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Diel patterns of photosynthate biosynthesis by phytoplankton in permanently ice-covered Antarctic lakes under continuous sunlight

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Abstract. Diel patterns of photosynthate biosynthesis by Antarctic freshwater phytoplankton growing under the variable but continuous sunlight of summer were found to be similar in many respects to those reported from other aquatic environments where light/dark periods alternate. Lipid synthesis by freshwater phytoplankton in Lakes Vanda and Fryxell predominated during periods when solar radiation and photosynthesis were most intense; the inverse was generally true of the protein and polysaccharide fractions. The major photosynthetic end-products in both lakes were protein and polysaccharide, which together accounted for 60–81% of the total cellular carbon incorporation. Less than 4% of the carbon was incorporated into lipid in Lake Vanda; >12% appeared in the lipid fraction in Lake Fryxell. The Lake Fryxell populations showed evidence of photoinhibition of complete photosynthesis during 'midday' when irradiance was most intense. I_k values, computed from the photosynthesis irradiance relationships in Lake Fryxell, corroborate other studies suggesting that the phytoplankton populations in permanently ice-capped Antarctic lakes are among the most shade-adapted yet reported.

Introduction

The influence of the light/dark cycle on patterns of photosynthate biosynthesis by microalgae has attracted the recent interest of phytoplankton ecologists (e.g. Morris, 1981; Cuhel *et al.*, 1984). Culture studies of phytoplankton have shown that dark catabolism of polysaccharides and lipids accumulated during the light period can sustain protein synthesis during the dark period (Foy and Smith, 1980). Studies of carbon partitioning patterns by natural phytoplankton populations incubated under *in situ* day/night conditions also indicate that protein synthesis occurs during the night at the expense of intracellular low-molecular-weight (LMW) metabolites, polysaccharides and lipids synthesized during the day (Morris, 1981; Priscu and Priscu, 1984).

Nearly all of the work on the intracellular distribution of photosynthate has been conducted on organisms that experience a periodic day/night regime in nature. The only study conducted on organisms under continuous irradiance of which we are aware showed little dependence of photosynthate distribution on diel irradiance in Arctic marine phytoplankton (Li and Harrison, 1982). This lack of dependence appears to be related to the fact that the light levels during the experiment were seldom below saturation. The dependence of complete photosynthesis (i.e. non-fractionated carbon incorporation) on continuous polar irradiance by freshwater phytoplankton has been reported by Goldman *et al.* (1969) and Whalen and Alexander (1984). Both of these latter studies showed that complete photosynthesis was in phase with the diel light regime.

We present here a description of photosynthate biosynthesis by Antarctic freshwater phytoplankton growing in habitats receiving a continuous supply of

solar energy over the 24 h cycle. Our study sites are unique in that they are permanently capped by a minimum of 3.5 m of ice and advective mixing from inflows and outflows is negligible. Consequently, the water columns of these lakes are extremely stable hydraulically and distinct vertical microbial layering occurs (Priscu *et al.*, 1987). Owing to this stability, the only major diel fluctuations in irradiance are from the variations in sunlight, i.e. the organisms are not exposed to short-term irradiance fluctuations associated with vertical water movement.

Materials and methods

Study sites

Studies were conducted on Lake Vanda (77°35′S,161°40′E) and Lake Fryxell (77°35′S, 163°15′E), two permanently ice-covered lakes located in the Wright and Taylor Dry Valleys, respectively, of southern Victoria Land, Antarctica. Lake Vanda is 5 km^2 in area, 70 m deep and permanently covered by 3.5 m of ice; Lake Fryxell is larger (7 km^2) and shallower (19 m) and is permanently covered throughout summer by 4.6 m of ice. Because of the permanent ice-cover, these lakes possess an extremely stable water column and conspicuous microbial layering (Vincent, 1981; Priscu, 1987). A distinct feature in these lakes, that results partially from this hydraulic stability, is a deep chlorophyll *a* and photosynthesis maximum that exists near the bottom of the trophogenic zone at irradiance levels of 0.1–1.0% of the solar radiation incident on the surface of the ice-cap. Both lakes are anaerobic and nutrient-rich below the deep-chlorophyll maxima. More thorough descriptions of these lakes can be found in the references cited above.

Methods

Diel partitioning patterns were determined on lake water samples collected just beneath the ice caps of both lakes and in the deep-chlorophyll layer of Lake Fryxell (9 m). A 1-l sample from each location was inoculated with 75–160 μCi of $Na_2{}^{14}CO_3$ and incubated in a lakeside hut near the light level and temperature of collection. It was necessary to incubate the samples 'indoors' to avoid freezing. Labeled organisms were harvested from the radiolabeled 1-l sample by filtration of 100 ml subsamples at specific times over a 24 h period onto Whatman GF/F glass-fiber filters. The filters were (wet) frozen and transported to New Zealand for biochemical fractionation using the method described by Priscu and Priscu (1984). This procedure apportions the incorporated ^{14}C into methanol–water-soluble (LMW metabolites), cholorform-soluble (lipid), hot trichloroacetic acid (TCA)-soluble (polysaccharide) and TCA-insoluble (protein) fractions. Solar irradiance was measured with a recording Li-Cor pyranometer located at Lake Vanda. Lake Fryxell lies about 45 km from Lake Vanda; therefore, irradiance measured at the latter location may not reflect completely small local variations at Lake Fryxell.

Results

The diel pattern of complete photosynthesis (i.e. total cellular carbon incorporation) for the 3.5 m populations in Lake Vanda generally tracked solar irradiance (Figure 1, Table I). The difference between 'day' (1300 h) and 'night' (2400 h) photosynthetic rates was ~5-fold in response to about a 10-fold difference in irradiance. On average (±SD), most of the ^{14}C flowed into the

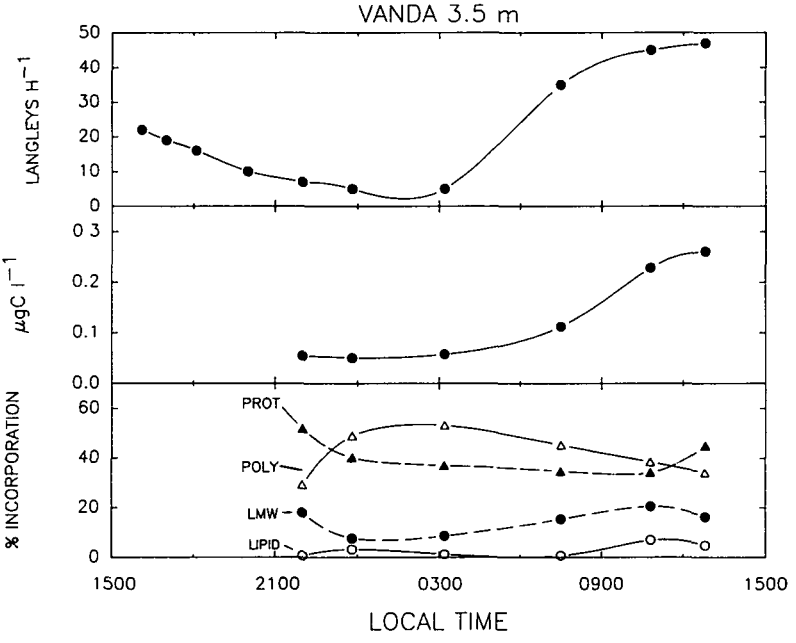

Fig. 1. Diel pattern of irradiance (ly h^{-1}), photosynthesis (μg C l^{-1}) and incorporation of C into the major end-products of photosynthesis (% of total) for the 3.5 m microbial population in Lake Vanda. ^{14}C was added to the sample at 1391 h local time.

Table I. Statistical comparisons among irradiance (ly h^{-1}), photosynthesis (μg C l^{-1}) and carbon incorporation into the major end-products of photosynthesis (%)

Variables		Vanda 3.5 m		Fryxell 4.6 m		Fryxell 9 m	
x	*y*	*r*	*P*	*r*	*P*	*r*	*P*
ly h^{-1}	μg C l^{-1}	0.94	0.01	0.50	0.14	0.40	0.26
	% lipid	0.87	0.03	0.10	0.10	0.49	0.15
	%LMW	0.68	0.14	0.06	0.06	−0.14	0.71
	% poly.	−0.37	0.47	0.36	0.36	−0.20	0.60
	% prot.	−0.30	0.56	−0.28	−0.28	0.05	0.88
μg C l^{-1}	% lipid	0.79	0.06	0.75	0.75	0.58	0.08
	% LMW	0.62	0.19	−0.60	−0.60	0.55	0.11
	% poly.	−0.44	0.40	−0.20	−0.20	−0.40	0.26
	% prot.	−0.17	0.77	0.24	0.24	−0.41	0.24

r = correlation coefficients; *P* = probability that slope will not differ from zero.

polysaccharide (41.5 ± 9.2%) and protein (40.4 ± 6.9%) fractions followed by incorporation into LMW metabolites (14.4 ± 5.1%) and lipid (3.7 ± 2.4%) over the diel period. Diel trends in labeling patterns indicate that percent lipid and LMW labeling were positively correlated with irradiance and photosynthesis (Table I). Conversely, maximum polysaccharide and protein synthesis in Lake Vanda occurred when irradiance was at a minimum, as indicated by the negative correlations in Table I.

Although not as tightly associated as in Lake Vanda, photosynthesis at 4.6 and 9 m in Lake Fryxell was positively correlated with irradiance (Figures 2 and 3, Table I). The lower intensity of association in Lake Fryxell is related to the fact that complete photosynthesis is not linearly related to irradiance, as is the case in Lake Vanda (Figure 4). Photosynthesis in the 4.6 m population appeared to be inhibited by irradiance levels greater than ~20 ly h^{-1}; photosynthesis in the 9 m population was inhibited above ~25 ly h^{-1}. The average (±SD) percentage ^{14}C incorporation over the period of measurement into the lipid, LMW, polysaccharide and protein fractions at 4.6 and 9 m was 20.9 ± 5.2, 18.4 ± 8.9, 23.5 ± 5.0 and 37.2 ± 10.3, and 12.8 ± 2.7, 13.4 ± 5.2, 30.8 ± 4.9 and 43.1 ± 6.0, respectively. Within Lake Fryxell, polysaccharide and protein labeling at 9 m was greater than in the 4.6 m populations, primarily at the expense of ^{14}C incorporation into lipid. Both of these biosynthetic patterns differed from those in Lake Vanda, where lipid received <4% and polysaccharide >40% of the label.

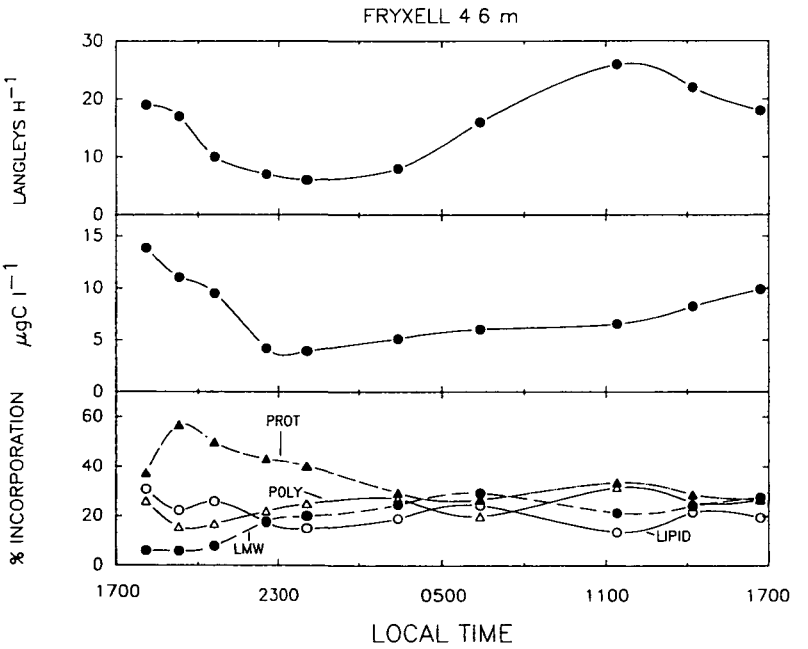

Fig. 2. As in Figure 1 except for the 4.6 m population in Lake Fryxell. ^{14}C was added to the sample at 1606 h local time.

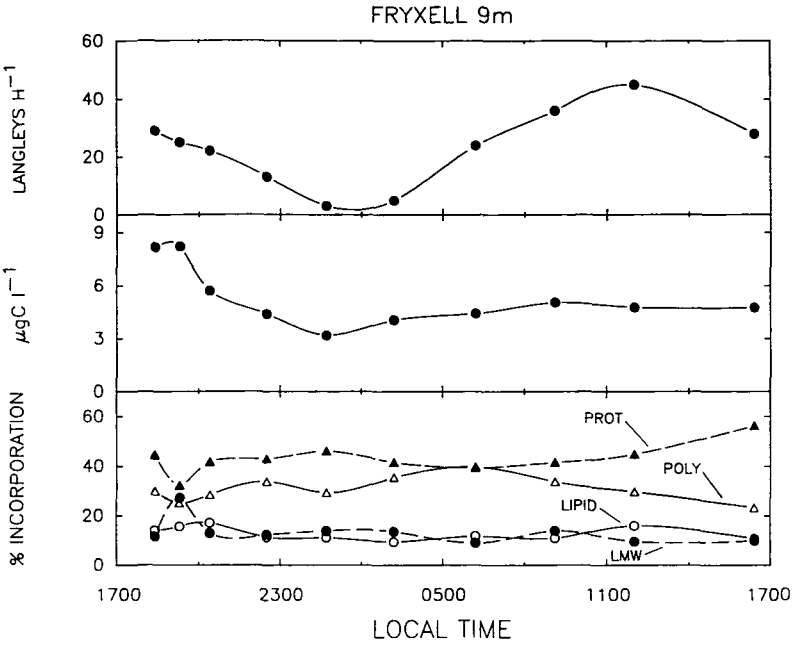

Fig. 3. As in Figure 1 except for the 9 m population in Lake Fryxell. ^{14}C was added to the sample at 1615 h local time.

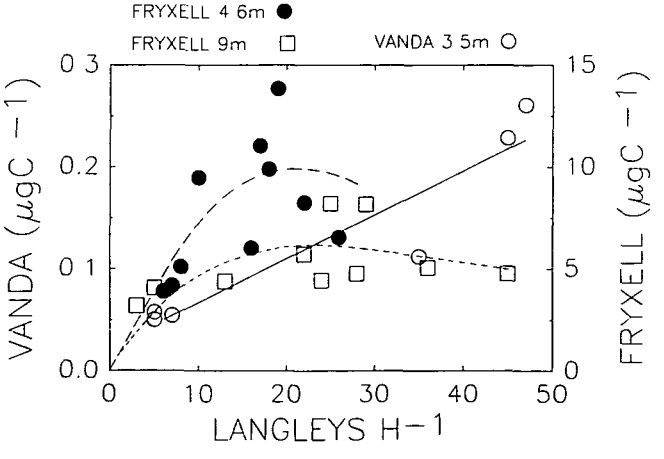

Fig. 4. Relationship between photosynthesis (μg C l^{-1}) and irradiance (ly h^{-1}) for the 3.5 m population in Lake Vanda and the 4.6 and 9 m populations in Lake Fryxell. The curve for Lake Vanda was drawn by inspection; the curves for the Lake Fryxell samples were derived from the equation of Platt *et al.* (1980). See text for details.

The photosynthate partitioning patterns in the 4.6 and 9 m populations of Lake Fryxell did not show the same relationships with irradiance and both were somewhat different from those observed in Lake Vanda. As in Lake Vanda, protein synthesis in the 4.6 m population was negatively correlated with irradiance but polysaccharide labeling showed an opposite trend (Table I). Both lipid and LMW labeling at 4.6 m in Lake Fryxell had little association with irradiance. A relatively high positive correlation occurred between lipid labeling and irradiance at 9 m in Lake Fryxell, a pattern observed in Lake Vanda. The negative correlation between polysaccharide labeling and irradiance at 9 m in Lake Fryxell also corresponds to the pattern in Lake Vanda.

Relatively strong positive correlations exist between percentage lipid synthesis and photosynthetic activity in samples from both lakes (Table I). Relationships between LMW, polysaccharide and protein labeling and photosynthetic activity were similar in the 3.5 m Lake Vanda and 9 m Lake Fryxell populations; LMW labeling showed relatively high positive correlations with photosynthesis whereas ^{14}C flow into polysaccharide and protein were negatively associated with photosynthesis.

Discussion

The diel labeling patterns we report here for the phytoplankton growing under the relatively constant light of the Antarctic summer are in many ways similar to those reported from lower latitudes where light/dark periods alternate. For example, a number of studies (e.g. Li *et al.*, 1980; Priscu and Goldman, 1983) have shown movement of ^{14}C from storage products, and sometimes LMW metabolites, to protein at night. Our results from Antarctic lakes indicate that the percentage ^{14}C incorporation into protein generally occurs during the period of lowest solar radiation and photosynthesis. Conversely, the highest relative lipid biosynthesis always occurred during periods of high irradiance. The primary difference between our Antarctic lakes and temperate lakes occurs in the polysaccharide labeling patterns. Virtually all studies of temperate phytoplankton have shown that polysaccharide labeling is greatest during periods of high irradiance and photosynthesis (e.g. Morris, 1981; Priscu and Goldman, 1983). Polysaccharide labeling at 3.5 m in Lake Vanda and 9 m in Lake Fryxell were negatively correlated with both irradiance and complete photosynthesis.

We are aware that the incorporation of ^{14}C into the major photosynthetic end-products may be due to pool turnover rather than to the rate of net synthesis (Oaks and Bidwell, 1970). However, because of our relatively long incubation period, the metabolite fractions should have reached equilibrium with the added label. We thus presume that our labeling patterns reflect net gains or losses of the fractions rather than turnover of existing pools. Recent studies have shown that 4–6 h is adequate to equilibrium-label natural populations of phytoplankton growing at rates similar to those estimated at Priscu *et al.* (1987) for Antarctic lake populations (Terry *et al.*, 1983; Priscu and Priscu, 1984).

It is generally believed that microorganisms subjected to a diel day/night cycle synthesize storage materials and LMW metabolites during the day because of an

excess supply of energy over the immediate metabolic requirements of the cells; these constituents are then used during the night for protein synthesis (Cohen and Parnas, 1976). The phytoplankton populations in our study lakes appear to follow this pattern in that the greatest C flow into lipid (a high energy storage product) occurs during periods of highest irradiance. The carbon stored as lipid during this period appears to be used to synthesize protein when irradiance levels diminish. The 4.6 m population in Lake Fryxell varies from this pattern in that polysaccharide is the major storage product synthesized during periods of high irradiance.

Based on theoretical constructs, Cohen and Parnas (1976) suggested that photosynthetic storage materials of microorganisms should be synthesized according to future requirements, taking into account their cost of production. The rather non-deterministic environment experienced by the phytoplankton in Antarctic lakes during the polar summer (e.g. little vertical mixing and relatively constant nutrient and light supply) presumably leads to relatively high ^{14}C incorporation into storage products during periods of relatively high solar radiation at the expense of ^{14}C flow into growth polymers (i.e. protein).

Interestingly, complete photosynthesis at both depths in Lake Fryxell appeared to be inhibited during periods of high 'midday' irradiance; no evidence of inhibition or saturation was observed in Lake Vanda (Figure 4). We parameterized the Lake Fryxell photosynthesis–irradiance relationships with the three-coefficient model of Platt *et al.* (1980) fitted with Marquardt's algorithm. The coefficients obtained from this model were used to compute I_k, which is a single photosynthetic parameter that is influenced by both the maximum rate of photosynthesis (P_m) and the slope of α light limitation (α). I_k, calculated from the identity $I_k = P_m/\alpha$, represents the irradiance level where extrapolation of P_m and α intersect. Talling (1957) introduced this parameter for use as an index of photoadaptation for complete photosynthesis. I_k values computed in this manner were 8.0 and 2.5 ly h^{-1} for the 4.6 and 9 m populations, respectively, in Lake Fryxell. Assuming (i) an average solar wavelength of 550 nm and (ii) that half of the solar radiation is available for photosynthesis, and (iii) taking into account the irradiance level at which the samples were incubated, the values can be converted to Photon Flux Densities of 0.43 and 0.07 uE m^{-2} s^{-1}. These I_k values are within the range of experimental values reported by Priscu *et al.* (1987), supporting their contention that phytoplankton populations in these lakes are among the most shade-adapted yet reported.

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