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Ecological investigations of blooms of colonial *Phaeocystis pouchetti*. II. The role of life-cycle phenomena in bloom termination

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Abstract. A bloom of the colonial stage of the prymnesiophyte *Phaeocystis pouchetii* was studied for 2 months in a 13-m^3 flow-through mesocosm. *Phaeocystis* increased in abundance for 6 weeks coincident with declining temperature and nutrient supply rates. Experiments suggested that colony growth was primarily nitrogen-limited during this period. An extended period of subzero temperatures and nutrient deprivation was associated with a mass exodus of cells from the colonies. Previously non-motile cells developed flagella, became motile and emigrated out of the colonies, accompanied by significant decreases in the chlorophyll *a* content and photosynthetic rates of the colonies. Concentrations of bacteria on the surfaces of such 'ghost' colonies were two orders of magnitude higher than on 'normal' colonies. Growth rate studies of field populations indicated that rapid declines in temperature induced development of motility and emigration from the colonies. Ancillary observations implied that chronic nutrient deprivation resulted in similar life-cycle events. Warming and nutrient addition did not halt release of swarmers, suggesting that, once initiated, the process proceeds to completion. The combined data indicate that blooms of colonial *Phaeocystis*, unlike many other phytoplankton, are not necessarily terminated by grazing or sinking out of the euphotic zone. The physiological option of motility and emigration provides *Phaeocystis* with an ecological alternative which has significant implications in interpreting the structure and function of plankton communities.

Introduction

A significant problem in understanding marine plankton ecology is the unknown role of life-cycle phenomena in the inception, duration and termination of plankton blooms. Several phytoplankton groups alternate between motile and non-motile vegetative states, the former flagellated, the latter often palmelloid or coccoid (i.e. vegetative cysts). These life-cycle phases regulate the temporal and spatial occurrence of such species, e.g. coccolithophorid (Lefort, 1975) and red-tide dinoflagellate blooms (Anderson *et al.*, 1983).

Phaeocystis pouchetii, a prymnesiophyte famous for prodigious blooms first documented more than a century ago (see Gran, 1902), has a remarkable life-cycle. It has at least two different plankton phases, perhaps a benthic phase, and is capable of rapid vegetative growth in each stage (Kornmann, 1955; Kayser, 1970). *Phaeocystis* occurs most prominently in the plankton as a large gelatinous, colonial aggregation of non-motile cells (3–8 μm). Thousands of cells can occur in colonies up to 10 mm in diameter *in situ* and in culture (Gieskes and Kraay, 1975; Verity *et al.*, 1988). In its other planktonic stage, *Phaeocystis* occurs as an asexual solitary cell of 3–6 μm diameter. These single swarmers may persist indefinitely in the plankton, form new colonies and may also be released from older colonies. There may also be other stages of solitary cells. In addition to asexual zoospores which can develop into colonies,

Kornmann (1955) described macrospores and microspores which he suspected to be gametes. Although in culture they behaved like asexual swarmers, neither regenerated the colonial phase, either alone or when mixed together. Kayser (1970) also described a sessile stage, in which single cells attached to surfaces, replicated themselves, and released planktonic swarmers.

The formation of colonies from solitary cells and the subsequent emigration of cells from the colonies has been observed in culture (Kornmann, 1955; Kayser, 1970; Parke *et al.*, 1971), but the induction and regulation of these processes is poorly understood. Numerous environmental factors have been invoked as bloom triggers, including temperature (Jones and Haq, 1963); decreased concentrations of silicate and phosphate (Jones and Spencer, 1970; van Bennekom *et al.*, 1975; Veldhuis and Admiral, 1987); trace metals (Morris, 1971); and edaphic effects (Jones and Haq, 1963). Conclusive evidence to support these hypotheses is lacking (Cadee and Hegemann, 1986; Weisse *et al.*, 1986). Moreover, factors influencing the emigration of cells out of colonies have not been determined, and the role of this life-cycle phenomenon in the terminating blooms of colonial *Phaeocystis* is poorly understood. This paper describes our observations of the fate of a winter bloom of *Phaeocystis* and potential regulatory mechanisms. It is part of a larger investigation of the *in situ* dynamics of this enigmatic species (Verity *et al.*, 1988; T.J.Smayda *et al.*, in preparation).

Methods

Phaeocystis pouchetti (Hariot) Lagerheim bloomed from late November to late January in one of the land-based mesocosms (tank 1) on Narragansett Bay, Rhode Island (41°30'N, 71°23'W). The tanks are scaled to the natural environment in terms of mixing, temperature and flushing rates (Pilson, 1985; Oviatt *et al.*, 1986). During this study, unfiltered seawater was fed into tank 1 during 12-min pulses every 6 h, equivalent to a 4% daily exchange, using a diaphragm pump shown to be non-destructive to plankton. Automatic vertical plungers ensured chemical homogeneity after 10 min of mixing. Substantial populations of *Phaeocystis* also developed in four other mesocosms (tanks 2, 4, 6, 8) from January through April (Verity *et al.*, 1988). These tanks differed from tank 1 in that they contained 36 cm of intact soft-bottom sediments from lower Narragansett Bay, and their temperatures were regulated by glass heat exchangers. This study focuses on events in tank 1, in which temperature was not controlled (Figure 1). *Phaeocystis* from tank 2 were used in one experiment to document further the role of temperature shocks in life-cycle processes. Additional details on tank operation are given in Verity *et al.* (1988). Data on chemical and biological variability among tanks and comparisons to Narragansett Bay are described elsewhere (Nixon *et al.*, 1984; Oviatt *et al.*, 1986).

Various physical, chemical and biological parameters were measured at regular intervals during the 2-month bloom. Surface irradiance was recorded continuously. Light attenuation, using a Secchi disk (Holmes, 1970), and concentrations of NH_4, $NO_3 + NO_2$, PO_4 and $Si(OH)_4$, using a Technicon

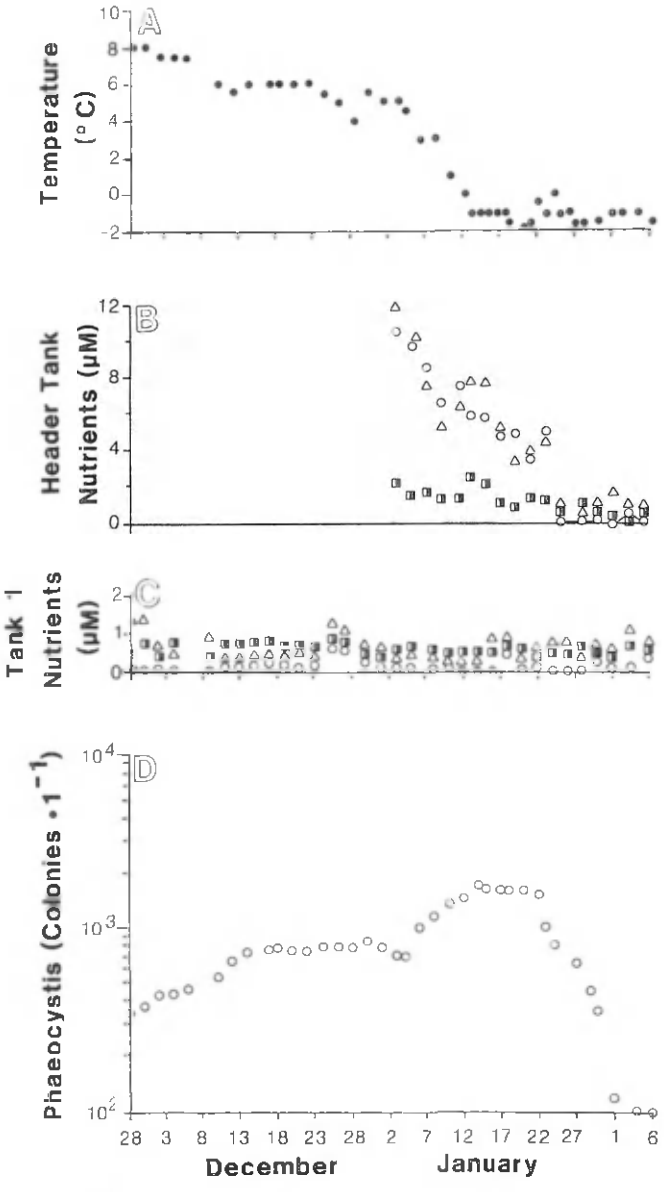


Fig. 1. Time course of temperature (**A**), nutrient concentrations in the header tank (**B**) which fed into tank 1 (**C**), and *Phaeocystis* abundance (**D**). Nutrients: $NO_3 + NO_2 + NH_4$ (○), $Si(OH)_4$ (△) and PO_4 (■).

AutoAnalyzer II (Glibert and Loder, 1977), were measured daily. *Phaeocystis* colonies in triplicate 100-ml samples were enumerated on alternate days using a Wild M5 stereomicroscope.

The role of temperature and nutrients in influencing the fate of blooms of *Phaeocystis* colonies was investigated through measurements of photosynthesis and growth rates. The methods are described in detail in Verity *et al.* (1988). Briefly, rates of photosynthesis were determined as follows. Sets of 250-ml polycarbonate bottles, soaked in acid and rinsed in deionized water, were gently filled with subsamples of the natural plankton community in tank 1. In the nutrient enrichment experiments (Tables I and II), duplicate bottles received various additions of reagent-grade chemicals, followed by inocula of 5 μCi of $NaH^{14}CO_3$ (New England Nuclear), prepared using trace metal clean techniques (Fitzwater *et al.*, 1982). These bottles were incubated for 7–24 h in growth chambers under a photoperiod of 12:12 L:D and 75–90 $\mu E\ m^{-2}\ s^{-1}$ (see Tables I and II for details). The incubation temperature of 6°C was similar to ambient tank temperature. In the photosynthesis–irradiance experiments (Figure 2), sets of duplicate bottles were inoculated with ^{14}C (no added nutrients) and were incubated outdoors for 24 h in flowing seawater exposed to 100, 50, 25, 10 and 5% of natural irradiance. Light intensity was attenuated using appropriate layers of neutral density screen. Additional bottles wrapped in aluminum foil served as dark controls.

After incubation, 5–20 colonies were pipette-isolated from each bottle and collected in triplicate on Gelman AE glassfiber filters under a vacuum pressure of 5 mmHg (= *Phaeocystis* photosynthesis). The remaining water in each bottle was split into triplicate 50-ml aliquots and collected on separate filters (= community production, including *Phaeocystis*). All filters were rinsed, placed in glass vials, acidified with 0.1 ml of 5 N HCl to drive off residual inorganic ^{14}C and suspended in 5 ml of Aquasol II scintillation cocktail. Samples were counted to an accuracy of at least 5% using a Beckman LS 150 liquid scintillation counter. Quenching was corrected by the channels ratio method utilizing a curve prepared from [^{14}C]toluene. An isotope discrimination factor of 1.05 was applied. Photosynthesis rates were corrected by subtracting dark bottle activity, which was always a small fraction of uptake in the light. Verity *et al.* (1988) discuss the effects of filter type and head pressure on radiolabel accumulation by *Phaeocystis* colonies.

Growth rates of *Phaeocystis* in various temperature and nutrient treatments were measured as the product of changes in colony abundance and cells per colony. Natural plankton communities dominated by *Phaeocystis* were collected from tanks 1 and 2, and incubated for 8 and 6 days respectively in 2½-l polycarbonate jugs. The jugs were wrapped in neutral density screening to simulate 50% of incident irradiance, and incubated outdoors in flowing seawater, at various temperatures. At 2-day intervals the *Phaeocystis* colonies contained in triplicate 100-ml samples were enumerated using a stereomicroscope. The number of cells per colony was determined using the method of Reynolds (1983), as modified by Verity *et al.* (1988). The abundance of *Phaeocystis* cells was calculated as (colony abundance) × (cells per colony).

Growth rates were expressed as population doublings according to:

$$K(\text{doublings day}^{-1}) = (1/t)\ \log_2(N_t/N_0)$$

where N_t and N_0 were abundances at times t and 0. These growth rates represent the net accumulation of cells within colonies, and incorporate any immigration and emigration of motile cells.

The chlorophyll *a* (chl *a*) content of *Phaeocystis* colonies was determined as described by Verity *et al.* (1988). Individual colonies were pipette-isolated into 0.45-μm filtered seawater, and transferred through several washes to remove other phytoplankton and particulate organic matter. Colonies were collected on 0.45-μm Gelman AE filters using low vacuum pressures (<5 mmHg). Chl *a* was extracted by grinding the filter in 90% acetone, and measured fluorometrically before and after acidification according to Holm-Hansen *et al.* (1965).

The abundance of bacteria suspended in the water column of the tanks, and those attached to the surface of *Phaeocystis* colonies, was enumerated in samples stained with acridine orange (Hobbie *et al.*, 1977; Davis and Sieburth, 1982). Bacterial concentrations in seawater were measured by *gently* pre-screening samples through a 20-μm Nitex mesh, staining with acridine orange and collecting the filtrate (devoid of *Phaeocystis* colonies) on 0.22-μm black Nuclepore filters. The number of bacteria attached to the colonies was determined by pipette-isolating stained *Phaeocystis* directly onto black Nuclepore filters, and enumerating the stained bacteria found on the colony surfaces. These surface counts were corrected for the abundance of bacteria found on portions of the filter not covered by *Phaeocystis* colonies. A minimum of 2×10^3 bacteria were enumerated in each sample.

Photomicrographs of *Phaeocystis* colonies were taken using a Zeiss Photomicroscope II equipped with phase-contrast optics.

Results

Tank 1 was filled with unfiltered water from Narragansett Bay in October 1984 but was not sampled until late November, by which time a bloom of *Phaeocystis* colonies had developed (Figure 1). During the next 8 weeks, *Phaeocystis* exhibited two consecutive periods of increasing colony abundance followed by stasis. Net colony growth rates were 0.07 and 0.13 doublings day^{-1} during the two periods of increasing abundance, or 0.13 and 0.19 doublings day^{-1} when corrected for the daily washout. A precipitous decline in colony number occurred in late January, with a decay constant of 0.25 day^{-1} in addition to the washout rate.

Temperature decreased from 8°C to 0°C during December and early January coincident with net increases in the number of *Phaeocystis* colonies (Figure 1). Cooling was gradual (−0.1°C day^{-1}) until early January, when extremely cold and windy conditions resulted in a rapid (−0.6°C day^{-1}) decay in mean water column temperature to <0°C. Water temperature in the unheated tank fluctuated between 0°C and −2°C during the ensuing 3 weeks, during which time there was frequent formation of surface ice.

Nutrient concentrations in the water entering the tank from Narragansett Bay were measured beginning in January (Figure 1). At that time, inorganic nitrogen ($NO_3 + NO_2 + NH_4$) and silicate were 10–12 μM, and PO_4 was 2 μM. Concentrations declined steadily during the next three weeks and approached limits of detection, presumably due to uptake by the winter–spring diatom bloom which developed in the Bay during January. Despite the variability in supply rate, ambient concentrations of all nutrients in tank 1 were relatively constant and low throughout the study (Figure 1). Combined inorganic nitrogen was consistently near or below detection limits (0.1 μM), while PO_4 and $Si(OH)_4$ were generally <1 μM.

The nutrient data implied that nitrogen might have limited the growth of *Phaeocystis*, at least prior to the rapid decline in temperature in January. Two photosynthesis experiments were conducted in December to evaluate this hypothesis. Both studies, of similar design, involved incubating natural plankton communities from tank 1 with ^{14}C and various nutrient treatments. The first experiment measured photosynthetic rates of *Phaeocystis* colonies and the entire community for 7 and 24 h (Table I). The second, separate experiment determined responses after 24 and 48 h (Table II). The nutrient additions, which are given in the tables, were of sufficient magnitude to ensure that concentrations remained well above detection limits during the incubations. This was confirmed using automated analyses of samples collected after the incubations (data not shown). *Phaeocystis* incorporated labelled carbon at rates of 0.3–0.4 μg C colony^{-1} during 7-h incubations, contributing 40–48% of the total carbon uptake of the entire community. There were no significant differences in incorporation of ^{14}C by *Phaeocystis* colonies among individual nutrient treatments after 7 h. However, after 24 h the highest photosynthetic rates characterized colonies in treatments containing either NO_3 or NH_4 alone or with added PO_4 (Table I). Lower colony-specific rates were found in treatments with $Si(OH)_4$ alone or combined with PO_4. The presence of N or N + P stimulated photosynthesis by *Phaeocystis* (56–62% of community uptake) compared with control treatments (42%). These effects were investigated over longer periods in a second study where additional nutrient treatments were imposed (Table II). In agreement with the first experiment, the highest colony-specific photosynthetic rates over 24 and 48 h were in treatments containing nitrogen; rates were usually higher over 0–24 h compared with the same treatments over 24–48 h due to the production of new, smaller colonies over time. The contribution by *Phaeocystis* to community production was also highest in nitrogen treatments, an effect especially evident after 48 h. These conclusions were supported by calculations of Spearman rank correlation coefficients, which demonstrated significant similarity ($P < 0.001$) in nutrient stimulation of *Phaeocystis* production among the three 24 h incubations (Tables I and II), with the highest ^{14}C incorporation into colonies occurring in treatments containing added nitrogen. The addition of trace metals and EDTA to N + P + Si resulted in the highest colony photosynthetic rates. The contribution by *Phaeocystis* to community production was not similarly stimulated, suggesting that potential limitation of photosynthesis by micronutrients may have affected all phytoplankton.

Table I. Photosynthetic rates of *Phaeocystis* colonies from plankton communities in tank 1 incubated with ^{14}C and various nutrient treatments for 7 and 24 h

Treatment	0–7 h		0–24 h	
	PS (μg C colony^{-1} 7 h^{-1})	PS (Phaeo/Σ) (%)	PS (μg C colony^{-1} 24 h^{-1})	PS (Phaeo/Σ) (%)
NO_3	0.40 ± 0.07	41	0.42 ± 0.03	59
NH_4	0.38 ± 0.06	48	0.42 ± 0.10	56
PO_4	0.34 ± 0.09	40	0.32 ± 0.08	44
$Si(OH)_4$	0.35 ± 0.02	40	0.27 ± 0.05	33
NH_4 + P	0.35 ± 0.02	40	0.40 ± 0.06	62
NO_3 + P	0.37 ± 0.02	42	0.42 ± 0.12	59
Si + P	0.40 ± 0.05	44	0.25 ± 0.02	44
NH_4 + Si	0.43 ± 0.04	48	0.34 ± 0.03	48
NO_3 + Si	0.34 ± 0.10	44	0.26 ± 0.02	44
Control	0.34 ± 0.06	44	0.33 ± 0.02	42

Rates are expressed as carbon uptake per colony and as a percentage of total carbon uptake by the entire community. Initial and control nutrient concentrations (μM) were: $NO_3 = 0.3$, $NH_4 = 0.1$, $PO_4 = 0.3$ and $Si(OH)_4 = 0.5$. Nutrient additions were as follows: $NO_3 = 20$, $NH_4 = 20$, $PO_4 = 3$ and $Si(OH)_4 = 36$ μM. Samples were incubated at 6°C in growth chambers under 12L:12D and ~90 μE m^{-2} s^{-1}. The 7-h incubation was done during the light phase. Error bars represent 1 SD.

Table II. Photosynthetic rates of *Phaeocystis* colonies from plankton communities in tank 1 incubated with various nutrient treatments for 24 and 48 h

Treatment	0–24 h		24–48 h	
	PS (μg C colony^{-1} 24 h^{-1})	PS (Phaeo/Σ) (%)	PS (μg C colony^{-1} 24 h^{-1})	PS (Phaeo/Σ) (%)
NO_3	0.38 ± 0.04	46	0.29 ± 0.01	69
NH_4	0.38 ± 0.28	54	0.23 ± 0.01	55
PO_4	0.29 ± 0.08	34	0.13 ± 0.04	33
$Si(OH)_4$	0.14 ± 0.02	20	0.15 ± 0.04	27
NH_4 + PO_4	0.49 ± 0.10	42	0.25 ± 0.03	58
NO_3 + PO_4	0.30 ± 0.08	43	0.27 ± 0.01	76
Si + PO_4	0.19 ± 0.01	31	0.12 ± 0.02	26
NH_4 + Si	0.23 ± 0.11	40	0.24 ± 0.03	–[a]
NO_3 + Si	0.31 ± 0.08	40	0.21 ± 0.06	35
NH_4 + PO_4 + Si	0.32 ± 0.06	42	0.27 ± 0.02	–[a]
NO_3 + PO_4 + Si	0.31 ± 0.08	36	0.31 ± 0.04	45
NPS + TM + EDTA	0.54 ± 0.20	46	0.51 ± 0.15	50
Control	0.30 ± 0.08	33	0.18 ± 0.02	46

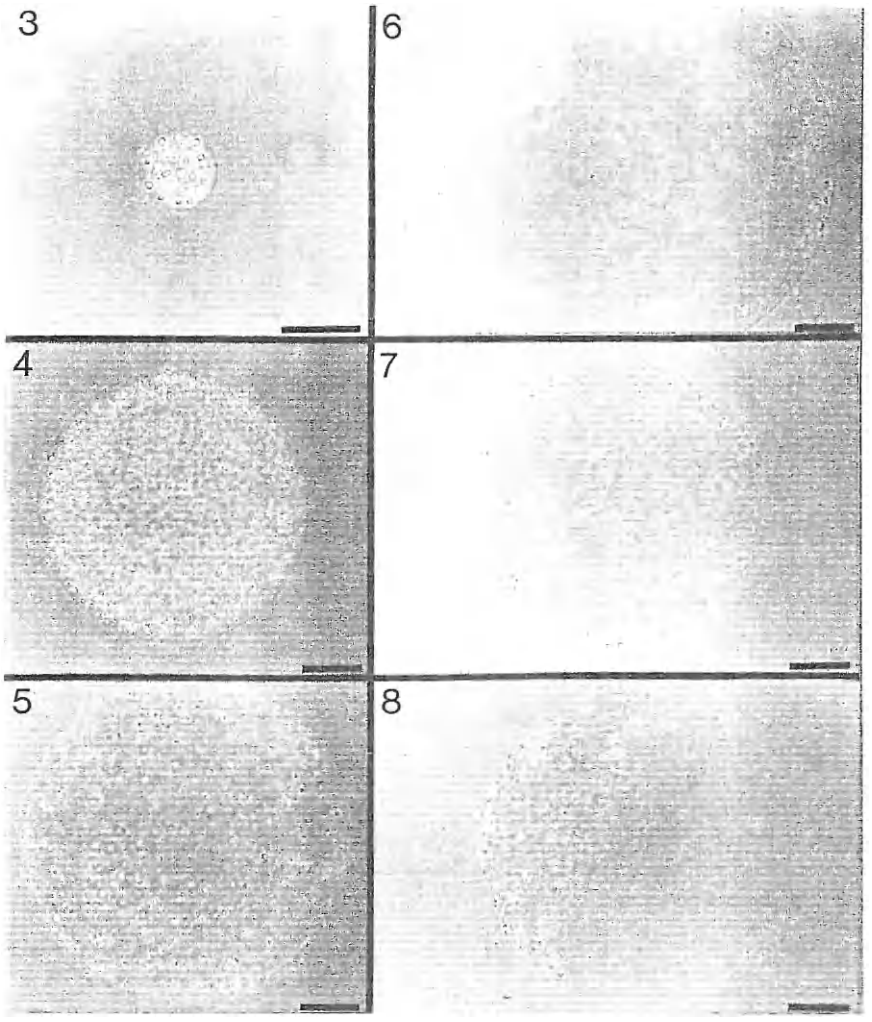
^{14}C was added for 24 h beginning at t = 0 and t = 24 h respectively. Rates are expressed as carbon uptake per colony and as a percentage of total carbon uptake by the entire community. Initial and control nutrient concentrations (μM) were: NO_3 = undetectable, NH_4 = 0.1, PO_4 = 0.7 and $Si(OH)_4$ = 0.3. Nutrient additions were made at t = 0 and t = 24 h as follows: NO_3 = 20, NH_4 = 20, PO_4 = 3 and $Si(OH)_4$ = 36 μM. Trace metals (TM) and EDTA were added according to Schone and Schone (1982). NPS = NO_3 + PO_4 + $Si(OH)_4$. Samples were incubated in growth chambers at 6°C, 12L:12D and ~75 μE m^{-2} s^{-1}. Error bars represent 1 SD.

[a]Lost.

The decline in water temperature during January was associated with significant declines in *Phaeocystis* photosynthesis (Figure 2). This trend was apparent when expressed on a colony- or pigment-specific basis. Carbon incorporation decreased from 0.18 μg C colony^{-1} day^{-1} at 3°C to 0.02 μg C colony^{-1} day^{-1} at −1.5°C, equivalent to 94 μg C μg Chl a^{-1} day^{-1} (3°C) and 22 μg C μg Chl a^{-1} day^{-1} (−1.5°C). The colonies contained similar amounts of particulate organic carbon, 317–364 ng C. However, they differed significantly in their Chl *a* content: 1.9 ng colony^{-1} on January 6 (3.0°C) compared with 0.6 ng colony^{-1} on January 18 (−1.5°C).

The decrease in pigment content was due to emigration of cells out of the colonies (Figures 3–8). Normal, healthy colonies were spherical, ~50 μm to several millimeters in diameter, and contained hundreds to thousands of cells (Figures 3 and 4). The cells were evenly distributed around the periphery of the colony, and were only occasionally deeply embedded within the gelatinous matrix. The number of cells per unit colony surface area began to decline in mid January (Figure 5), eventually resulting in gelatinous spheres nearly devoid of cells (Figure 6). Colonies with decreased cell densities were also occasionally observed with apparent extracolonial protrusions (Figure 7), the function of which is unknown. More commonly, cells were aggregated, with one side of the

Fig. 2. Photosynthetic rates (PS) of *Phaeocystis* colonies in tank 1 on January 6 (○) and January 18 (●). Rates are expressed on a pigment-specific (**A**) and colony-specific (**B**) basis. Error bars represent 1 SD. Irradiance (*I*) was daily incident solar radiation.



Figs 3–8. *Phaeocystis* colonies from tank 1. **3.** A young colony. **4.** A mature 'healthy' colony. Focal plane is through the center of the colony. Note that cells at the periphery are in focus, while the center remains empty, indicating that most cells are found in proximity to the colony–seawater interface. **5.** A typical colony observed shortly after temperature plummetted below 0°C. Note the reduced density of cells compared with the previous figure. **6.** A larger colony, almost devoid of cells, collected a few days later during the period of declining abundance. **7.** A colony similar to that in Figure 5, but with a gelatinous protrusion of unknown function. **8.** A second type of colony often found during the period of subzero temperatures. Note the increase in cell density from one polar region devoid of cells to the opposite pole, where cells were closely packed and many were motile. All scale bars = 50 μm.

colony completely devoid of cells. Cell density gradually increased along the colony surface to a maximum (Figure 8), at which point the cells were packed together, almost in contact, and many were motile.

Emigration of cells out of the colonies coincided with rapid declines in both temperature and nutrients. Two growth experiments were conducted to investigate the relative roles of these factors in this life-cycle event. The first, initiated on January 22, was designed to determine if an increase in temperature or an addition of nutrients would reverse the trend of declining abundance and cell density in the colonies (Figure 9). Two 2½-l jugs were filled with natural plankton communities from tank 1 (−1.5°C), wrapped in neutral density screen to simulate 50% I_0, and incubated outdoors in flowing seawater (+1.0°C) for 8 days. One jug received nitrate, phosphate, silicate, trace metals and EDTA similar to those in Table II, while the other was undisturbed. Colony abundance and cell density declined linearly over 8 days, despite the warmer temperature. Nutrient-replete colonies consistently contained more cells and were more numerous than nutrient-depleted colonies, but variability obscured possible statistical differences. *Phaeocystis* colonies in both treatments were essentially 'ghosts' when the experiments were terminated.

A second similar experiment was initiated on February 6 to investigate the effects of temperature on inception of cell motility and emigration (Figure 10). Three 2½-l jugs were filled with natural plankton communities from tank 2, and were incubated under 50% I_0 at various temperatures for 6 days. The *Phaeocystis* in tank 2 at this time were nutrient-replete and growing actively at

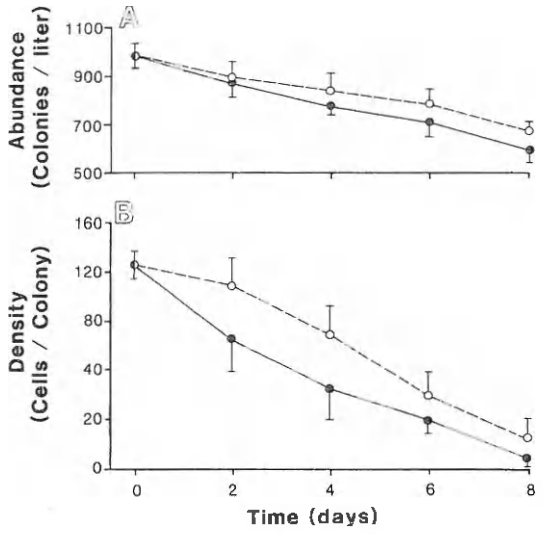


Fig. 9. Time course of *Phaeocystis* abundance (**A**) and cell density within the colonies (**B**) in tank 1 plankton communities collected at −1.5°C and incubated at +1.0°C. A control jug lacking nutrients (●) and one with added N + P + Si + TM + EDTA (○) were incubated at 50% I_0 for 8 days. Error bars represent 1 SD. See text for details.

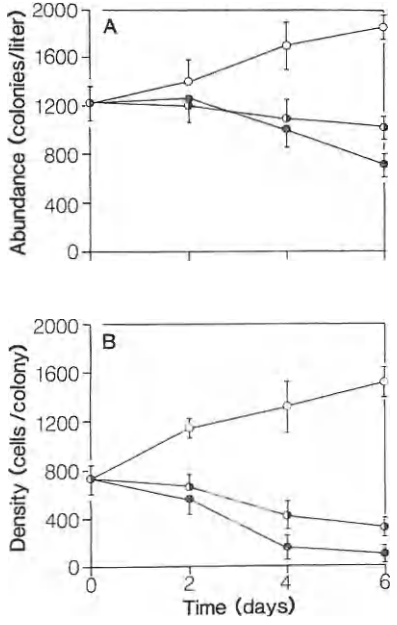


Fig. 10. Time course of *Phaeocystis* abundance (**A**) and cell density within the colonies (**B**) in tank 2 plankton communities incubated at 3°C (○), −0.5°C (◑) and −1.5°C (●). Jugs were incubated at 50% I_0 for 6 days. Error bars represent 1 SD. See text for details.

3°C (Verity *et al.*, 1988). Colonies in the jugs kept at 3°C approximately doubled in cell density and showed small but steady increases in abundance (Figure 10). In contrast, colonies incubated at −0.5°C and −1.5°C decreased in abundance and lost cells throughout the experiment. This effect was especially apparent at the lower temperature.

The concentrations of bacteria in seawater and on the surfaces of colonies were determined in two tanks which differed substantially in the physiological state of their *Phaeocystis* populations (Table III). Colonies in tank 1 were temperature-stressed, nutrient-starved, contained few cells and had low pigment-specific photosynthetic rates. Populations in tank 8 were growing actively at 3°C in the presence of excess nutrients, contained numerous cells and exhibited high photosynthetic rates (Verity *et al.*, 1988). Despite relatively similar concentrations of suspended bacteria in the two tanks, colonies in tank 1 contained 880 bacteria cells per 10^4 μm^2 of surface area, compared with five cells in an equivalent surface area in colonies from tank 8.

Discussion

Enormous stands of colonial *Phaeocystis* exceeding 10^8 colonies m^{-3} occur in both coastal and oceanic waters (Kashkin, 1963). In polar regions, epidemics of

Table III. Abundance of bacteria in seawater (SW) and on the surface of colonies (C) in two tanks on 29 January

Tank	DIN (μM)	Chl *a* (ng colony^{-1})	PS (gC g Chl a^{-1} day^{-1})	Bacteria	
				SW (cells ml^{-1})	C (cells 10^{-4} m^2)
1	0.3	0.1	4.1	$5.5 \cdot 10^6$	880
8	10.7	3.1	55.8	$1.6 \cdot 10^6$	4.9

DIN = $NO_3 + NO_2 + NH_4$. Chl *a* = chlorophyll *a* content of colonies. PS = pigment-specific colony photosynthesis.

Phaeocystis are a classical bloom phenomena (Smayda, 1958; El-Sayed *et al.*, 1983; Palmisano *et al.*, 1986). This species accounts for as much as 80% of the phytoplankton biomass during blooms in Norwegian waters (Eilertsen *et al.*, 1981; Haug *et al.*, 1973; Nost-Hegseth, 1982), and it exhibits a remarkable year-round occurrence in Balsfjord (70°N) over an annual temperature range of 1–7°C. *Phaeocystis* is equally abundant along the Dutch, German and Belgian coasts, where there is the provocative historical implication that it has become a 'weed' species in the progressively eutrophic Wadden Sea (Cadee and Hegemann, 1974, 1979, 1986). Blooms exceed 10^7 colonies m^{-3} in Narragansett Bay (Verity *et al.*, 1988), the eastern Irish Sea and Liverpool Bay (Jones and Haq, 1963; Jones and Spencer, 1970), and the North Sea (Weisse *et al.*, 1986).

The fate of this production is unknown. Most phytoplankton blooms are assumed to be ingested by herbivores, or to settle out to be remineralized at depth, to provide food for the benthos, or to be buried in the sediments. The evidence that grazers terminate colonial *Phaeocystis* blooms is confused and contradictory. For example, from early studies of gut contents it was concluded that *Phaeocystis* serves as food for copepods, cladocerans and meroplanktonic larvae (Lebour, 1922; Nicholls, 1935; Jones and Haq, 1963; Fretter and Montgomery, 1968). In contrast, oyster larvae suffer significant reductions in growth rate during *Phaeocystis* blooms (Walne, 1970), and the species is unsuitable as food for adult oysters (Gabbott and Walker, 1971) and mussels (Pieters *et al.*, 1980). Some euphausiids ingest *Phaeocystis* (Marr, 1962), whereas others apparently do not (Falk-Petersen *et al.*, 1982). Temperate copepods of intermediate size do not ingest colonies >200–300 μm in diameter (Weisse, 1983; Foffonoff *et al.*, 1986; P.G.Verity *et al.*, submitted), whereas some large suspension-feeding Arctic herbivores (*Calanus hyperboreus*) ingest *Phaeocystis*, at least in unialgal experiments (Huntley *et al.*, 1987). Schnack (1983) reported that suspension-feeding Antarctic copepods did not prey on colonies, in contrast to active feeding by copepods using a 'mixed and raptorial feeding mode'. Thus, while nutritionally acceptable to some herbivores, *Phaeocystis* is apparently unpalatable or inedible to others.

Few data are available concerning sedimentation of *Phaeocystis* blooms. Remnants of colonies have been identified in shallow-depth sediment traps in the Barents Sea (Wassmann, 1987). In the North Sea, extensive accumulations of sea foam coincide with the demise of blooms of *Phaeocystis* colonies (Eberlein

et al., 1985; Batje and Michaelis, 1986). These events have been attributed to the disintegration and decay of colonies. Our data indicate that bacteria concentrations are elevated on ghost colonies, suggesting that microbial activity in the water column may enhance their remineralization. The gelatinous matrix of *Phaeocystis* colonies is composed primarily of medium to high molecular weight polysaccharides (Guillard and Hellebust, 1971; Veldhuis and Admiraal, 1985), theoretically a good substrate for bacterial metabolism. However, *Phaeocystis* produces copious quantities of acrylic acid (Guillard and Hellebust, 1971), a substance with known antibiotic properties (Sieburth, 1960). *Phaeocystis* also secretes large amounts of dimethylsulfide (DMS), a by-product of the reaction which forms acrylic acid, prompting the hypothesis that this species and others synthesize acrylic acid primarily to inhibit bacterial attack, with DMS providing a metabolic tracer (Barnard *et al.*, 1984). Although this postulate has not been investigated, the present observations and similar ones in culture (Guillard and Hellebust, 1971) indicate that *Phaeocystis* cells have some mechanism which prevents attachment of significant numbers of bacterial cells to the colony surface. This mechanism is much less effective when the cells leave the colonies. These data are supported by low rates of uptake of [^{3}H]thymidine in *Phaeocystis* cultures (Veldhuis and Admiraal, 1985), and elevated bacterial productivity and biomass following the 'collapse' of *Phaeocystis* colony blooms in the Wadden Sea (Laanbroek *et al.*, 1985; Billen and Fontigny, 1987). The resulting sea foam also contains high concentrations of saprophytic bacteria (Gunkel, 1982, cited in Eberlein *et al.*, 1985).

The collapse of blooms of *Phaeocystis* colonies does not require the death of cells contained within them. Under favorable conditions, non-motile cells may develop flagella and exit the colonies. This release of single-celled 'swarmers' has been documented in culture (Kayser, 1970; P.G.Verity, unpublished) and in other field studies (Jones and Haq, 1963; Parke *et al.*, 1971). The emigration of cells from colonies in tank 1 occurred relatively quickly even at −1 to −2°C, with most colonies devoid of cells in <10 days. This phenomenon may be more rapid at warmer temperatures. For example, the emergence of motile cells from colonies of the related prymnesiophyte *Pleurochrysis* required only 12–24 h at 15°C (Merrick and Leadbeater, 1979). The new flagella of *Pleurochrysis* were short and difficult to detect, though their development was rapid.

Several factors may regulate the inception and rate of swarmer release from colonies of *Phaeocystis*. In tank 1, emigration coincided with simultaneous declines in nutrient concentrations and temperature. Actively growing, nutrient-replete colonies at 3°C rapidly released motile cells when transferred to subzero temperatures (Figure 10), indicating that low temperatures may induce swarmer release independent of nutrient availability. In tanks 2 and 6, which were thermally regulated to 2–4°C (Verity *et al.*, 1988), chronic nutrient deprivation gradually led to ghost colonies, suggesting that nutrient stress alone may also induce this life-cycle event. In agreement with these field observations, *Phaeocystis* colonies in culture produced large numbers of swarmers when nutrients were depleted (Kayser, 1970). Provocatively, high nutrient concentrations in culture may also result in proliferation of single cells (Guillard and

Hellebust, 1971). Increasing temperature and resupply of nutrients to colonies in tank 1 did not stop the emigration of cells, suggesting that, once initiated, swarmer release must proceed to completion. These field data are not conclusive, however, and the role of abiotic factors in mediating life-cycle events in this alga requires documentation under controlled conditions.

Thus, the fate of *Phaeocystis* blooms remains an enigma. The physiological option of producing motile unicells may provide a refuge from stressful or lethal conditions for cells contained within or constrained by a gelatinous matrix. If so, this life-cycle event might be the functional equivalent of resting spore and cyst formation in diatoms and dinoflagellates. However, the swarmers are not physiologically idle, but instead are photosynthetically active and capable of rapid growth in culture (P.G.Verity and T.J.Smayda, in preparation). The production of motile unicells may also be only the first step in a more complex process, since the occurrence and role of sexual reproduction in *Phaeocystis* is unknown. Likewise, regulatory mechanisms remain speculative. Development and release of swarmers may be the direct result of temperature or nutrient effects on the metabolism of colony cells, or an indirect effect of chemical inducers released extracellularly by stressed colony cells. In the colonial green alga *Volvox*, heat shocks induced somatic cells to manufacture and release a sexual inducer, which caused the asexual females to generate egg-bearing daughters (Kirk and Kirk, 1986). Such an autoinduction of sexuality would be adaptive in nature, where *Volvox* reproduces asexually in the spring but requires sexual reproduction to produce dormant zygotes prior to evaporation of temporary ponds in the summer. Unfortunately, the life-cycle of *Phaeocystis* is not sufficiently understood to speculate on the adaptive significance of swarmer release. It is not even clear whether swarmers are sexual or asexual, whether they remain planktonic or whether they can produce new colonies.

Despite these uncertainties, the transition from colonies to solitary cells has significant implications for food web structure and function. This life-cycle event incorporates a decrease in diameter of 10^2–10^3 and a decrease in volume of 10^6–10^9. The opposite phenomenon, colony development from solitary cells, has been hypothesized as an adaptation to minimize losses to suspension-feeding zooplankton (Reynolds *et al.*, 1982). Thus, the demise of large colonies and concurrent release of perhaps millions of 3- to 6-μm cells per liter may significantly alter the community composition and feeding behavior of herbivores. Solitary *Phaeocystis* cells are a good food source for phytophagous microzooplankton, but they are too small to be efficiently collected by many herbivorous copepods (P.G.Verity *et al.*, submitted). Moreover, bacterial production is enhanced following 'collapse' of a colony bloom. These combined data suggest that swarmer release may induce substantial alterations in the size structure and perhaps the ecological efficiency of planktonic food webs. However, the small size and non-distinctive morphology of solitary cells preclude their identification and enumeration by standard light microscopy in samples containing other photosynthetic nanoplankton. The challenge for future studies is to overcome this obstacle and quantify the role of life-cycle phenomena in the *in situ* dynamics of *Phaeocystis*.

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Estimating nutrients recycling by tropical oceanic macroplankton–micronekton from biomass data

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Abstract. Simultaneous measurements of biomass and excretion of macroplankton–micronekton lead to similar results in tropical Atlantic and Pacific Oceans. Ratios are thus proposed to roughly estimate regenerated production in oligotrophic tropical waters from biomass data: biomass figures (mg dry wt m^{-2}) should be multiplied by 0.32 to estimate the amount of total nitrogen excreted (μg at m^{-2} day^{-1}) and by 0.027 to estimate the amount of total phosphorus excreted (μg at m^{-2} day^{-1}).

Introduction

From seven cruises taken in 1978–1979 in the tropical Atlantic Ocean 4°N–10°S/4°W (Roger, 1982a,b) and in 1982–1984 in the tropical Pacific Ocean 21°S/168°E (Roger, 1986, 1988), data have been gathered simultaneously on (i) the biomass of macroplankton–micronekton defined as individuals in the range 0.5–10 cm long, and (ii) nitrogen and phosphorus release by animals of the different taxa based on individual measurement of the excretion of 391 animals, mainly crustaceans.

Materials and methods

Biomass data were obtained from 221 non-closing oblique tows performed with a 160-cm-diameter ORI net (Omori, 1965) fitted with 2-mm mesh, operated by night (20.00–24.00 hours local time) between the surface and a depth of ~400 m. Net speed was maintained close to 3 knots. Precise depth and volume filtered were given by a depth–distance recorder (DDR from TSK). The 0–400 m stratum is considered to contain at least 90% of the whole micronekton biomass in the size range 0.5–10 cm at night (Roger, 1986). Samples were preserved in 10% formalin, and after a few weeks or months, having been rinsed with tap water and dried at 65°C for 48 h were sorted before measuring dry weight.

Excretion experiments were conducted as follows. The ORI net was slowly towed for ~10 min between the surface and a depth of ~50 m, between 20.00 and 23.00 hours local time, i.e. at a time where the vertical distribution of animals is at its most shallow. The sample was carefully poured into a vessel previously half-filled with surface seawater. Apparently healthy animals were gently transferred to 1-l brown glass flasks containing seawater taken at a depth of 10 m, filtered through 0.8-μm mesh and cooled to the experimental temperature. These flasks had been washed previously with 10% chlorhydric acid, then rinsed with flowing seawater. All the experiments reported here were carried out at 17°C, which is a temperature intermediate between those of deep