

## **Primary and Extracellular Production during Austral Summer at 70°S, 12°E, Antarctica**

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### **Abstract**

Much of the  $^{14}\text{C}$  radioactivity supplied as  $\text{NaH}^{14}\text{CO}_3$  accumulates in the low molecular weight fraction of the particulate matter, into polysaccharides and proteins. There was little radioactivity in the lipid component. Phytoplankton exudation showed a significant decline with greater depth within the euphotic column. The quality of material exuded also varied. In some cases, a greater amount of aminoacids was released whereas in other cases organic acids formed the greater fraction. These data are discussed in relation to physical oceanographic factors.

### **Introduction**

Earlier studies have shown that Antarctic phytoplankton excrete from 1 to 30% of assimilated carbon as extracellular products (Horne *et al.*, 1969). Recent studies on marine phytoplankton near the ice-edge at 70°S, 12°E during an austral summer show that several low molecular weight metabolites are exuded by natural assemblages of Antarctic phytoplankton and that past physiological history of sampled populations may determine the nature of exuded material in these colder waters as it does in other regions of the world oceans. This hypothesis is discussed with reference to biomass estimated as chlorophyll *a* and ATP, growth rates of phytoplankton and primary and extracellular product determinations.

The data presented here was collected during the Fifth Indian Expedition to Antarctica, 1985-1986.

### **Material and Methods**

Seawater samples were collected using 5 litre Niskin samplers from 5 depths within the euphotic zone, and this zone was estimated on the basis of Secchi disc readings and occasional quanta measurements with a LiCor quantameter with deck and submersible cells.

Chlorophyll *a* was measured on a Turner<sup>(R)</sup> fluorimeter using 90% acetone extracts of phytoplankton contained in 1 litre or less of seawater filtered through auto Whatman<sup>(R)</sup> GF/C papers.

ATP content of 1 litre or less seston material filtered through a 0.45 $\mu$  porosity sterile Millipore<sup>(R)</sup> membrane paper and extracted with Tris buffer was determined using the Luciferin-Luciferase system on an SAI<sup>(R)</sup> Photometer. ATP and chlorophyll *a* were determined on board as were pH and alkalinity in accordance with techniques in Strickland and Parsons (1972).

Carbon-assimilation was measured using radioactive bicarbonate (Steeman-Nielsen, 1952) in 1 ml of 5 $\mu$ Ci NaH<sup>14</sup>CO<sub>3</sub> per 100 ml sample of seawater. Incubations were done in pumped surface seawater in a deck incubator for a minimum of 6 hours although in most cases samples were incubated for 24 hours. All incubations were done in triplicate. After incubation samples were filtered onto 0.45  $\mu$  porosity membrane filters. Duplicates from each set were put into well-stoppered glass vials with 1 ml each of distilled water and methanol and stored at -20°C for later analysis of particulate and products. The third set of filter papers, one from each depth, was put into Beckman Unisolv<sup>(R)</sup> scintillation cocktail for subsequent <sup>14</sup>C determination. No dark correction has been made in any of the values expressed as  $\mu$ gC/l/hr although background has been subtracted. Radioactive disintegrations were measured in a Beckman<sup>(R)</sup> automatic sample changer standardized by the channels ratio method.

Products of particulate carbon-assimilation were assayed by successive extractions of filtered material in chloroform, methanol and trichloroacetic acid to determine lipid, low molecular weight water soluble compounds, polysaccharides and structural proteins in the four fractions by the method of Li *et al.* (1980). All results presented are the average of duplicate determinations.

Following filtration, acidification of filtrate to pH 4 and bubbling with air for 45 minutes to remove inorganic <sup>14</sup>C, the filtrates were neutralized back to pH 8 and separated into three fractions successively on cation, anion and charcoal columns eluted with NH<sub>4</sub>OH, HCl and ethanol in accordance with methods published elsewhere (Shailaja and Pant, 1986). 5 ml aliquots of each eluate were freeze-dried and the radioactivity determined as above. All dpm values have been expressed as  $\mu$ gC/l/hr or  $\mu$ gC/Chl *a*/hr. Only those dpm values which were significantly above background have been used in these calculations.

### Results and Discussion

The correlation between chlorophyll *a* and Adenosine triphosphate (ATP) is positive and significant ( $r^2 = 0.67$ , Fig. 1) and most of the ATP is of phytoplankton origin although the *y*-intercept in Fig 1 suggests that bacterial ATP also forms a substantial part of the extracted material. The vertical distribution of organisms shows a clear sub-surface peak in both biomass and productivity (Fig.2) and appears to be a consequence of inhibiting surface light intensity.

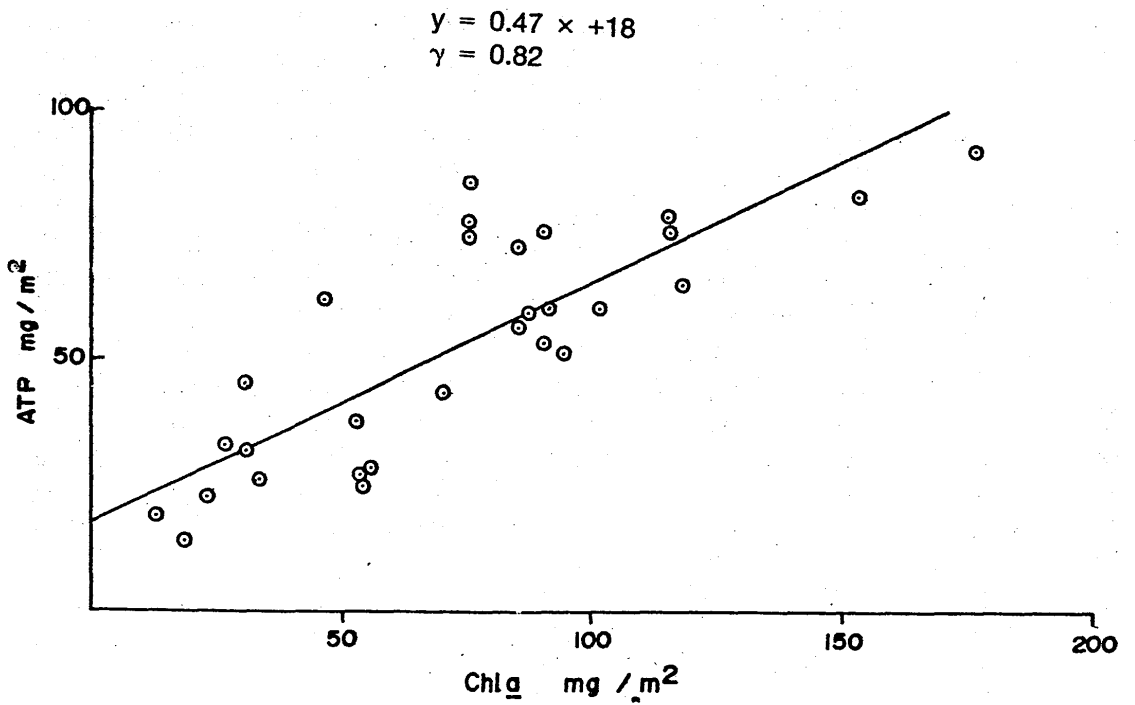


Fig. 1. Relationship of chlorophyll a and ATP at a single station off the ice edge in an austral summer.

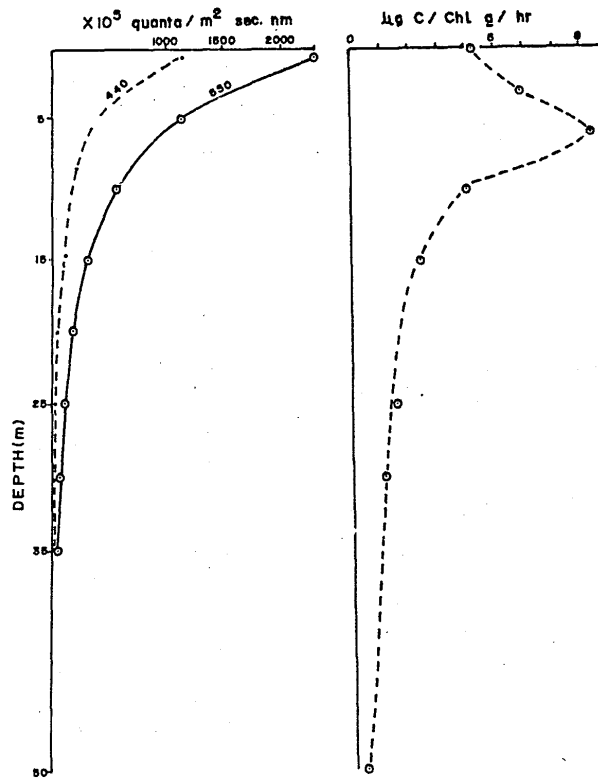
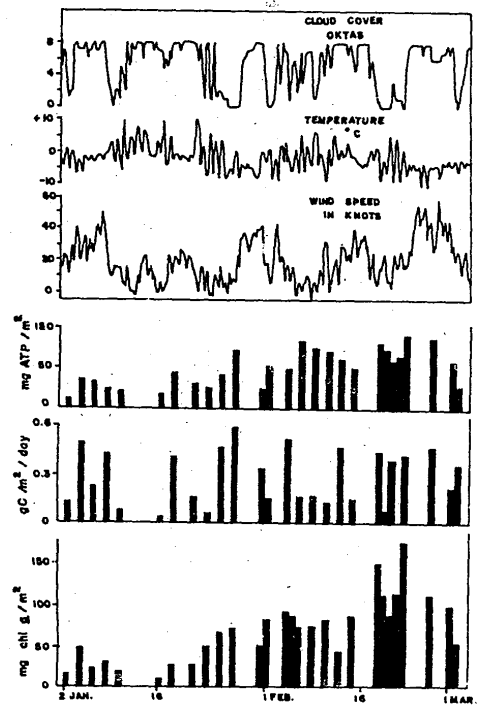


Fig. 2. Vertical profile of assimilation number in comparison to disappearance of light at two wave lengths.

There is a large day to day variation in both biomass and productivity near the ice-edge. To some extent this variation is related to local weather, particularly wind-velocity and (Fig. 3) cloud cover. Lateral drift of water and associated phytoplankton patchiness may also result in observed biomass fluctuations. The values of chlorophyll *a* vary between 10 mg/m<sup>2</sup> to 190 mg/m<sup>2</sup> and primary production from 0.05 to 5gC/m<sup>2</sup>/day between 2nd January and 2nd March, 1986 and massive blooms of the kind reported by Mandelli and Burkholder (1966) or El-Sayyed (1971) in Bransfield straits and Bellingshausen Sea were not observed during this period in this sector of the Antarctic ocean. The mean generation time calculated from specific growth rate ( $\mu$ ; Eppley, 1972) using a C:Chl *a* ratio of  $30 \pm 10$  (Steele, 1962) for nutrient-rich water is about 4 days, the range being 2.5 to 9 days. As compared to tropical phytoplankton with generation times between 1.5 to 3 days the growth in Antarctic waters is slow. However, as discussed by Pant (1986) the rapid changes in local weather, wind-driven circulation of the water and the depth of the well-mixed layer which reaches 200m in the water column will influence activity of phytoplankton by determining the time they spend in the euphotic layers with a consequent effect on productivity rates and generation times. Thus *in situ* generation times may be different from the values presented here.

Fig. 3. Effect of climate on production at 70°S-12°E during January to March 1986. (Euphotic zone)

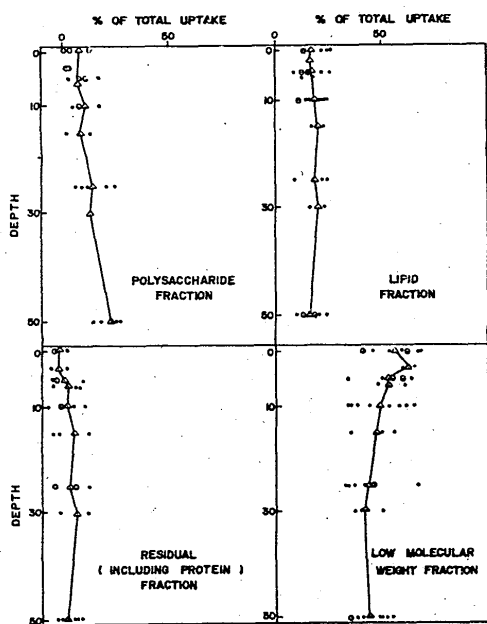


Size analysis of phytoplankton populations (Table I) indicates that organisms smaller than 20  $\mu$  contribute 30 to 80% of total primary production near the ice-edge. Brockel (1985) has shown the importance of nannoplankton in the Antarctic ecosystem and the results presented here agree with earlier findings.

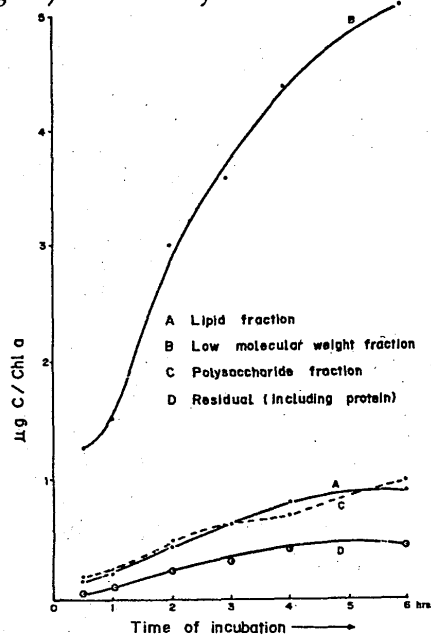
**Table I. Studies on fractionation by filtration of Antarctic marine phytoplankton productivity.**

Date	Whole	<50 $\mu$	% Total	<20 $\mu$	% Total
4 Feb	1.1	0.4	36	0.28	25
8 Feb	0.35	0.2	57	0.12	34
14 Feb	0.70	0.81	100	0.59	84
26 Feb	1.86	1.23	66	0.59	32
2 Mar	0.95	0.53	56	0.4	42

Smith and Morris (1980) have suggested that at low temperatures lipids may accumulate in the end-products of phytoplankton photosynthesis. Several experiments at the ice-edge showed, however, that accumulation of radioactivity in the lipid fraction (Fig. 4) is not significantly higher than similar data obtained from the tropical Arabian Sea (Goes and Devassy, 1986) and a greater percentage of inorganic  $^{14}\text{C}$  ends up in the low molecular weight methanol fraction at the termination of a 4 hours incubation with  $\text{NaH}^{14}\text{CO}_3$  than in any other fraction. Representative results of time-course studies (Fig. 5) show very similar results.



**Fig. 4. Percent of total radioactive uptake in different fractions**



**Fig. 5. Time course of  $\text{NaH}^{14}\text{CO}_3$  uptake into particulate matter by phytoplankton**

The rate of accumulation of  $^{14}\text{C}$  in the low molecular weight fraction is also much faster than that in the other products. These results do not substantiate the hypothesis of Smith and Morris (1980). On the contrary, they suggest that short-term products of Antarctic phytoplankton photosynthesis are similar to those of temperate or tropical phytoplankters.

Extracellular products vary in both quality and quantity at each time of sampling. In general, however, as shown in Fig. 6 there is a decline in amount of extracellular matter produced at increased depths and the marked sub-surface peak in particulate production is not repeated in the exuded products. There is, however, a measurable radioactivity in all the components segregated by ion-exchange chromatography and recovery in all three fractions combined in terms of  $^{14}\text{C}$  is about 85%. In most cases (representative examples 27th Jan, 18th Feb;

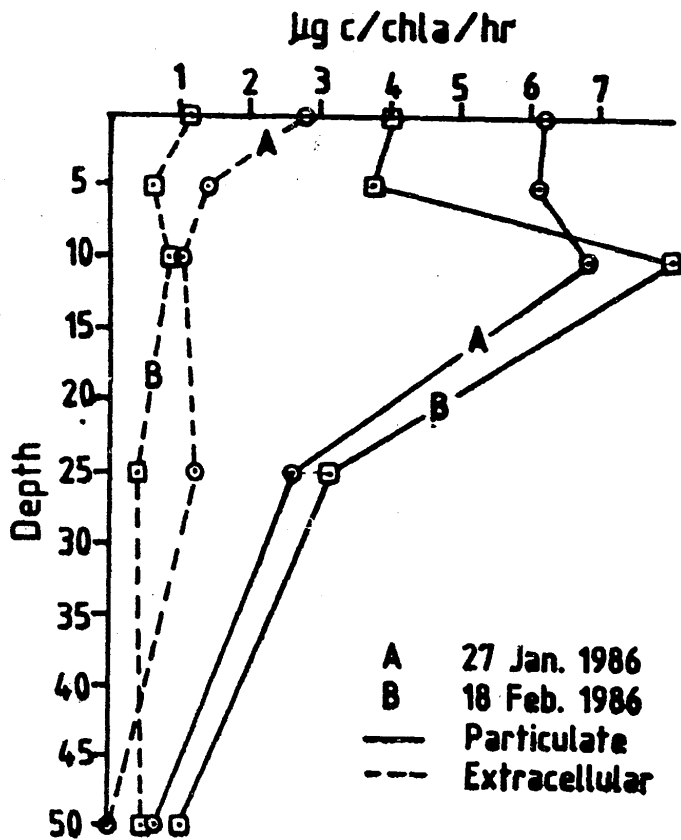
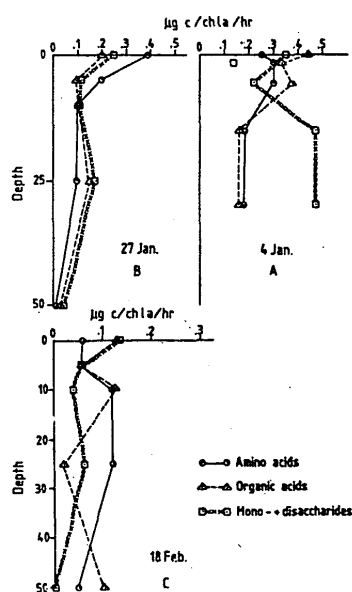


Fig. 6. Vertical profile of particulate and extracellular production at an ice-edge station, 70°S - 12°E

Fig. 7) show more or less similar decrease with depth except in a few instances (4th Jan; Fig. 7) when radioactivity in the mono- and disaccharide fractions increased at greater depth. On the basis of available information it is not possible to assign any unique reason for this observation. For example, chlorophyll *a* values in the water column on 4th Jan and 27th Jan are not significantly different (Fig. 3) although on the 18th Feb chlorophyll *a* was higher and in all three cases the chlorophyll *a*: phaeopigment ratios are in excess of one indicating a "healthy" population. Several workers have shown that some organisms in culture (Guillard and Wangersky, 1958; Hellebust, 1965; Myklestad, 1974) or decaying blooms (Ittekkot *et al.*, 1981) release polysaccharides. Physiological states of sampled populations may thus determine both quality and quantity of extracellular matter in colder Antarctic waters as they do in other environments.

It is interesting to note in the present series that of the total four time-course studies conducted between 2 Jan to 2 March 1986, the two most dissimilar sets of results (Fig. 8) occur on the 19th and 22nd of February. Although there is a gradual increase in extracellular products with time on both days, fractionation by ion-exchange shows a greater amount of radioactivity in mono- and disaccharides on the 19th February when particulate production was also low, than in the similar fraction on the 22nd February where carbon-assimilation was high. On the latter day organic acids formed a larger percentage of exuded matter, particularly at the end of 6 hours. Percentage extracellular release (inset, Fig. 8) on both days was also very different, from an average 50% on the 19th to 20% on the 22nd of February. Both the initial samples were collected one metre below the surface and chlorophyll *a* contents were similar though not identical. One possible explanation for this dissimilarity in quantities may be the physiological

Fig. 7. Results of ion-exchange separation of extracellular products into amino acid, organic acid and mono- & disaccharide components after 4 hr simulated *in situ* incubations.



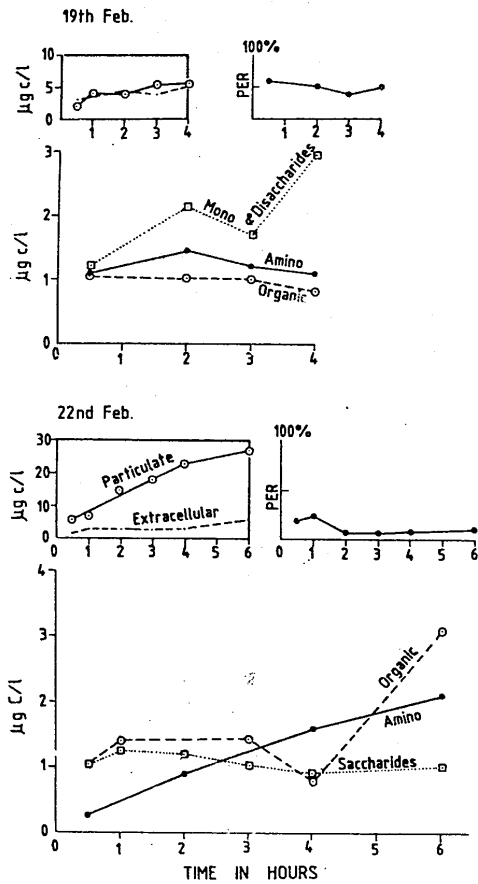


Fig. 8. Ion-exchange fractionation of extracellular products from two representative time-course studies. Insets present particulate and total extracellular data for the same experiments.

PER = Percentage extracellular release

history of the sampled populations. The rapidly changing local weather and the observed movement of ice floes near the ice-edge suggest that sampling intervals of even 24 hours may result in data from different populations. One method of determining physiological states of environmental phytoplankton may be by analysis of exudates.

The data on extracellular products of phytoplankton in the Antarctic also suggest that considerable energy transfer from phytoplankton to bacterioplankton occurs via the exudate pathway. Such bacterial dependence on phytoplankton has been convincingly demonstrated in other oceanic and estuarine regions (Nalewajko *et al.*, 1976; Bell and Sakshaug, 1980; Jensen, 1983) and bacterial uptake of low molecular weight reduced carbon compounds originating from Antarctic phytoplankton cannot be ruled out. It would be of considerable interest to explore further relationships between algae-bacteria, particularly those in ice-algae.



### Acknowledgements

I thank my colleagues both on board M.V. Thuleland and at NIO for the help given to me. I am grateful to the late Dr. H.N. Siddique, Director, NIO for permission to participate in the Expedition and to Dr. S.Z. Qasim, Secretary, DOD for the opportunity.

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