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The composition and bacterial utilization of DOC released by phytoplankton

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Abstract

The primary production, quantity and molecular weight distribution of DOC released by phytoplankton, and the subsequent utilization and transformation of these compounds by bacteria in an estuary was studied. High primary production rate and DOC release was observed. The amount and composition of DOC released by phytoplankton varied with time and changing algal population. Low molecular weight products with MW less than 500, fraction with MW 10 000 – 30 000, and high molecular weight compounds greater than 300 000 dominated in the algal DOC. Bacteria utilized a significant portion of released DOC. The role of bacteria in the transformation of released DOC by algae is discussed.

Introduction

The potential ecological significance of photosynthetically produced and released dissolved organic carbon (DOC) in secondary production, and the pathway of its return to the particulate phase of the food web have been discussed by several authors (POMEROY 1974; NALEWAJKO 1977). One of the significant pathways of this DOC return to the food web is bacterial utilization of algal extracellular products. Heterotrophic use of released DOC represents not only a portion of energy flow in the environment, but also represents a fraction of particulate organic carbon (POC), which is used as a food source for higher trophic levels.

Release of DOC can be of importance in investigating quantitative aspects of primary production of phytoplankton, as well as trophic relationships in waters (CHRÓST 1978, CHRÓST and SIUDA 1978).

The purpose of the present study was to characterize the primary production, quantity, and molecular weight distribution of DOC released by phytoplankton, and the subsequent utilization and transformation of these compounds by bacteria.

Material and methods

The studies were performed *in situ* at Rhode River estuary. The Rhode River is a tidal subestuary on the western shore of Chesapeake Bay, south of Annapolis, Maryland, USA. The estuary has a length of 6.7 km, a mean depth of 2.0 m, and a volume of 349 x 10⁴ m³ at mean low tide.

Water samples were taken 20 cm below the surface of the estuary, and incubated *in situ* at the sampling site.

Primary production, and extracellular release of DOC by phytoplankton were determined weekly with the use of ^{14}C -bicarbonate (New England Nuclear) according to CHRÓST and FAUST (1981).

An Amicon Corp. ultrafiltration stirred cell, model 202, and Diaflo ultrafiltration membranes, with nominal MW cutoffs in parentheses; UM 05 (500), UM 2(1 000), UM 10 (10 000), PM 30 (30 000), XM 50 (50 000), XM 100A (100 000), XM 300 (300 000), were used for the fractionation and estimation of the molecular weight (MW) of ^{14}C -DOC released by algae in 0.2 μm filtrates, as previously described by CHRÓST and FAUST (1980).

Bacterial utilization and transformation of algal DOC were measured according to CHRÓST and FAUST (1981).

Total viable counts of bacteria were estimated by the epifluorescence microscopy method of ZIMMERMANN and MEYER-REIL (1974).

Table 1

Range of temperature, salinity, dissolved oxygen content, chlorophyll a and phaeophytin at the site of experiments in Rhode River estuary (in parenthesis mean values for month)

Month 1978-79	Temperature °C	Salinity ppt	Oxygen mg/l	Chlorophyll a $\mu\text{g/l}$	Phaeophytin $\mu\text{g/l}$
October	13.5-15.0 (14.2)	14.5-14.8 (14.6)	8.7-8.8 (8.7)	14.75-22.40 (16.25)	0.15-4.60 (1.15)
November	7.0-13.0 (10.4)	13.0-14.0 (13.4)	8.4-9.3 (8.9)	7.19-16.85 (12.49)	1.43-10.17 (4.79)
December	2.0-9.5 (4.3)	12.5-14.5 (13.6)	11.2-12.8 (12.1)	9.38-43.40 (19.66)	0.0-0.71 (2.04)
January	1.8-2.0 (1.7)	7.2-8.4 (7.7)	13.2-15.0 (14.2)	6.36-24.02 (12.95)	0.0-2.19 (1.06)
February	1.0-2.0 (1.3)	6.0-8.7 (7.9)	12.5-15.0 (14.0)	4.35-9.81 (6.18)	0.0-1.52 (0.81)
March	2.0-12.5 (9.6)	2.8-5.6 (3.7)	11.8-12.5 (12.1)	2.18-63.33 (26.18)	0.0-2.59 (0.31)
April	10.5-16.5 (12.6)	3.2-4.2 (3.8)	10.0-12.0 (10.5)	5.32-42.86 (34.89)	0.0-4.46 (2.04)
May	17.0-20.5 (19.3)	4.4-5.3 (4.6)	6.0-10.8 (8.5)	8.19-97.19 (38.44)	0.0-5.28 (1.03)
June	20.0-25.0 (22.9)	4.0-5.8 (5.4)	9.0-10.0 (9.6)	37.12-328.96 (165.31)	0.15-1.24 (0.35)
July	24.5-25.0 (24.5)	6.0-6.8 (6.6)	7.0-8.0 (7.6)	18.56-169.26 (56.85)	0.0-0.87 (0.12)
August	24.5-29.0 (26.4)	6.0-6.8 (6.3)	6.2-7.2 (6.4)	23.47-54.05 (39.24)	0.0-0.89 (0.11)

Chlorophyll a and phaeophytin were determined daily according to GOLTERMAN and CLYMO (1971).

Temperature, salinity, and dissolved oxygen content in water were monitored continuously during the study period with recording instruments.

Results and discussion

The Rhode River is a tidal estuary, which receives runoff from its watershed primarily through Muddy Creek, the largest fresh water source. The brackish water enters the system from the Chesapeake Bay of Atlantic Ocean. The runoff, as well as the tides affect the physico-chemical properties of this environment.

Table 1 presents the range of temperature, salinity, dissolved oxygen content, chlorophyll a and phaeophytin in the water during the study period. The salinity of water was higher in the fall and winter, and varied from 6.0 to 14.5 ‰.

Unusually high rainfall during the spring and summer of 1979 resulted in low salinity of water with fluctuation from 2.8 to 6.8 ‰. Dissolved oxygen varied from 6.0 to 15.0 mg O₂/l.

A higher concentration of oxygen in the period December to March was observed. In spring and summer the fluctuations of dissolved oxygen were higher, but the general tendency of decreasing oxygen content with increased water temperature was observed. The water temperature varied from 1.0°C in winter to 29.°C in August.

Chlorophyll a concentration varied strongly with sampling time. During the late fall and winter the chlorophyll a content varied from 4.35 to 43.40 µg/l, and the phytoplankton community had a more diverse population composed of small flagellates, cryptophytes, chrysophytes and dinoflagellates. In December, an algal bloom occurred, and was mainly composed of *Skeletonema costatum*, *Prorocentrum* sp., and *Synedra* sp. In March, *Chlamydomonas vernalis* bloom was observed (63.33 µg/l of chlorophyll a).

From May to August almost permanent algal blooms occurred. The mean chlorophyll a content varied from 38.44 to 165.31 µg/l. The phytoplankton community was dominated by *Katodinium rotundatum* (May), *Gymnodinium nelsoni*, *Gymnodinium estuariale*, *Katodinium* sp. (June), *Cryptomonas* sp., *Calycomonas* sp., *Prorocentrum* sp. (July), and very small dinoflagellates with size less than 7 µm (August). Phaeophytin concentration in water varied constantly, and fluctuated from 0 to 10.17 µg/l. This indicates that the dead algae did not strongly affect the properties of the water.

High primary production rate ranged on the average from 44.75 to 285.7 µgC/l/h, except January and February, see Table 2. The amount of DOC released by algae varied with time and changing phytoplankton populations. Low release rate of DOC by algae ranging on the average from 1.50 to 4.95 µgC/l/h, from January to April, was observed. Excluding this period of studies, the release rate of DOC varied in average from 17.6 to 96.26 µgC/l/h, and constituted from 7.91 to 46.07 % of total carbon fixed in photosynthesis.

In previous studies on release of DOC by natural populations of phytoplankton, an inverse relationship between productivity, chlorophyll or biomass of algae, and the PER was observed (ANDERSON and ZEUTSCHEL 1970; RYTHER et al. 1971; CHRÓST and WAZYK 1978). Our results gave no indication of such an inverse relationship between release of DOC, POC production, and chlorophyll concentration. It is possible that also the phytoplankton composition affected the photosynthetic fixation of carbon

Table 2

Range of primary production rate and extracellular release of DOC by phytoplankton (in parenthesis mean values for month)

Month 1978–79	Primary production ($\mu\text{gC/l/h}$)			PER
	POC	DOC	Total	
October	39.50–54.60 (52.27)	28.55–50.65 (44.66)	69.15–100.21 (96.93)	46.07
November	45.80–58.90 (54.60)	27.40–45.65 (38.75)	76.15–101.17 (93.30)	41.53
December	8.80–46.50 (24.50)	16.30–35.40 (20.25)	25.34–75.42 (44.75)	45.25
January	3.10–3.60 (3.40)	1.30–1.80 (1.50)	4.40–5.40 (4.90)	30.61
February	3.00–5.85 (4.70)	1.40–2.65 (1.89)	4.45–8.15 (6.60)	28.63
March	45.15–147.10 (128.20)	4.60–6.65 (4.95)	51.20–152.10 (133.10)	3.71
April	16.20–63.40 (58.27)	2.75–4.46 (4.01)	18.90–66.45 (62.30)	6.43
May	59.10–161.60 (90.24)	40.15–69.72 (50.41)	110.20–224.80 (140.60)	35.85
June	84.72–155.75 (141.23)	44.26–58.76 (47.24)	130.60–205.40 (188.50)	25.06
July	116.90–227.80 (189.48)	40.05–123.70 (96.26)	157.40–302.40 (285.70)	33.69
August	128.60–272.50 (204.89)	6.80–18.30 (17.60)	134.60–280.50 (222.50)	7.91

POC – Particulate Organic Carbon, DOC – Dissolved Organic Carbon,
PER – Percentage of Extracellular Release

and its subsequent release. One of the most important factors regulating this process in the Rhode River estuary is light penetration depending on turbidity of water.

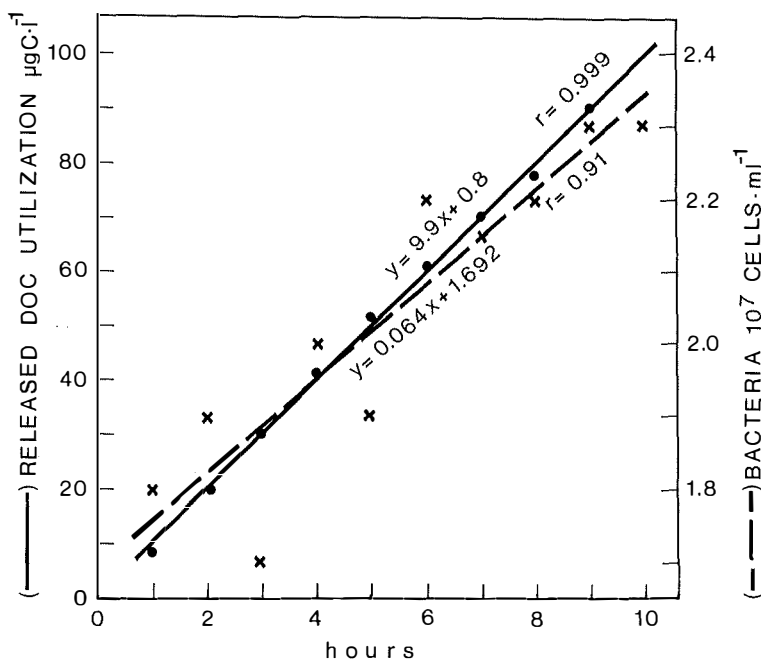
An ultrafiltration procedure for the fractionation and molecular weight study of released DOC was used. The data from these experiments are shown in Table 3. Molecular weight distribution of DOC released by algae varied with different composition of algal species in phytoplankton community. However, 3 fractions dominated in released DOC after 4 hours of photosynthesis, i.e. low molecular weight fraction of DOC with MW less than 500 (14.6 – 31.1 %), fraction with MW 10 000 – 30 000 (26.8 – 47.3 %), and high molecular weight compounds with MW greater than 300 000 (4.6 – 31.0 %).

Simultaneously with the determination of DOC released by phytoplankton, its utilization by bacteria was studied.

Table 3

Molecular weight (MW) distribution of DOC released by phytoplankton (mean values for month)

Month	% DOC released in various MW fractions							
	< 500	500– 1 000	1 000– 10 000	10 000– 30 000	30 000– 50 000	50 000– 100 000	100 000– 300 000	>300 000
1978–79								
October	26.4	2.8	4.8	33.4	10.9	6.2	1.8	13.7
November	30.0	4.3	2.5	39.2	13.4	5.2	0.8	4.6
December	14.9	2.7	2.2	47.3	10.0	2.0	3.6	17.3
January	28.5	12.6	8.4	30.2	1.4	1.8	0.8	16.3
February	16.4	8.4	5.2	28.4	4.6	5.9	6.2	24.90
March	14.6	5.7	7.4	31.4	19.2	4.2	2.4	15.1
April	23.3	14.0	6.4	33.2	1.7	4.3	2.1	15.0
May	15.6	1.4	1.3	27.5	20.4	15.8	4.4	13.6
June	16.7	4.4	5.8	34.8	5.3	4.9	8.7	19.4
July	18.0	3.1	0.8	30.9	8.2	6.4	1.6	31.0
August	31.1	12.3	4.7	26.8	2.7	5.8	3.1	13.5
average	21.4	6.5	4.5	33.0	8.9	5.7	3.2	16.8

**Figure 1**

The kinetics of bacterial utilization of DOC released by phytoplankton.

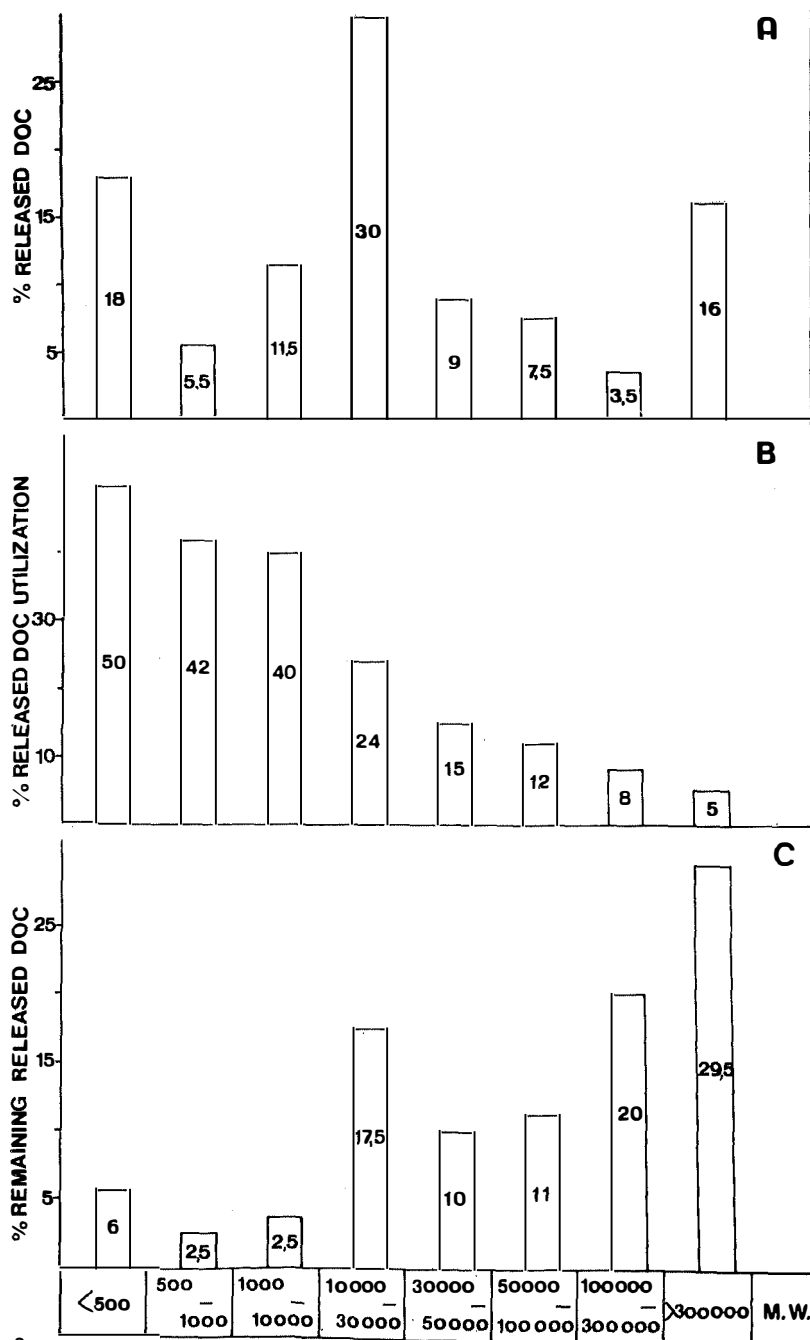


Figure 2

Bacterial utilization and transformation of DOC released by phytoplankton; A – MW composition of DOC released by phytoplankton (% of total release), B – bacterial utilization of different MW fractions of DOC released by phytoplankton (% of the released MW fraction), C – MW composition of remaining algal DOC after 10 hours of bacterial utilization (% of remaining release).

Figure 1 presents the kinetics of bacterial utilization of released DOC. The initial amount of algal DOC available for bacteria was 165 $\mu\text{gC/l}$. After 10 hours of bacterial utilization the amount of DOC decreased to 65 $\mu\text{gC/l}$, i.e. 60.7 % of the total available algal DOC was utilized. The utilization rate of released DOC was linear for 10 hours. It was estimated that bacteria took and retained in the cells approximately 40 % of utilized DOC. The second portion, 60 % of utilized DOC, bacteria respired in the same time. An analysis of changes in quantity of bacteria showed an increase of total number of bacterial cells with time. Bacteria utilized the released algal DOC as a source of nutrients and energy for their growth and metabolism.

Figure 2 presents changes in molecular weight distribution of DOC released by phytoplankton after its bacterial utilization. Organic compounds with MW less than 10 000 were preferentially utilized by bacteria. The utilization rate of different MW fractions of released DOC decreased with the increase in their molecular weight. The remaining DOC, after bacterial utilization, was composed of high molecular weight compounds. The composition of DOC after these experiments showed a high level of organic products with MW of 100 000 – 300 000, and the fraction with MW greater than 300 000, which constituted 20 % and 29.5 % of remaining DOC. These fractions in the released algal DOC accounted for 3.5 % and 16.0 %, respectively. Bacteria took up a large amount of the lower MW-fractions and in consequence the remaining DOC consists of a greater percentage of high molecular fractions. Thus the bacteria might be indirectly responsible for the predominance of high molecular weight organic compounds in water.

The presented results show a close relationship between phytoplankton and bacteria in the food chain. The release of DOC by phytoplankton is a direct link between autotrophic and heterotrophic processes in aquatic environments.

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