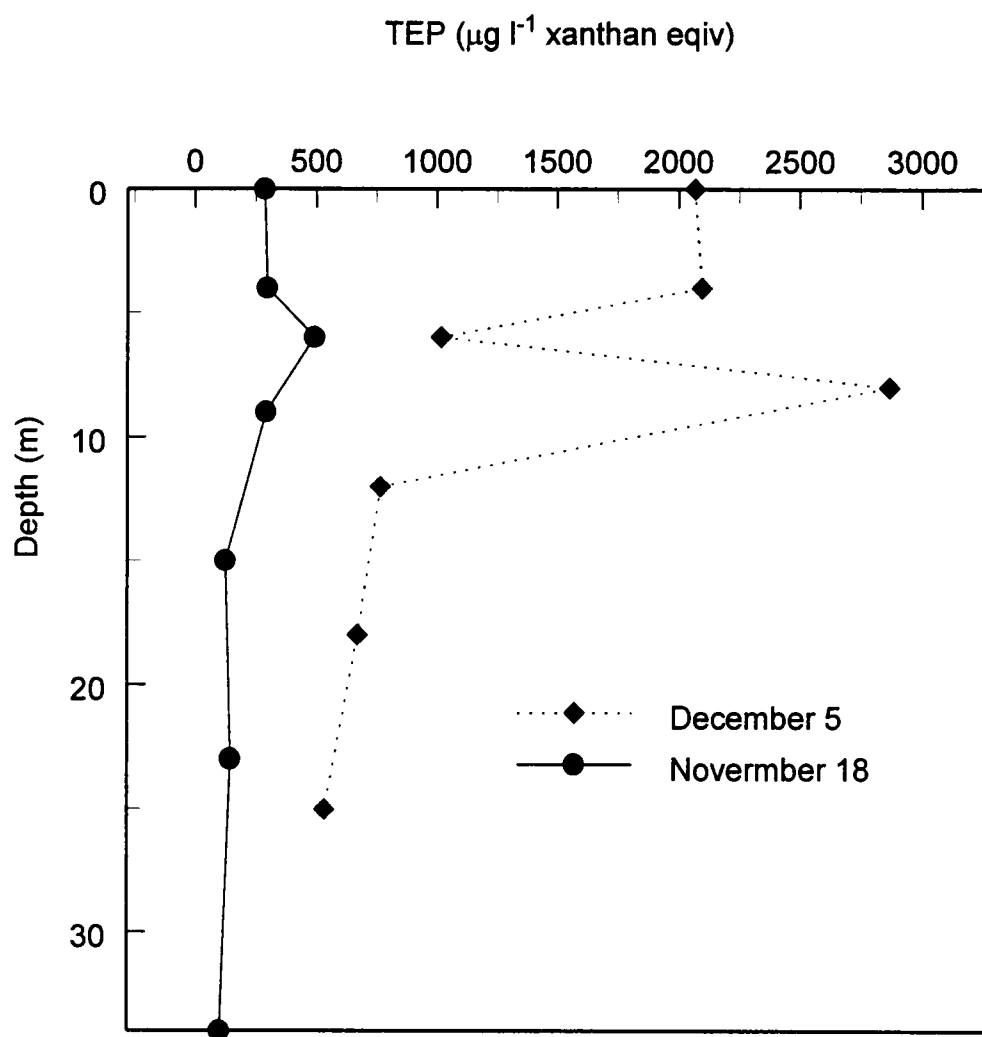


Figure 20. The changes of TEP concentrations at the station surveyed at November 18 and December 5

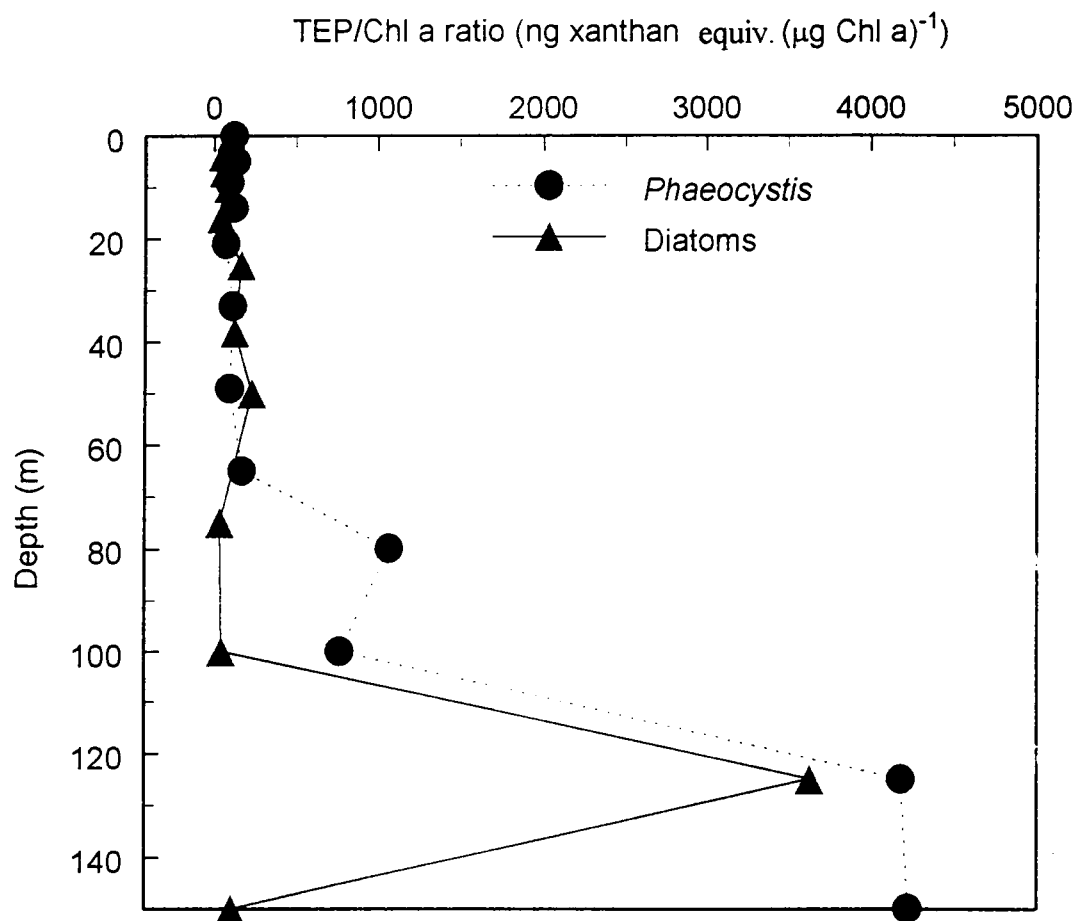


Figure 21. Vertical distribution of TEP/Chl a ratio at station dominated by *Phaeocystis* and diatoms

TEP Horizontal Distribution

The highest concentrations of TEP were found in the *Phaeocystis* bloom area in the southeastern Ross Sea. A decrease of TEP concentrations away from shore, from east to west and from south to north was observed (Figs. 22-25). Maximum concentrations of TEP coincided with maximum chlorophyll *a* levels (Fig. 26), which suggests a correlation between TEP and chlorophyll *a*. This correlation was highly significant ($R^2=0.001$). Some parameters that correlated with chlorophyll *a*, such as nitrate and phosphate, were also correlated with TEP concentration (Table 7). Salinity ranged from 34.2 to 34.5 over the period of the survey and did not influence the change of TEP ($P=0.151$).

TEP Vertical Distribution

Vertically, there were three types of TEP vertical distribution patterns. In Section III TEP exhibited a surface maximum and decreased with depth (Fig. 23). In the second type, subsurface peaks in TEP abundance were observed as well as at about 120 m, as shown in Section I (Fig. 24). The third type was characterized by the highest concentrations located in the bottom layer (Fig. 25). Unlike the TEP, the vertical distributions of chlorophyll *a* at most stations were characterized by surface maxima within the mixed layer (Figs. 27-30). Because the different vertical distribution between TEP and chlorophyll, maxima of TEP/Chlorophyll *a* ratios appeared in the bottom of the water column at most stations investigated (Figs. 31-33).

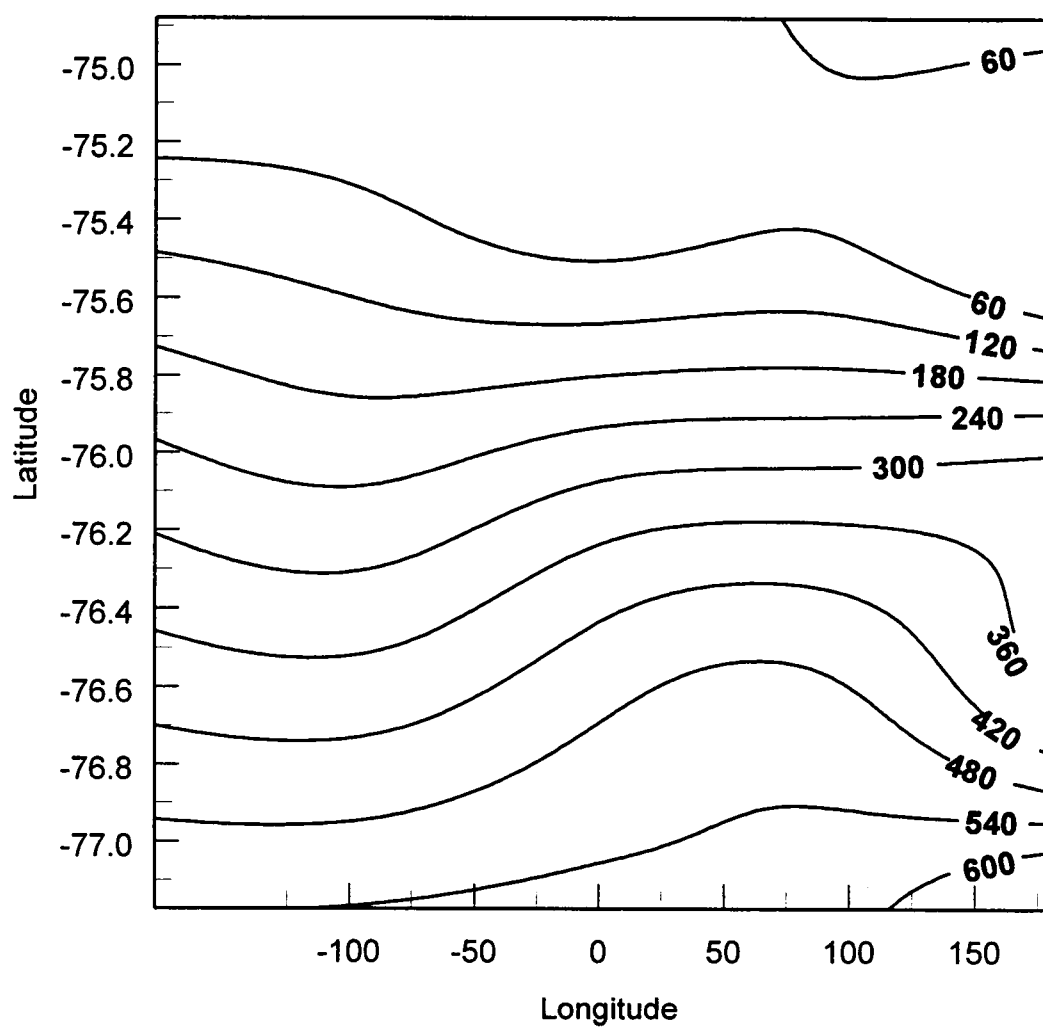


Figure 22. Surface TEP concentrations ($\mu\text{g l}^{-1}$ xanthan equiv) distribution; negative latitude represents south, positive and negative 100 longitude represent 170° west and 170° east respectively

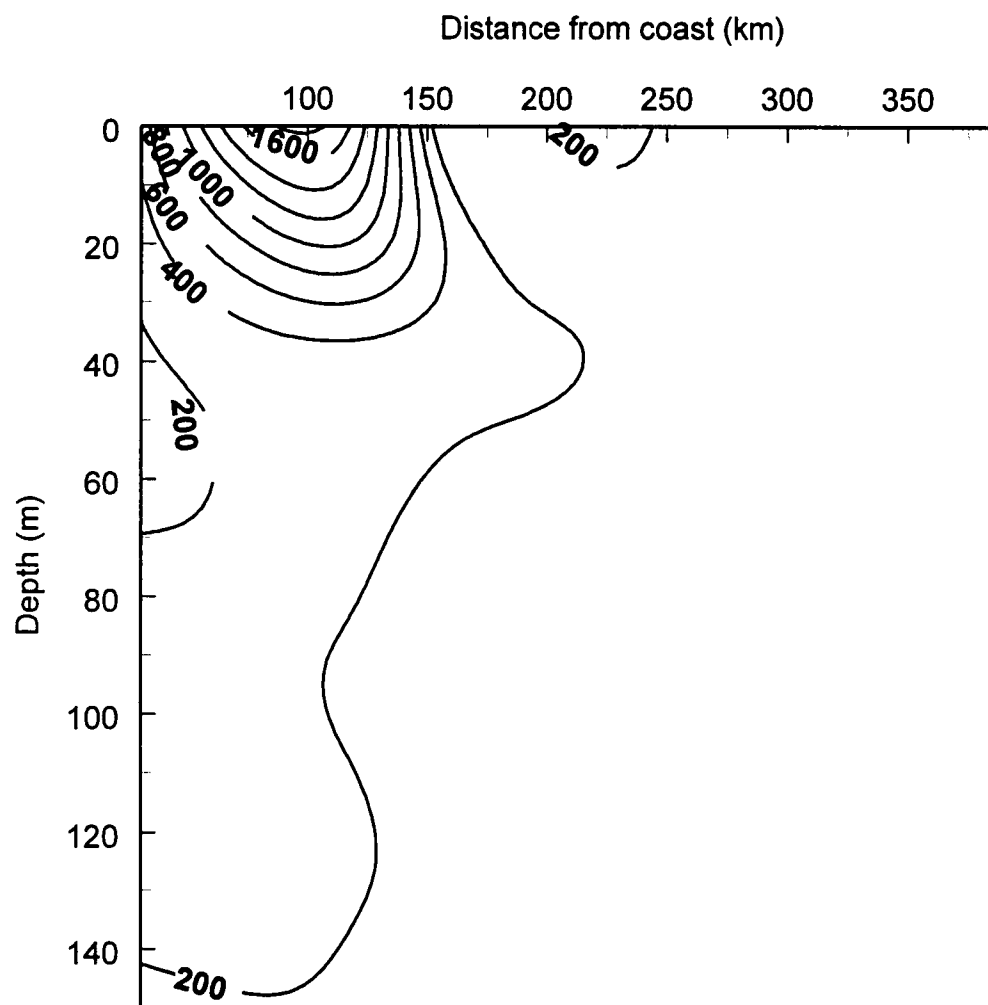


Figure 23. The vertical distribution of TEP concentrations across Transect 2 (173°E) in the Ross Sea.

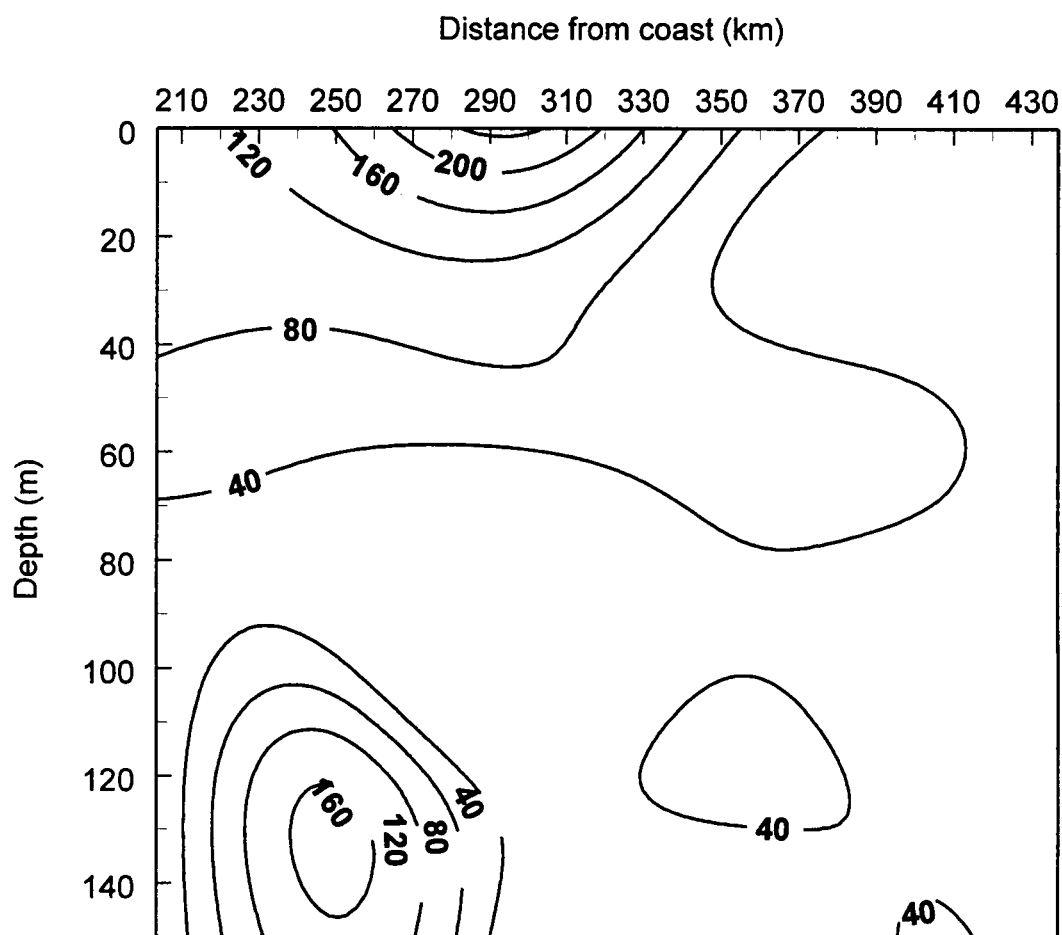


Figure 24. The vertical distribution of TEP concentrations across Transect 3 (at 75° S) in the Ross Sea.

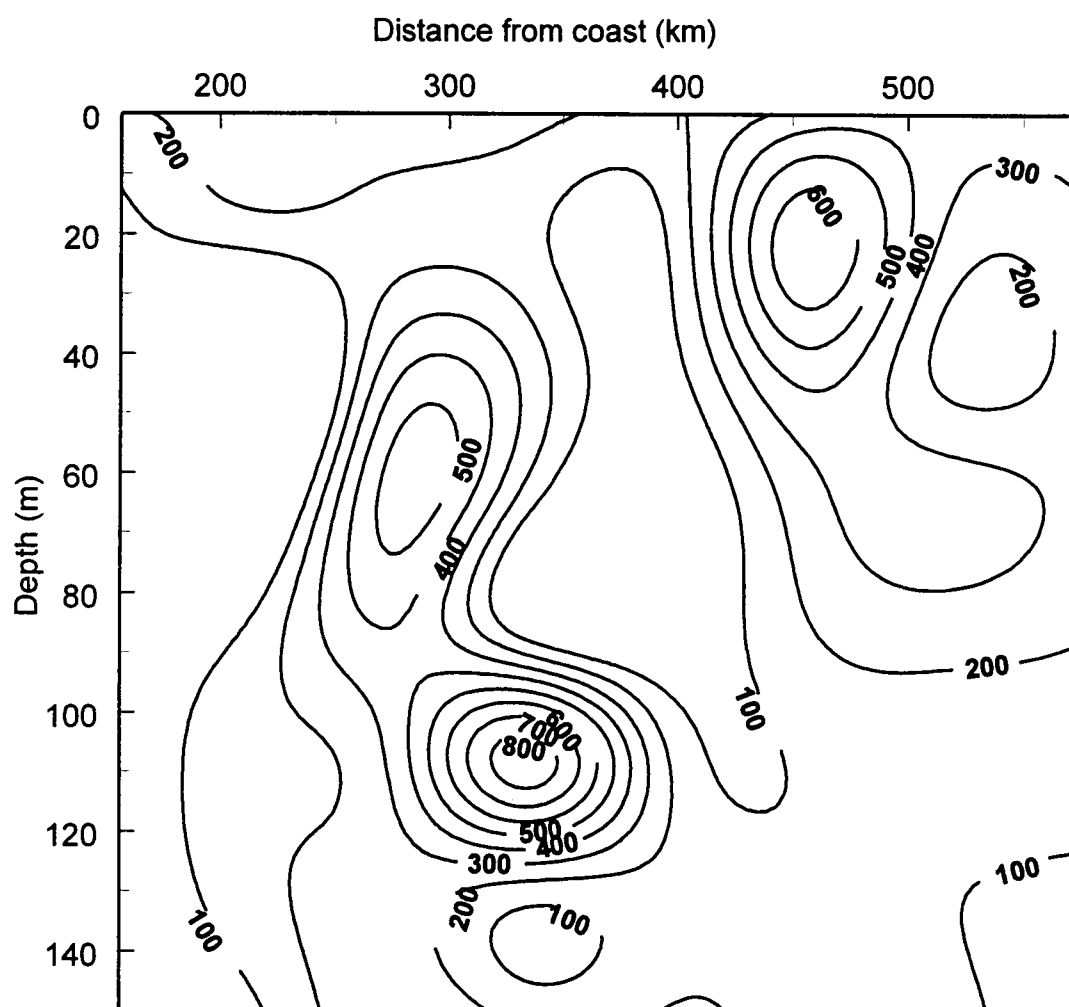


Figure 25. The vertical distribution of TEP concentrations across
Transect 1 (at 76° S) in the Ross Sea

Table 7. Significant correlation coefficients of environmental parameters with TEP concentrations in the mixed layer

Parameter	Correlation Coefficients	Probability >F
Chlorophyll a ($\mu\text{g l}^{-1}$)	0.65	<0.001
Nitrate (μM)	-0.77	<0.001
Phosphate (μM)	-0.58	<0.001
Temperature ($^{\circ}\text{C}$)	0.46	<0.001
Wind speed (m s^{-1})	-0.25	0.03

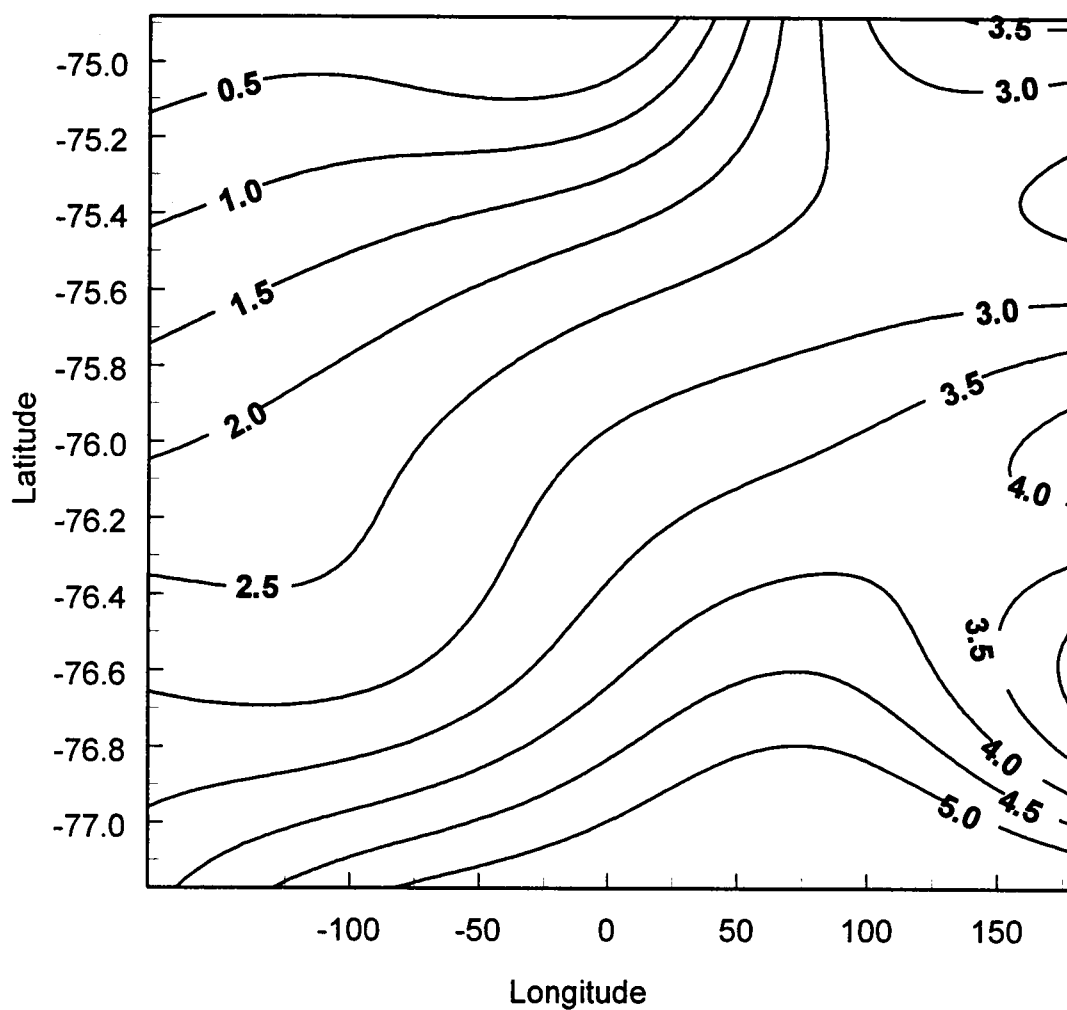


Figure 26. Surface chlorophyll a concentrations ($\mu\text{g ml}^{-1}$) distribution; negative latitude represents south, positive and negative one hundred longitude represent 170° west and 170° east

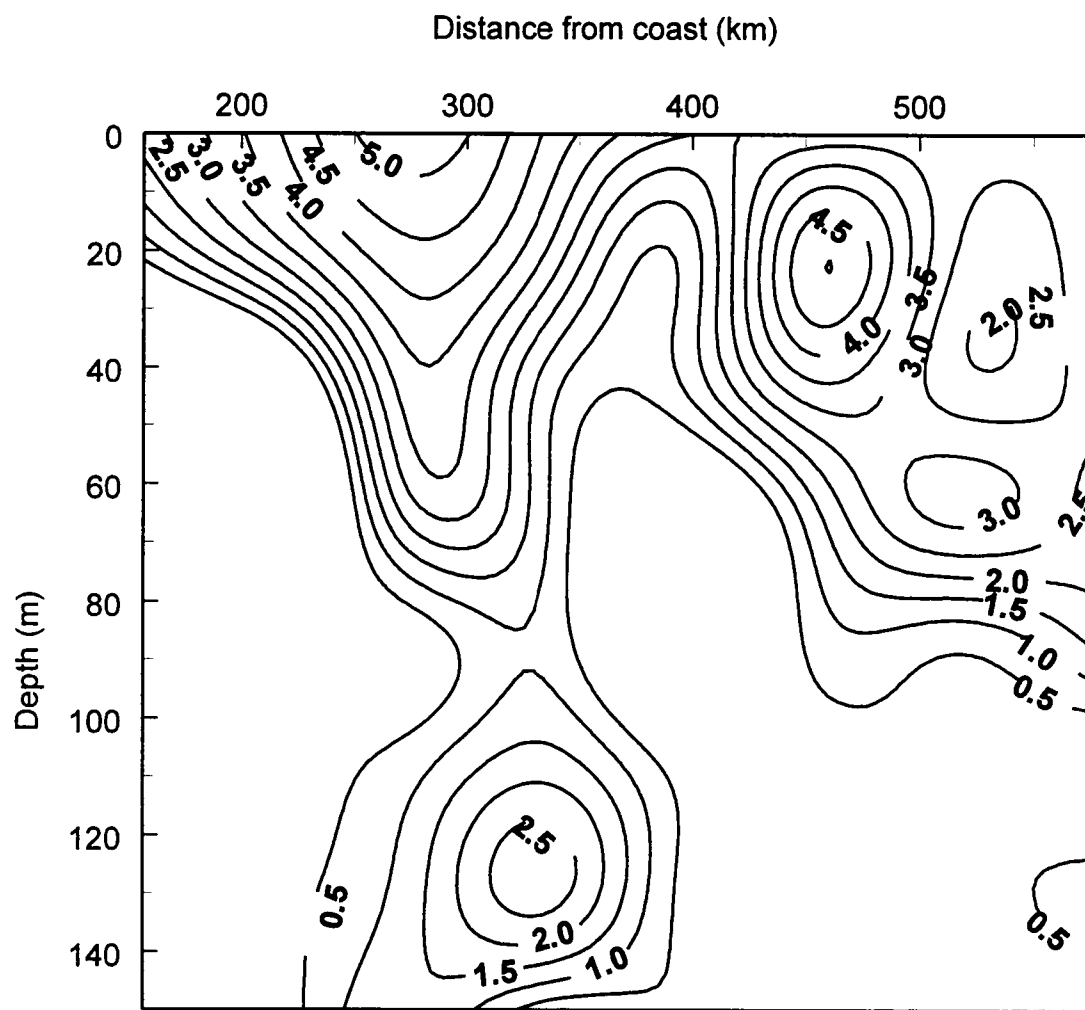


Figure 27. The vertical distribution of chlorophyll concentration across Transect 1 (at 76° S) in the Ross Sea.

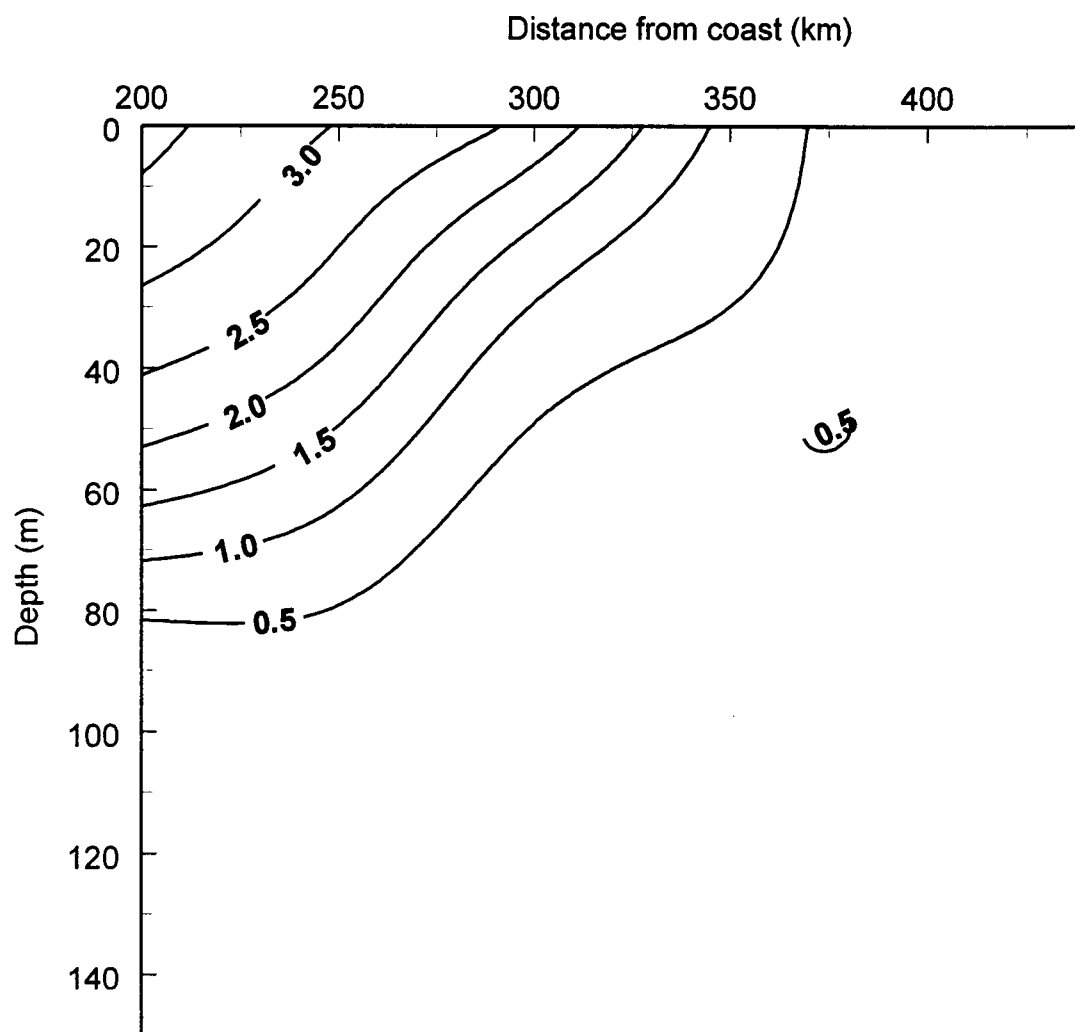


Figure 28. The vertical distribution of chlorophyll a concentrations across Transect 3 (at 75°S) in the Ross Sea

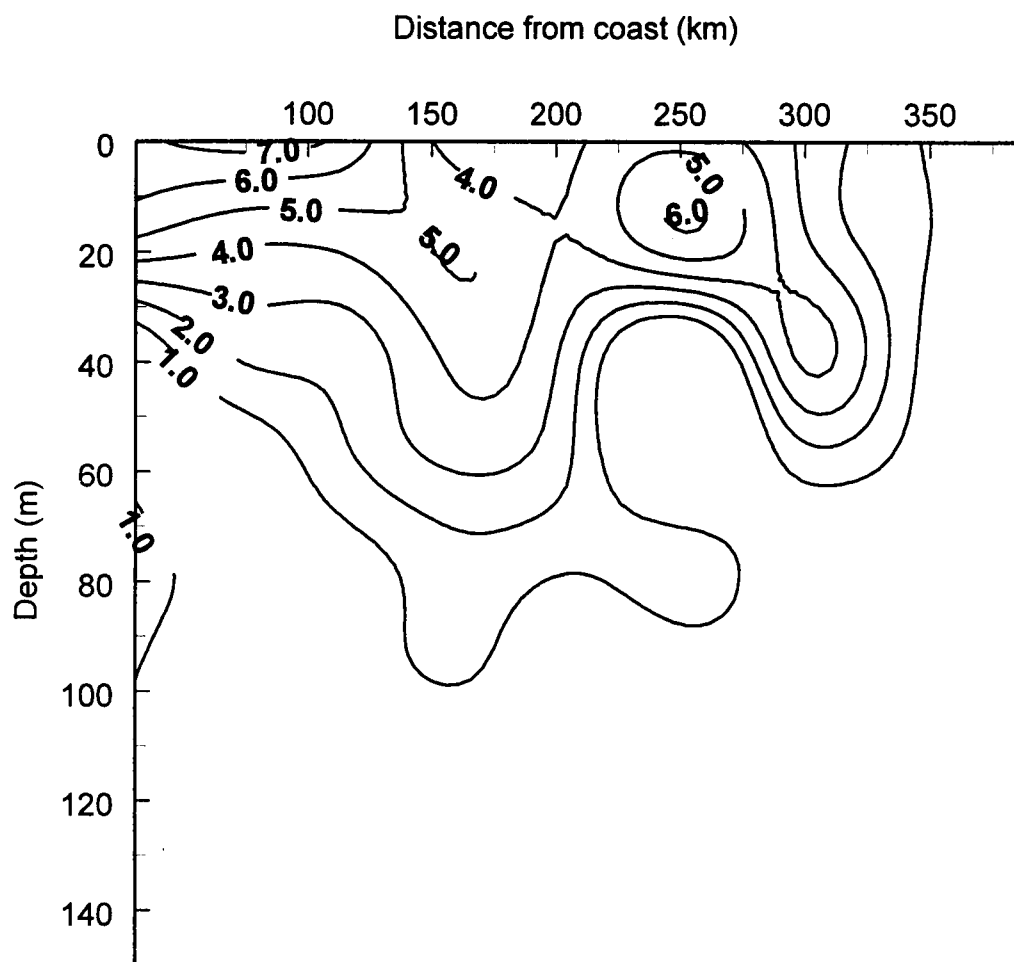


Figure 29. The vertical distribution of chlorophyll a concentrations across Transect 2 (at 173° E) in the Ross Sea

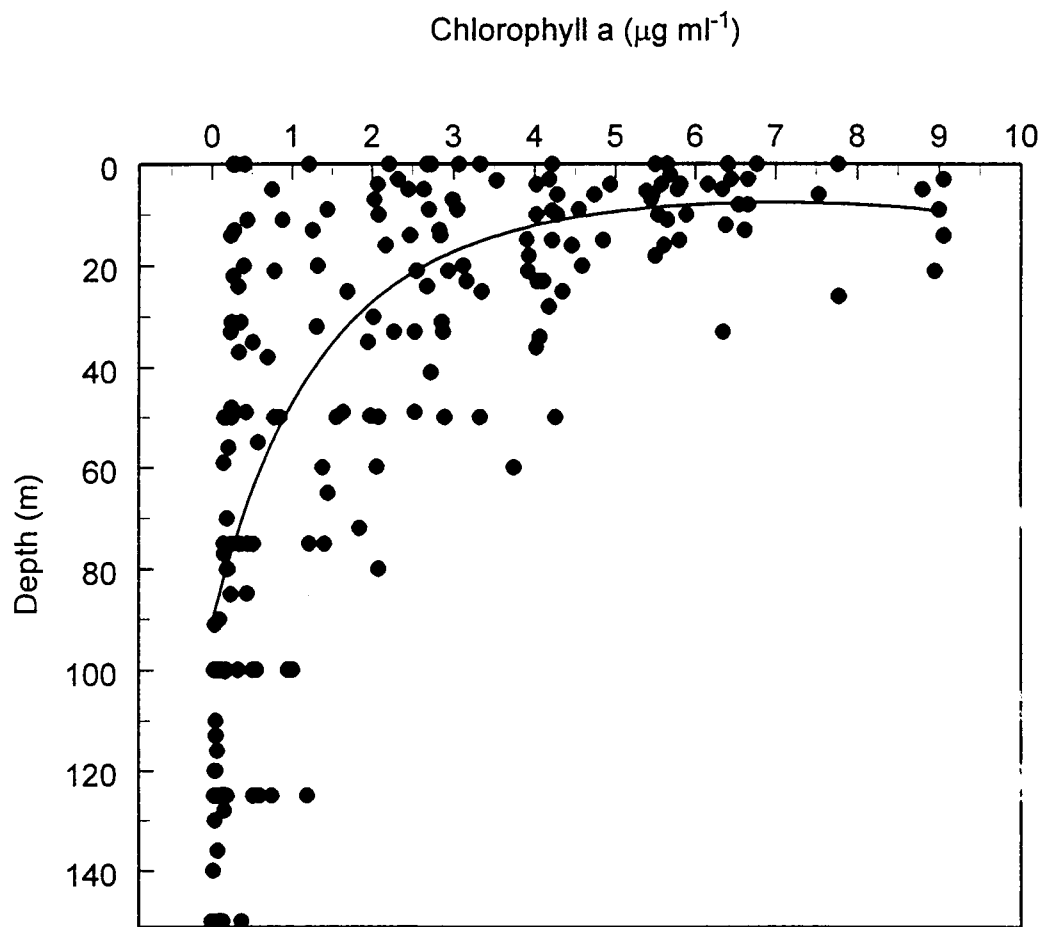


Figure 30. The vertical distribution of chlorophyll a concentrations at all stations.

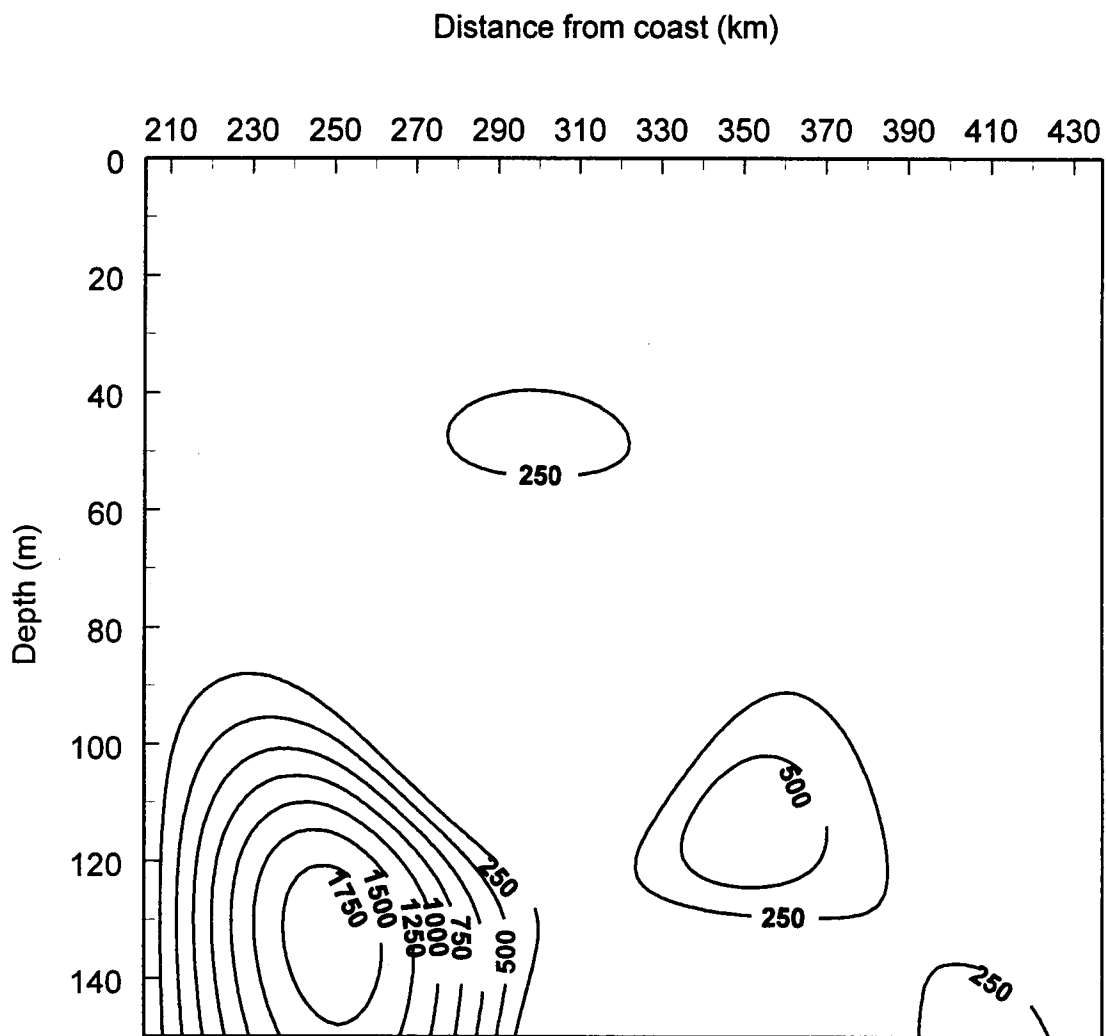


Figure 31. The vertical distribution of TEP/Chl a ratio across Transect 3 (at 75° S) in the Ross Sea

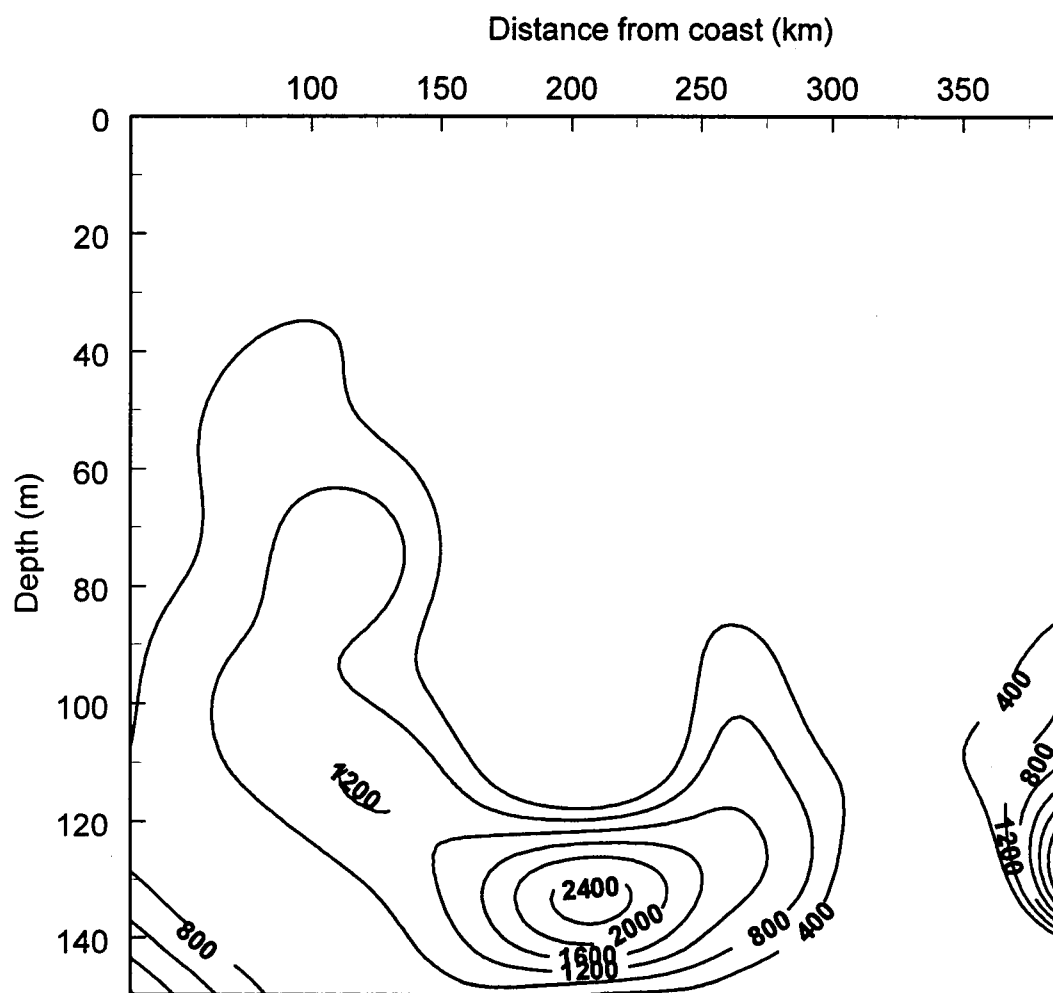


Figure 32. The vertical distribution of TEP/Chl a ratio across
Transect 2 (at 173° E) in the Ross Sea

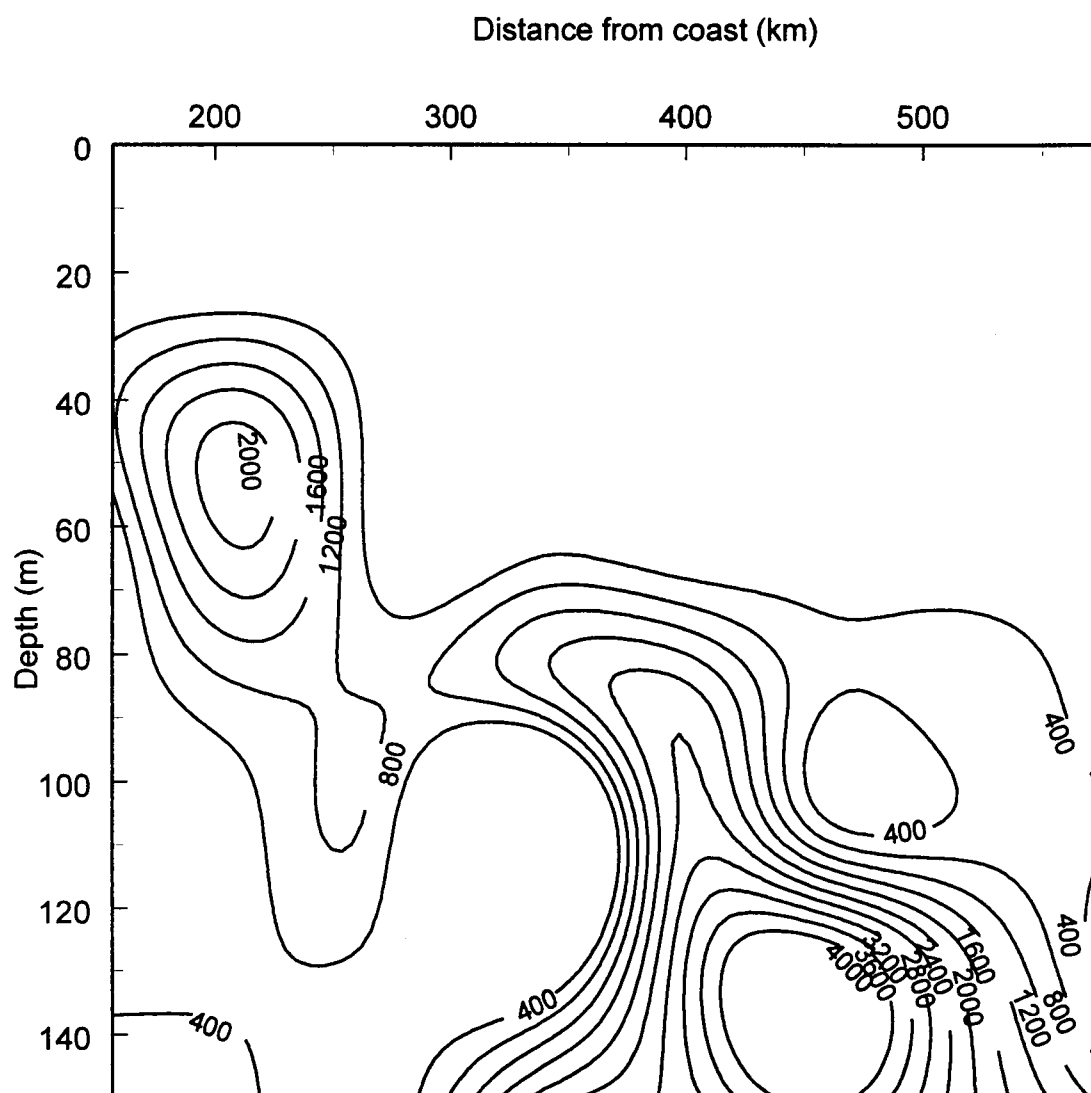


Figure 33. The vertical distribution of TEP/Chl a ration across Transect 1 (at 76° 33'S) in the Ross Sea

TEP vertical distributions varied at the same site. For example, Stations 38 and 47, which were dominated by diatoms, and sampled on Nov. 20 and 28, respectively, had distinctly different distributions of TEP (Fig. 34). Station 47 showed a peak TEP concentrations at 120 m, but the maximum of TEP at Station 38 was at 60 m and above. The stations dominated by *Phaeocystis* showed similar trends (Fig. 35).

Correlation analysis was conducted to determine if environmental parameters could account for observed variation in TEP. For the independent variables tested in a mixed (a layer of water that is mixed by wind and is therefore isothermal from 0-35 m), chlorophyll a closely correlated with TEP. It displayed a 2-state system in which TEP concentrations were low when chlorophyll a concentrations were below a certain threshold level. Above this threshold concentration, a rapid transition to the second state resulted in a large increase in TEP with increased chlorophyll a concentrations. The accumulative effective form of Weibull model αx^β describes the distribution of variables which accumulate through time (Casella and Berger, 1990). The change of TEP concentrations against chlorophyll a fit this kind of distribution, and α equals 1.014, β equals 3.632 (Fig. 36). TEP concentrations also showed a correlation with temperature and wind speed at the time of sampling. Because these two parameters were also correlated with chlorophyll a, they may indirectly impact TEP concentrations through the influence on the chlorophyll a.

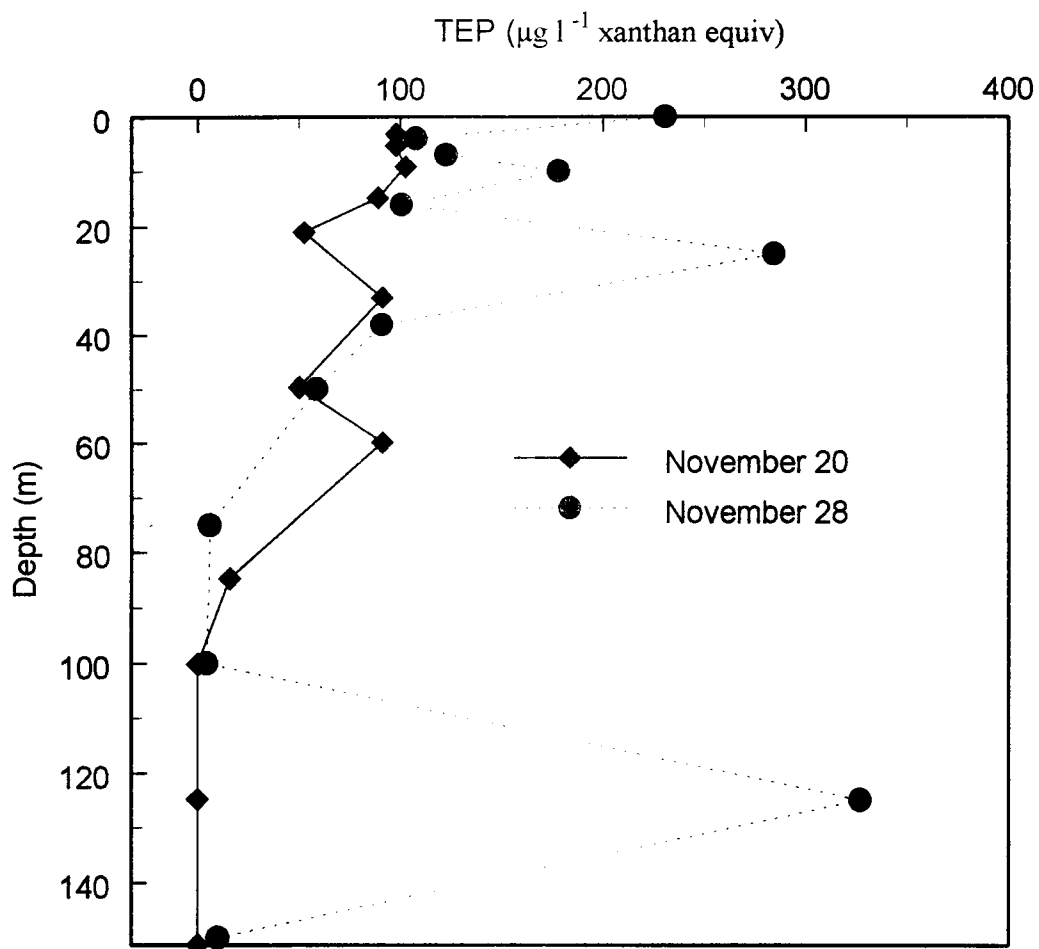


Figure 34. The vertical distribution of TEP concentrations at the station dominated by diatoms and surveyed at November 20 and 28.

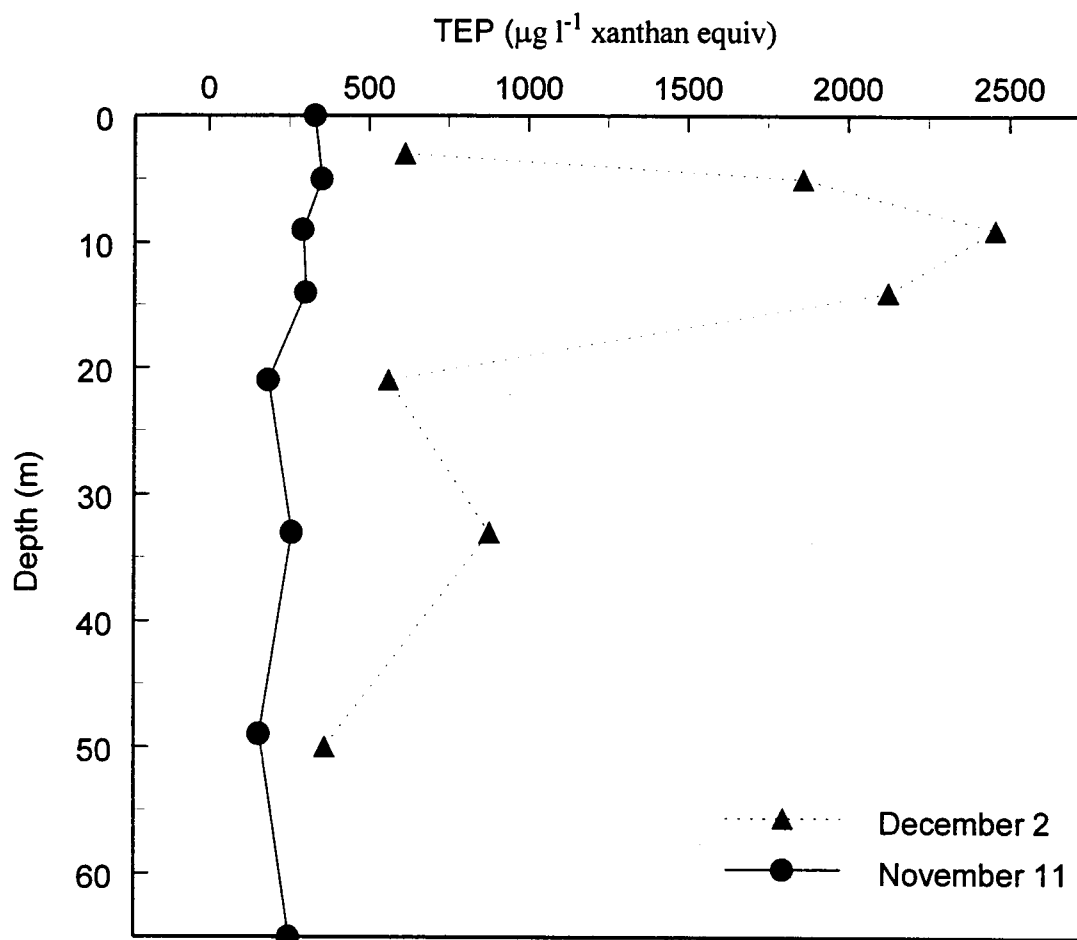


Figure 35. The vertical distribution of TEP concentration at the station dominated by *Phaeocystis* sp. on November 11 and December 2.

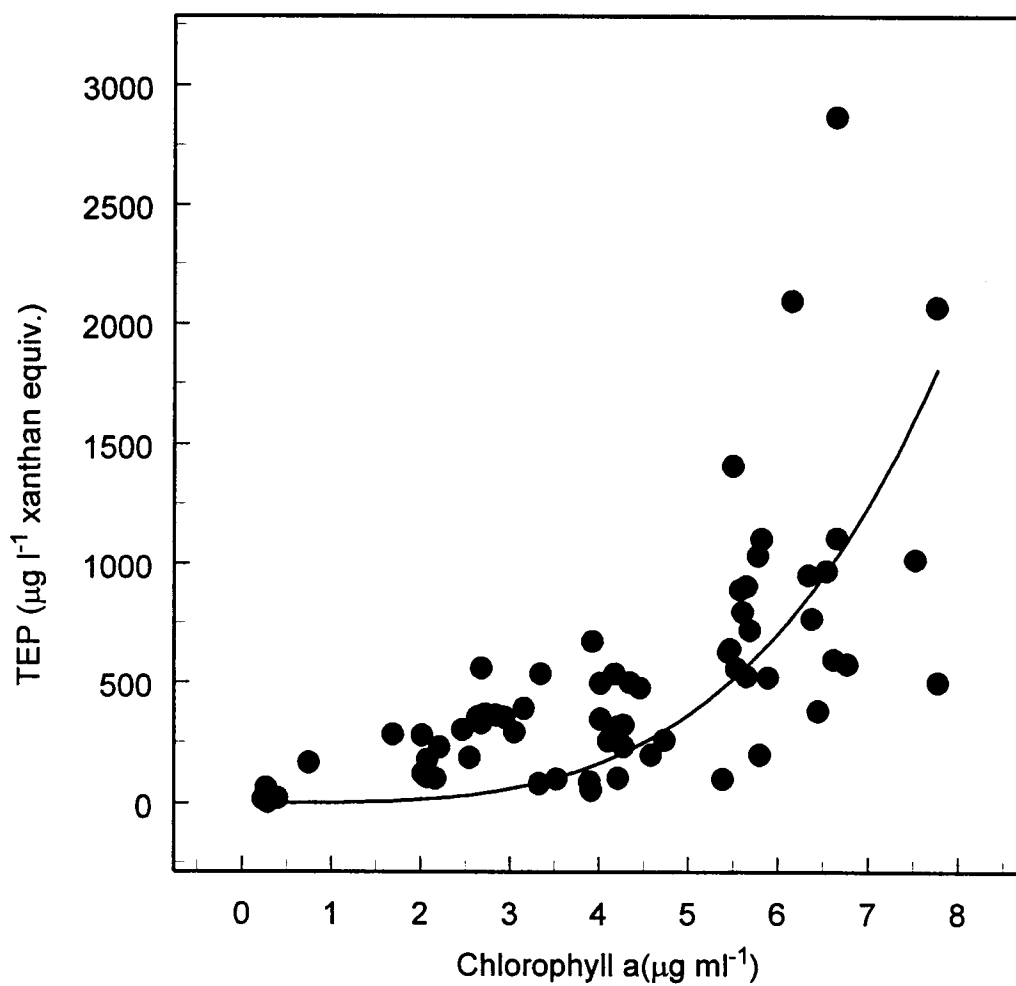


Figure 36. Relationship between TEP and Chlorophyll a concentration in the mixed layer. The model is $y = (\alpha x)^\beta$, and $\alpha = 1.01$; $\beta = 3.63$.

CHAPTER IV

DISCUSSION

TEP Produced by *Phaeocystis*

Transparent exopolymer particles (TEP), a new class of particles, were reported recently by Alldredge et al. (1993). Their quantities have been measured colorimetrically by Alldredge and Passow (in press) in the Pacific and Atlantic Oceans and in phytoplankton cultures. The values for natural waters ranged from 10 to 500 $\mu\text{g l}^{-1}$ xanthan equiv. No measurements have been conducted in the Ross Sea, and thus these data represent an important comparison to other regions which have been studied. TEP concentrations observed during this study were high when compared with values reported by Alldredge and Passow (in press), but most were within the range they found. In the Ross Sea TEP concentrations ranged from 100 to 1000 $\mu\text{g l}^{-1}$ xanthan equiv. in 94% samples and were less than 500 $\mu\text{g l}^{-1}$ xanthan equiv. in 80% of samples. The mean concentration was 308 $\mu\text{g l}^{-1}$ xanthan equiv.

The present study was conducted during a *Phaeocystis* bloom in the Ross Sea. Therefore, *Phaeocystis* might contribute to the high concentrations of TEP during the study. Passow and Wassmann (1994) found that when *Phaeocystis* colonies began to disintegrate during senescence, the fragments of the disrupted colonies were stainable, and these particles were considered to be TEP. This study indicates that *Phaeocystis* has a potential to produce high concentrations of TEP during bloom when *Phaeocystis* cells were not at the

High concentrations of TEP were also observed in *Phaeocystis* cultures during stationary phases. The average concentration of TEP was $4,590 \mu\text{g l}^{-1}$ xanthan equiv, and TEP concentrations ranged from 1,200 to $3,660 \mu\text{g l}^{-1}$ xanthan equiv. in 81% of samples. These levels were higher than those reported by Alldredge and Passow (in press), who observed an average concentrations of TEP in diatom cultures to be ca. $1000 \mu\text{g l}^{-1}$ xanthan equiv. The greater TEP concentrations observed in this study might be due to the fact that different salinities and irradiances were used, which clearly influenced growth.

The results of this study showed that irradiance strongly impacted TEP production, and this influence was closely related to growth status such as growth stage and photosynthetic performance. The response of TEP to irradiance during a short incubation was similar to that of the photosynthesis / irradiance relationship when *Phaeocystis* colonies were incubated under the same conditions. Long-term culture experiments also showed that much more TEP was produced by *Phaeocystis* when irradiance was high.

Salinity impacted TEP production and photosynthesis in a different fashion. It was observed that P_{max} and α were a function of salinity when salinity ranged from 28 to 36 ppt. Further experiments might consider salinity as a variable in the photosynthetic model. However, TEP production was not impacted by salinity in the range from 32 to 36 ppt, which was similar to both photosynthetic response and growth rate data. Furthermore, there was a synergistic effect between irradiance and salinity in terms of TEP production by *Phaeocystis*. When

between irradiance and salinity in terms of TEP production by *Phaeocystis*.

When irradiance was not limiting *Phaeocystis* growth, salinity had less influence on TEP production. When salinity was around 34 ppt, all three irradiances tested did not obviously influence TEP production. This may be one explanation for the lack of correlation between TEP and irradiance in the field investigation, where salinity varied little and was around 34 ppt.

The production of TEP may also be influenced by temperature and nutrients. Verity et al. (1986) reported that nutrient-stressed cells excreted more dissolved organic carbohydrate materials than did nutrient-replete cells. The multiple regression analysis showed that nitrate was a better predictor of TEP than any of other variables during the survey, and temperature was also highly correlated with TEP, although there was no linear relationship between them. How these two parameters influenced TEP was not clear because multiple regression analysis also suggested the large influence of these two parameters on chlorophyll a concentrations. Therefore, further experiments on the effects of temperature and nutrients on TEP production are needed.

Formation of TEP Produced by *Phaeocystis*

TEP produced by diatoms was suggested to be formed from dissolved carbohydrates because (1) TEP contain polysacharides; (2) the increases in TEP concentrations were always paralleled by a decrease in dissolved carbohydrate levels; and (3) TEP contain cation bridges (Alldredge et al., 1993, Passow et al., 1994). The mechanism of TEP formation by *Phaeocystis* was

considered to be fragments of the disrupted *Phaeocystis* colonies (Alldredge and Passow, 1993), and Passow and Wassmann (1994) observed that Alcian blue stainable polysaccharide particles (5 to 50 μm) were formed from dissolved carbohydrates derived from the colonial matrix of *Phaeocystis*. In our studies, when senescent *Phaeocystis* cultures were filtered through 0.2 μm filters, and the supernatant was stained with alcian blue after drying, a large amount of stainable material was observed in the supernatant and which also paralleled an increase in TEP concentrations (data not shown). Accordingly, dissolutions of matrices of senescent colonies might, at least in part, contribute to the accumulation in TEP of *Phaeocystis* dominated assemblages.

The formation of TEP has been suggested to occur through the process of aggregation or coagulation (Alldredge et al., 1993, Passow and Wassmann 1994, Passow et al., 1994). It was hypothesized that the dissolved polysaccharide aligned to form elongated bundles of organic macromolecules called fibrils (Alldredge et al., 1993). These fibrillar colloids further coagulate through the process of aggregation, or otherwise coalesce to form the larger TEP particles. The present studies suggest that TEP produced by *Phaeocystis* might involve the coagulation process.

It was observed that the relationship between TEP and chlorophyll a concentrations in the Ross Sea exhibited a two-state system (Fig.36) (e.g., the change of TEP was minimal at chlorophyll concentrations below 3 $\mu\text{g l}^{-1}$, but above this concentration a rapid increase in TEP with increasing chlorophyll a

concentrations was observed). Because dissolved carbohydrate is a function of chlorophyll and the two were positively correlated to *Phaeocystis* cell numbers (Eberlein, 1985), DOC should be an implicit function of TEP, and a two-state system might exist between DOC and TEP concentrations. This two-state system was proposed in the model of aggregate formation during algal blooms (Jackson, 1990). According to the coagulation theory (Jackson, 1990, Riebesell, 1991, Kiørboe et al., 1994), aggregation of suspended particles depends on the concentration of particles, the size of the particles, and the mechanisms responsible for particle collision, such as shear. Shear rates in the ocean depend on variety of factors, including wind stress and surface mixed layer depth. TEP concentrations in our study were related with wind speed. This might provide support for the hypothesis that the formation of TEP involves aggregation or coagulation. Further investigation on the relationship between TEP and DOC concentrations, as well as turbulent adherence rates, are needed to understand TEP production by *Phaeocystis*, and assessment of chlorophyll a in surface waters and other factors involving shear process may allow prediction of TEP concentrations.

Distribution of TEP Produced by *Phaeocystis*

The general distributional pattern of TEP in the Ross Sea was similar to that of phytoplankton biomass. TEP was found to be more abundant near the coast than in the central Ross Sea and more abundant in the mixed layer than below.

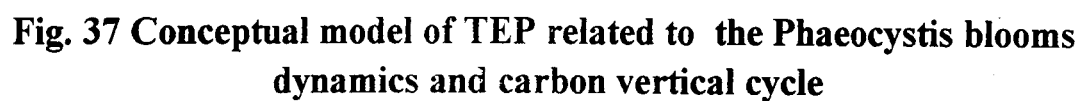
phytoplankton composition (Passow and Alldredge, 1994), in that no simple overall relationship between algal composition and TEP distribution was observed. Within Section I, which was dominated by *Phaeocystis*, maximum TEP values were found at or near the surface in some stations, and maximum TEP values in other stations were found near 120 m, whereas within transect II, which had a mixed assemblage of *Phaeocystis* and diatoms, TEP concentrations were high in surface water and decreased with depth (Fig. 23). In addition, a similar level of TEP concentration per chlorophyll in the stations dominated by *Phaeocystis* and diatoms, respectively, was observed (Fig. 21). It appears that TEP concentrations were related more to chlorophyll concentrations (i.e. phytoplankton biomass) than taxonomic composition in our limited survey, and the fact that TEP concentration maxima appeared in deep water in a later survey suggests that as the *Phaeocystis* cells grew and the bloom developed, vertical distribution of TEP changed. Further time series observations are needed to understand the vertical distribution of TEP in the Ross Sea.

Riebesell (1993) and Wassmann et al. (1990) observed that *Phaeocystis* attached to the large aggregates during a bloom, so that *Phaeocystis* colonies were suggested to be important for the biologically enhanced physical aggregation during vertical transport, which in turn gives rise to marine snow. However, Passow and Wassmann (1994) proposed that cells and colonies of *Phaeocystis* may not contribute significantly to vertical flux, but that the sedimentation of mucous flocs represents a secondary pathway for the flux of

sedimentation of mucous flocs represents a secondary pathway for the flux of photoassimilated carbon to depth. Because TEP was considered to contribute significantly to the formation mucous flocs (Alldredge et al., 1993, Passow and Alldredge 1994, Passow and Wassmann 1994, Passow et al., 1995), studies on the vertical distribution of TEP and *Phaeocystis* might help to understand the relationship between *Phaeocystis* and marine snow. These studies have demonstrated that there was a pronounced difference in the vertical distribution of TEP and chlorophyll a. During this study, water of the Ross Sea was repeatedly characterized by a surface chlorophyll a maximum. However, the TEP vertical distribution varied widely in the stations surveyed, and the ratio of TEP to chlorophyll a was high at depth in most of the stations. These results provide indirect evidence in support of the hypothesis that mucous flocs instead of *Phaeocystis* colonies contribute significantly to carbon vertical flux. Further studies on the relationship between TEP and marine snow in the Ross Sea are needed to quantify the role of TEP in the cycling and loss of carbon from the surface layer during *Phaeocystis* blooms.

Our experimental and field data may suggest the following sequence of events during *Phaeocystis* blooms in the Ross Sea: (1) During the early bloom (chlorophyll a concentrations $3 \mu\text{g l}^{-1}$ or less), few TEP were produced by *Phaeocystis*; (2) As biomass increased to the annual maximum (chlorophyll a concentrations greater than $10 \mu\text{g l}^{-1}$) accumulated dissolved carbohydrates caused the rapid increase of TEP concentrations; (3) During decline of the

bloom, a large amount of TEP will be produced by senescent *Phaeocystis* and the processes that enhance aggregation, such as shear, may also enhance the formation of TEP; and (4) The high abundance of TEP with elevated stickiness coefficients (Kjørboe and Hansen, 1993) contributes to flocculation, and flocs consisting of polysaccharide particles, bacteria and abiotic particles are formed. These flocs in turn provide a concentrated substrate for an increased metabolic activity of bacteria and may provide food particles for zooplankton and protozoan grazers; (5) Finally, large flocs sink and cellular detritus, inorganic particles as well as TEP are lost from the euphotic layer (Fig.37).



CHAPTER V

CONCLUSIONS

1. *Phaeocystis* sp. has the potential to produce transparent exopolymer particles (TEP) during blooms and senescent cultures, and TEP concentrations produced by *Phaeocystis* sp. were no less than those produced by diatoms.

2. TEP production was the function of growth rate and photosynthetic response, and was influenced by those factors which influence *Phaeocystis* growth. These studies indicated that both growth rate and TEP production were tightly correlated with irradiance and salinity, and these two factors were interacted with each other.

3. In the Ross Sea TEP production was a function of chlorophyll concentration, and was minimal at chlorophyll levels less than $3 \mu\text{g l}^{-1}$, but increased rapidly above this concentration.

4. In the Ross Sea TEP production may involve a coagulation mechanism, which is supported by the correlation between TEP concentration and wind speed. Those factors that correlated with chlorophyll a, such as, nitrate and temperature, were also correlated with TEP production.

5. Vertical distribution patterns of TEP and chlorophyll were different, which lead a maximum ratio of TEP to chlorophyll in the bottom layer at most of the stations investigated. When the results of two surveys at the same station are compared, higher TEP concentrations appeared at deep layer at the station

sampled later. This evidence supports the hypothesis that mucous flocs, which were tightly related with TEP, contribute significantly to carbon vertical flux.

6. Comparing the results of laboratory and field study, TEP concentration produced by *Phaeocystis* sp. were greater in cultures than those in the field. The results of experiments showed that TEP was influenced by salinity and irradiance, and the influence of these two factors were dependent on each other, which provides one possible explanation for the lack of correlation between TEP and irradiance in the field investigation, where salinity varied little and was around 34 ppt.

7. A conceptual model of TEP related to *Phaeocystis* bloom dynamics and carbon vertical cycles is proposed. Further studies are needed to confirm this model, such as the correlation between TEP and aggregates during *Phaeocystis* blooms and the relationship between TEP concentration and wind stress.

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APPENDICES

APPENDIX A

Cell numbers and chlorophyll a concentrations of *Phaeocystis* cultured at different salinities and irradiances

Time (hours)	Light ($\mu\text{E m}^{-2}\text{s}^{-1}$)	Salinity (ppt)	Fluorescence (arbitrary units)	Chl a ($\mu\text{g ml}^{-1}$)	Cell number (10^5 ml^{-1})
0	15	28	0.7	4.61	0.93
24	15	28	0.7	4.61	0.93
60	15	28	1.85	13.1	3.28
84	15	28	2.25	16.1	4.09
108	15	28	2.2	15.7	3.99
144	15	28	2.25	16.1	4.09
168	15	28	0.9	6.09	1.33
210	15	28	0.3	1.66	0.11
0	15	32	0.7	4.61	0.93
24	15	32	0.7	4.61	0.93
60	15	32	1.85	13.1	3.27
84	15	32	2.15	15.3	3.89
108	15	32	2	14.2	3.58
144	15	32	2.3	16.4	4.19
168	15	32	1.15	7.94	1.84
210	15	32	0.7	4.61	0.93
0	15	36	0.7	4.61	0.93
24	15	36	0.7	4.61	0.93
60	15	36	2.6	18.7	4.8
84	15	36	2.6	18.7	4.8
108	15	36	2.9	20.9	5.41
144	15	36	2.7	19.4	5
168	15	36	2.7	19.4	5
210	15	36	1.05	7.2	1.64
0	50	28	0.7	4.61	0.93
24	50	28	0.7	4.61	0.93
60	50	28	2.25	16.1	4.09
84	50	28	2.05	14.6	3.68
108	50	28	2.25	16.1	4.09
144	50	28	1.7	12	2.97
168	50	28	2.15	15.3	3.89

Time (hours)	Light ($\mu\text{E m}^{-2}\text{s}^{-1}$)	Salinity (ppt)	Fluorescence (arbitrary units)	Chl a ($\mu\text{g ml}^{-1}$)	Cell number (10^5 ml^{-1})
210	50	28	1.15	7.94	1.84
0	50	32	0.7	4.61	0.93
24	50	32	0.7	4.61	0.93
60	50	32	2.63	18.9	4.87
84	50	32	2.37	16.9	4.33
108	50	32	2.7	19.4	5
144	50	32	2.67	19.1	4.93
168	50	32	2.13	15.2	3.85
210	50	32	2	14.2	3.58
0	50	36	0.7	4.61	0.93
24	50	36	0.7	4.61	0.93
60	50	36	2.57	18.4	4.73
84	50	36	2.5	17.9	4.6
108	50	36	2.83	20.4	5.28
144	50	36	3.13	22.6	5.89
168	50	36	2.87	20.6	5.35
210	50	36	2.3	16.4	4.19
0	100	28	0.7	4.61	0.93
24	100	28	0.7	4.61	0.93
60	100	28	1.25	8.68	2.05
84	100	28	0.9	6.09	1.33
108	100	28	0.75	4.98	1.03
144	100	28	1.65	11.6	2.86
168	100	28	0.45	2.77	0.42
210	100	28	0.2	0.91	
0	100	32	0.7	4.61	0.93
24	100	32	0.7	4.61	0.93
60	100	32	1.57	11	2.69
84	100	32	1.27	8.8	2.08
108	100	32	1.17	8.03	1.88
144	100	32	1.9	13.5	3.37
168	100	32	0.95	6.46	1.44
210	100	32	0.63	4.12	0.79
0	100	36	0.7	4.61	0.93

Time (hours)	Light ($\mu\text{E m}^{-2}\text{s}^{-1}$)	Salinity (ppt)	Fluorescence (arbitrary units)	Chl a ($\mu\text{g ml}^{-1}$)	Cell number (10^5 ml^{-1})
24	100	36	0.7	4.61	0.93
60	100	36	2.23	15.9	4.05
84	100	36	2.53	18.2	4.67
108	100	36	2.87	20.6	5.35
144	100	36	2.27	16.2	4.12
168	100	36	1.5	10.5	2.56
210	100	36	0.73	4.86	0.99

APPENDIX B
Photosynthetic production of *Phaeocystis* cultured at different salinities

IN*	P _{max15}	P ^B _{max15}	P _{max25}	P ^B _{max25}	P _{max35}	P ^b _{max3}
1	0.75	0.03	2.25	0.07	1.83	0.069
1	1.02	0.04	2.02	0.07	1.86	0.07
1	0.91	0.03	2.44	0.08	2.15	0.08
5	7.51	0.28	15.5	0.51	14.8	0.56
5	5.97	0.22	14.1	0.46	17.9	0.67
5	6.84	0.26	15.3	0.5	15	0.56
15	11.4	0.43	25.6	0.84	30.3	1.14
15	12.1	0.46	27.9	0.91	34.8	1.31
15	12.5	0.47	26.1	0.85	32.1	1.21
30	9.79	0.37	18.5	0.6	27.3	1.03
30	9.12	0.34	22	0.72	23.8	0.9
30	9.44	0.356	20.4	0.67	26.6	1
50	11.2	0.42	26.1	0.85	29.3	1.1
50	12.1	0.46	25.6	0.84	31.7	1.19
50	11.5	0.43	27.4	0.89	32.2	1.2
100	12.8	0.48	29.7	0.97	32.8	1.23
100	14.5	0.55	29.4	0.96	324	1.21
100	12.7	0.48	27.4	0		

*Irradiance: $\mu\text{E m}^{-2} \text{s}^{-1}$

**P_{Max} and P: $\text{mg C (mg Chl)}^{-1} \text{h}^{-1}$

***subscript represent salinity (ppt)

APPENDIX C
The data of production vs irradiance of *Phaeocystis* sp.

irradiance ($\mu\text{E m}^{-2} \text{s}^{-1}$)	^{14}C (dpm)	P (mg C(mg Chl) $^{-1} \text{h}^{-1}$)	P _B (mg C(mg Chl) $^{-1} \text{h}^{-1}$)
0	106	0	0
0.16	409	636.9	1.501
0.27	689	1226	2.89
0.33	981	1839	4.333
0.38	1320	2561	6.036
0.48	1130	2158	5.087
0.59	1370	2658	6.265
0.6	1447	2819	6.643
0.61	1255	2415	5.692
0.62	1328	2568	6.053
0.63	1303	2518	5.934
0.79	1647	3240	7.63
0.85	1627	3198	7.537
0.86	1414	2750	6.481
0.88	1503	2938	6.924
0.88	1571	3080	7.259
0.9	1522	2977	7.016
1.1	1558	3051	7.192
1.2	1407	2736	6.448
1.3	1691	3332	7.853
1.3	1353	2622	6.179
1.6	1455	2835	6.681
1.7	1832	3629	8.552
1.8	1883	3735	8.803
2	1930	3835	9.039
2.3	1684	3316	7.816
2.5	1823	3610	8.509
2.5	1901	3773	8.893
2.6	1355	2625	6.187
4	1954	3885	9.155
5.7	1644	3233	7.62

APPENDIX D
TEP production in short-term incubation (4h)

Irradiance ($\mu\text{E m}^{-2}\text{s}^{-1}$)	TEP ($10^3 \mu\text{g l}^{-1}$ xanthan equiv)	TEP/Chl a (10^3 ng xanthan equiv. ($\mu\text{g Chl}$) $^{-1}$)
0.0	3.60	0.08
11.6	17.15	0.40
23.2	33.55	0.79
26.6	17.84	0.42
39.8	23.39	0.55
43.2	12.35	0.29
43.2	51.93	1.22
63.1	62.01	1.46
66.4	36.02	0.85
86.3	32.33	0.76
94.6	32.72	0.77
99.6	28.33	0.67
106.3	58.72	1.38
116.2	50.42	1.19
116.2	35.88	0.85
117.9	25.45	0.60
146.1	39.10	0.92
152.7	40.68	0.96
156.1	22.09	0.52
166.0	49.46	1.17
166.0	39.99	0.94
199.2	45.76	1.08
199.2	23.21	0.55
232.4	37.80	0.89
265.6	43.84	1.03
332.1	38.55	0.91
332.1	23.46	0.55
365.3	28.74	0.68
365.3	41.71	0.98

APPENDIX E

TEP production of *Phaeocystis* sp. Cultured at different irradiances and salinities

Time (hours)	Irradiance ($\mu\text{E m}^{-2} \text{ s}^{-1}$)	salinity (ppt)	TEP ($\mu\text{g l}^{-1}$ xanthan equiv.)	Chl a ($\mu\text{g ml}^{-1}$)	TEP/Chl (ng xanthan ($\mu\text{g Chl}$) $^{-1}$)
60	15	28	3104	13.59	228.3
108	15	28	1235	16.80	73.5
168	15	28	3464	4.90	707.2
210	15	28	3413	0.00	0.0
60	15	32	2590	13.59	190.5
108	15	32	3053	14.97	203.9
168	15	32	4956	7.19	689.6
210	15	32	4459	3.07	1453.0
60	15	36	3499	20.46	171.0
108	15	36	2504	23.21	107.9
168	15	36	11730	21.38	548.8
210	15	36	4751	6.27	757.5
60	50	28	3001	17.26	173.9
108	50	28	1201	17.26	69.6
168	50	28	9398	16.34	575.1
210	50	28	10580	7.19	1472.0
60	50	32	4459	20.77	214.7
108	50	32	4207	21.38	196.8
168	50	32	2927	16.19	180.8
210	50	32	5945	14.97	397.2
60	50	36	3018	20.16	149.8
108	50	36	2367	22.60	104.7
168	50	36	3579	22.90	156.3
210	50	36	6986	17.71	394.4
60	100	28	2178	8.10	268.8
108	100	28	6980	3.53	1979.9
168	100	28	11420	0.78	14660.0
210	100	28	5488	0.00	0.0
60	100	32	2550	11.00	231.8
108	100	32	2127	7.34	289.7

168	100	32	5362	5.36	1001.0
210	100	32	4482	2.46	1824.0
60	100	36	3030	17.10	177.1
108	100	36	2115	22.90	92.4
168	100	36	3407	10.39	327.9
210	100	36	9467	3.37	2807.0

APPENDIX F

TEP concentration at the sampling station of the Ross Sea

Station	Depth (m)	TEP ($\mu\text{g l}^{-1}$ xanthan equiv)	Chl a ($\mu\text{g ml}^{-1}$)	TEP/Chl (ng xanthan equiv ($\mu\text{g Chl}$) $^{-1}$)
7	0	331.3	2.68	123.6
7	5	354.2	2.64	134.2
7	9	294.8	3.05	96.6
7	14	303.9	2.47	123.0
7	21	187.3	2.55	73.4
7	33	260.5	2.27	114.7
7	49	159.8	1.64	97.5
7	65	253.6	1.45	174.9
7	80	214.7	0.20	1073.6
7	100	139.3	0.18	773.7
7	125	125.5	0.03	4184.6
7	150	169.0	0.04	4224.6
9	3	170.8	2.32	73.6
9	5	107.4	2.45	43.8
9	9	21.6	1.44	15.0
9	13	158.8	1.25	127.0
9	21	71.3	0.78	91.5
9	33	47.3	0.24	197.2
9	50	62.8	1.56	40.2
18	31	0.0	0.25	0.0
18	90	0.0	0.10	0.0
18	113	0.0	0.06	0.0
18	120	0.0	0.04	0.0
18	130	0.0	0.04	0.0
18	140	10.0	0.02	500.8
18	150	0.0	0.03	0.0
19	0	287.9	5.50	52.3
19	4	299.3	4.94	60.6
19	6	496.0	5.43	91.3
19	9	294.8	4.55	64.8
19	15	127.8	4.85	26.4
19	23	148.4	4.03	36.8

Station	Depth (m)	TEP ($\mu\text{g l}^{-1}$ xanthan equiv)	Chl a ($\mu\text{g ml}^{-1}$)	TEP/Chl (ng xanthan equiv ($\mu\text{g Chl}$) $^{-1}$)
19	34	102.7	4.07	25.2
22	3.2	98.1	3.53	27.8
22	5.3	98.1	5.39	18.2
22	9.2	102.7	4.22	24.3
22	14.8	89.0	3.90	22.8
22	21	52.4	3.92	13.4
22	33	91.2	2.53	36.1
22	50	50.1	1.98	25.3
22	60	91.2	2.06	44.3
22	85	15.8	0.44	35.9
22	100	0.0	0.17	0.0
22	120	0.0	0.15	0.0
22	150	0.0	0.08	0.0
23	0	0.0	0.41	0.0
23	11	0.0	0.44	0.0
23	20	19.6	0.40	49.0
23	31	26.5	0.36	73.6
23	49	0.0	0.43	0.0
23	75	0.0	0.45	0.0
23	113	72.9	0.05	1458.9
25	0	33.3	1.21	27.6
25	11	16.9	0.88	19.2
25	20	26.5	1.32	20.1
25	32	0.0	1.31	0.0
25	50	1.0	0.78	1.3
25	77	0.0	0.15	0.0
25	116	0.0	0.07	0.0
31	0	316.2	4.22	74.9
31	4	347.1	4.02	86.3
31	6	261.4	4.74	55.1
31	10	498.0	4.02	123.9
31	16	477.5	4.41	107.1
31	25	501.5	4.35	115.3
31	36	196.2	4.02	48.8
31	50	247.6	3.03	0.1
31	75	62.4	1.40	44.6
31	100	103.6	0.95	109.0

Station	Depth (m)	TEP ($\mu\text{g l}^{-1}$ xanthan equiv)	Chl a ($\mu\text{g ml}^{-1}$)	TEP/Chl (ng xanthan equiv ($\mu\text{g Chl}$) $^{-1}$)
31	125	161.9	0.11	1471.8
31	150	41.8	0.07	597.8
36	0	244.2	3.07	79.5
36	7	203.1	2.99	67.9
36	13	79.6	2.83	28.1
36	20	189.3	3.12	60.7
36	31	161.9	2.86	56.6
36	50	316.2	2.90	109.1
36	72	261.4	1.84	142.0
37	0	577.6	6.77	85.3
37	3	381.0	6.45	59.1
37	5	952.6	6.34	150.3
37	8	970.9	6.54	148.5
37	13	595.9	6.62	90.0
37	20	198.0	4.59	43.1
37	30	280.3	2.02	138.8
37	50	115.7	0.85	136.1
37	75	234.6	1.21	193.9
37	100	367.2	1.01	363.6
37	125	316.9	0.60	528.2
37	150	143.1	0.07	2044.9
38	0	556.3	6.41	86.8
38	2	721.0	5.69	126.7
38	4	892.5	5.58	159.9
38	6	642.1	5.47	117.4
38	10	560.1	5.54	101.1
38	15	199.6	5.80	34.4
38	23	391.7	3.16	124.0
38	50	137.9	0.18	766.0
38	75	69.3	0.52	133.2
38	100	76.1	0.33	230.7
38	125	4.1	0.19	21.7
38	150	100.2	0.11	910.5
39	0	3.2	0.29	10.9
39	14	15.5	0.24	64.6
39	24	12.8	0.33	38.7
39	37	0.4	0.34	1.2

Station	Depth (m)	TEP ($\mu\text{g l}^{-1}$ xanthan equiv)	Chl a ($\mu\text{g ml}^{-1}$)	TEP/Chl (ng xanthan equiv ($\mu\text{g Chl}$) $^{-1}$)
39	48	0.0	0.25	0.0
39	59	0.0	0.15	0.0
39	70	0.0	0.19	0.0
39	91	0.0	0.04	0.0
39	110	0.0	0.05	0.0
39	120	0.0	0.05	0.0
39	136	0.0	0.08	0.0
39	150	0.0	0.05	0.0
45.1	0	34.7	0.27	128.6
45.1	13	0.0	0.28	0.0
45.1	22	60.8	0.27	225.1
45.1	35	30.6	0.51	60.0
45.1	55	99.2	0.58	171.0
45.1	85	0.0	0.24	0.0
45.1	128	41.6	0.16	259.8
47.1	0	230.9	2.21	104.5
47.1	4	107.8	2.07	52.1
47.1	7	122.5	2.03	60.4
47.1	10	178.2	2.08	85.7
47.1	16	100.6	2.17	46.3
47.1	25	284.4	1.69	168.3
47.1	38	90.96	0.7	130
47.1	50	58.61	0.25	234
47.1	75	5.87	0.15	39.3
47.1	100	4.528	0.09	50.3
47.1	125	326.9	0.09	3630
47.1	150	10.01	0.09	111
53	0	906.2	5.65	160
53	4	1105	5.82	190
53	7	628.4	5.45	115
53	11	526.7	5.65	93.2
53	18	1410	5.5	2560
53	28	312.8	4.18	74.8
53	41	281.9	2.73	103
53	60	165.3	1.38	120
53	80	148.2	0.19	780
53	100	41.85	0.12	349

Station	Depth (m)	TEP ($\mu\text{g l}^{-1}$ xanthan equiv)	Chl a ($\mu\text{g ml}^{-1}$)	TEP/Chl (ng xanthan equiv ($\mu\text{g Chl}$) $^{-1}$)
53	125	576.9	0.14	4120
53	150	508.3	0.14	3630
56.1	0	79.58	3.33	23.9
56.1	3	535.8	4.18	128
56.1	6	323.1	4.28	75.5
56.1	10	233.9	4.28	54.7
56.1	15	261.3	4.22	61.9
56.1	23	257.9	4.11	62.8
56.1	35	336.8	1.95	173
56.1	56	158.4	0.21	755
56.1	75	65.85	0.35	188
56.1	100	38.4	0.03	1280
56.1	125	96.72	0.04	2418
56.1	150	631.8	0.03	21060
60.1	0	366.7	2.73	134
60.1	5	166	0.75	221
60.1	9	364	2.7	135
60.1	14	364	2.84	128
60.1	21	355.7	2.94	121
60.1	33	210.1	2.88	73
60.1	49	173.6	2.53	68.6
60.1	60	919	3.74	246
60.1	80	365.6	2.08	176
60.1	100	208.2	0.56	372
60.1	125	88.95	0.75	119
60.1	150	66.08	0.06	1100
64.1	3	614.2	9.06	67.8
64.1	5	1860	8.8	211
64.1	9	2457	9	273
64.1	14	563.2	9.06	62.2
64.1	21	2124	8.95	237
64.1	33	878.7	6.35	138
64.1	50	364.2	4.26	85.5
70.1	0	2069	7.77	266
70.1	4	2096	6.16	340
70.1	6	1018	7.53	135
70.1	8	2869	6.66	431

Station	Depth (m)	TEP ($\mu\text{g l}^{-1}$ xanthan equiv)	Chl a ($\mu\text{g ml}^{-1}$)	TEP/Chl (ng xanthan equiv ($\mu\text{g Chl}$) $^{-1}$)
70.1	12	768.9	6.38	121
70.1	18	673.6	3.93	171
70.1	25	536.5	3.35	160
70	5	1035	5.78	179
70	10	522.7	5.89	88.7
70	16	797.1	5.61	142
70	24	559.3	2.68	209
70	50	170.6	0.16	1070
70	75	225.5	0.25	902
70	100	124.9	0.07	1780
70	125	266.7	0.52	513
70	150	152.3	0.37	412
77	3	1108.6	6.66	166
77	26	501.2	7.78	64.4
77	50	477.5	2.08	230
77	100	330	0.51	647
77	125	247.6	1.19	208
77	150	271.7	1.35	201

VITA

Hong Ying was born in Lanzhou City, China, on September 15, 1960. She attended primary schools in Ningbo city, and graduated from The Third High Schools of Lanzhou, Lanzhou in August 1976. She began studies at Nanking University, Nanking in September 1978, and graduated from Nanking University in August 1982, having earned a Bachelor of Science degree with a major in Botany. She worked as a Research assistant in Institution of Biology, Gusu Academy of Sciences from September 1982 to August 1984. In September 1984, she began work towards a Master of Science degree in Institution of Hydrobiology, Chinese Academy of Sciences. She received her degree in August 1987. From September 1987 to December 1992 she worked as a lecturer in Jinan University at Guangzhou, China. In January 1993, she began graduate studies in the Department of Botany at The University of Tennessee, Knoxville, working under graduate research assistantships or graduate assistantships until receiving a Master of Science degree in May 1996.