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Lipid production in natural phytoplankton communities in a small freshwater Baltic lake, Lake Schöhsee, Germany

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SUMMARY

1. Seasonal patterns in lipid production and the photosynthetic parameters describing lipid production in Lake Schöhsee (a small freshwater, mono-dimictic and moderately eutrophic North German Baltic lake) were determined.
2. The mean lipid fraction of C fixation (LIP-FCF) was 13.2% (range 7.6–21%), measured bimonthly from January to December 1995. Periods of high nutrient concentration, low temperatures and particular species of algae (diatoms and/or cryptophytes), were associated with the highest LIP-FCF values.
3. None of the static indicators of nutrient status [dissolved nitrate, soluble reactive silica (SRS); C/N or N/P] are apparently related to the allocation of carbon to lipid in Lake Schöhsee.
4. The light saturation parameters (I_k : I_k-LIP) indicated that carbon fixation into lipid was saturated at much lower light than that of total carbon fixation. This suggested that carbon fixation into lipid was more light efficient than total carbon fixation.
5. The relative allocation of carbohydrate and protein was related to daylength and temperature, respectively.

Introduction

The production and storage of lipids by phytoplankton is regulated by environmental factors in a non-systematic and species-specific manner. Despite years of research on the subject, pioneered by Spoehr & Milner (1949), Collyer & Fogg (1955), Iwamoto, Yonekawa & Asai (1955), Fogg (1956), Iwamoto & Suzuki (1958), Klyachko-Gurvich *et al.* (1967) and Zhukova *et al.* (1969) few accurate generalizations have emerged regarding the regulation of this essential cellular component.

One pattern that has become apparent is the enhancement of lipid storage during nitrogen- and silica-deficient conditions. This has been demonstrated mainly in green algae (Iwamoto *et al*., 1955 Iwamoto & Suzuki, 1958; Fogg, 1959; Zhhukova *et al.* 1969; Ben-Amotz,Tornabene & Thomas, 1985; Suen *et al*., 1987; Renaud *et al.*, 1991), but also in some diatoms (Badour & Gergis, 1965; Opute, 1974; Shifrin & Chisholm, 1981; Taguchi, Hirata & Laws, 1987) and cyanobacteria (deVasconcelos & Fay, 1974). Nitrogen supply has also been noted to influence the relative distribution of fatty acids (FA) in several green algae (Pohl, 1974). Moreover, lipid storage has been shown to be influenced by the form of nitrogen supplied to the cells and by the duration of the N-stress period (Conover, 1975), as well as by culturing conditions (Richardson, 1969).

In marine ecosystems, an increase in the concentra-

tion of particulate lipid often accompanies nutrient depletion associated with unialgal blooms (Kattner, Gercken & Eberlein, 1983; Parrish, 1987). In freshwater ecosystems, Groeger & Kimmel (1988, 1989); Groeger & Tietjan (1993) and Arts, Robarts & Evans (1997), reported that nutrient limitation may shift the allocation of recently fixed algal carbon to lipid.

In a study of three mesotrophic to oligotrophic headwater Canadian lakes, Wainman & Lean (1992) found that the proportion of carbon entering the lipid pool was related to water temperature and daylength. They further report that allocation of recently fixed carbon into lipid components was not correlated with nutrient concentration.

Lipid storage is sensitive to a wide range of environmental factors. Consequently, different case studies, different laboratory conditions and different analytical techniques can all lead to discrepancies between reported values. Therefore, whilst individual studies may help to shed light on certain trends in lipid production, caution must be applied when making generalizations and broad-sweeping predictions.

Algal lipid production is of central importance to a number of aquatic processes. The calorific content of lipids is very high, and their production by photoautotrophs provides a primary source of energy, as well as essential FA, to the foodweb; for example, many species of herbivorous zooplankton and benthic invertebrates rely particularly heavily on algal lipids (Goulden & Hornig, 1980; Ahlgren *et al.*, 1990; Wainman *et al.*, 1993; Müller-Navarra, 1995) because they cannot themselves synthesize sufficient amounts of lipids for reproduction and survival.

In natural populations of planktonic algae, seasonal trends in lipid synthesis, the photosynthetic parameters describing this production, and the variation among lakes are largely unknown. The aim of this study therefore was to determine whether lipid production is related to static indicators of nutrient status, or the relative abundance of species which can synthesize large amounts of lipid. The study was conducted in a small freshwater lake.

Materials and methods

In lake water it is not possible, at present, to separate algae quantitatively from dead cells, detritus and heterotrophs which make up a significant proportion of the biomass. For these reasons patterns of synthesis of particulate macromolecular fractions may have a questionable relationship with algal contents and production in natural waters.

Water samples were collected between 08.00 and 09.00 h from Lake Schöhsee, an oligo-mesotrophic Baltic lake (54°10′N, 10°27′E; maximum depth = 30.2 m; mean depth = 13.0 m; surface area = 82 ha) at 2-m depth using a 5-l PVC Niskin bottle. The water was prefiltered through 35-μm mesh Nytal screen, to remove large, inedible, net phytoplankton and macrozooplankton. The water samples were then transported in Nalgene polyethylene carboys back to the laboratory.

Because of the low algal biomass [1.1–8.4 μg^{-1} chlorophyll *a* (Chl *a*)] it was necessary to concentrate the phytoplankton in order to obtain reliable measurements of their metabolic processes. A versatile technique (tangential flow filtration) was used to facilitate the concentration of phytoplankton samples while the cells were kept in suspension. An 'Ultrasette' Tangential Filtration Device (Omega Series; Filtron Technology Corporation, MA, U.S.A.) was used, which is a self-contained open-channel filter, with a filter area of 700 cm^2. It was especially designed for harvesting or cell removal and has a polyethersulphone membrane with a molecular weight cut-off at 1×10^6 Da, and a pressure range of 5–60 p.s.i. Samples of 20–30 l were concentrated using the above-mentioned filtration device to about 3 l. The retentate was stored at ambient lake temperatures.

Experimental incubations

Retantate was poured into a 2.5-l dispenser and spiked with sterile ^{14}C-labelled sodium bicarbonate to obtain a specific activity of between 5 and 6×10^5 d.p.m. ml^{-1}. Less than 1 h elapsed between sampling and incubation and the total temperature change during this time was < 2 °C. After thorough mixing, two 0.5-ml aliquots were pipetted into scintillation vials for the determination of specific activity. In addition, two 60–65-ml aliquots were taken at time zero (as controls) filtered through pre-combusted (450 °C for 4 h) glass fibre filters (Whatman GF/F), and rinsed three times with 10 ml of sterile, distilled water. The plankton-covered filters were immediately frozen and stored (–20 °C) in Eppendorf micro test tubes with safety lid locks until analysis. Aliquots (60 ml) were then quickly dispensed by an automatic dispenser into sterile glass

Table 1 Concentration and composition of nutrients and elemental ratios (all in $\mu g\ \mu g^{-1}$) of Lake Schöhsee

Sampling date	Days of the year	SRP* ($\mu g\ l^{-1}$)	Total P ($\mu g\ l^{-1}$)	Nitrite ($\mu g\ l^{-1}$)	Nitrate ($\mu g\ l^{-1}$)	Ammonia ($\mu g\ l^{-1}$)	Silica** soluble ($\mu g\ l^{-1}$)	Silica total ($\mu g\ l^{-1}$)	Inorganic carbon ($mg\ l^{-1}$)	C/N	N/P	C/P	Temp. (°C)
03.01	3	9.3	19.7	1.1	162.6	10.6	270.3	316.2	17.3	99.3	8.85	878.2	4.0
17.01	17	8.5	16.5	1.1	167.4	9.7	287.7	322.6	19.3	108.3	10.80	1169.7	2.2
01.02	32	6	14.1	1.2	201.4	11.9	261.9	281.4	16.2	75.5	15.21	1148.9	2.6
15.02	46	5.6	14.9	1.3	212.6	9.7	231.6	287.2	17.6	78.7	15.01	1181.2	3.2
08.03	67	0	9.7	1.1	182	11.7	2.7	27.3	18.4	94.5	20.08	1896.9	3.4
22.03	81	0	8.9	1	165.8	13.2	13.7	58.3	17	94.4	20.22	1910.1	3.6
05.04	95	1.6	12.5	1.1	143.8	10	17.9	33.8	18.3	118.1	12.39	1464.0	4.7
19.04	109	0.4	11.7	1.8	131.7	9.7	26.9	45.4	15.7	109.6	12.24	1341.9	5.9
8.06	158	0.4	11.9	1	66.2	9.3	26.9	40.9	20.8	271.9	6.43	1747.9	15.4
27.09	272	0	10.9	0.2	2.3	8.2	275.5	257.4	13.8	1289.7	0.98	1266.1	14.5
04.10	279	1.2	12.1	0.8	7.5	11.2	307.7	307.7	13.6	697.4	1.61	1124.0	13.1
11.10	286	0	12.9	0.3	4.9	9.1	332.2	325.8	14.4	1007.0	1.11	1116.3	11.6
25.10	300	1.6	14.9	1.5	9.5	11.9	275.1	343.8	13.1	572.1	1.54	879.2	11.4
01.11	307	2.4	12.9	1.6	18.5	25.2	335.4	290.9	13.5	298.0	3.51	1046.5	9.1
15.11	321	4.8	16.1	2.2	23.8	49.8	288.4	312.2	16.7	220.3	4.71	1037.3	7.8
29.11	335	5.6	13.7	2.5	53.9	45.5	255.5	267.2	16.5	161.9	7.44	1204.4	7.3
14.12	350	7.2	17.3	2.2	105.9	29.2	247.1	282.6	16.5	120.2	7.94	953.8	6.5

* = soluble reactive phosphate; ** = SRS.

sample vessels which were placed in a Phototron (light-gradient incubator; Rai & Krambeck, 1992) and the incubation started. The Phototron is designed to maintain a constant temperature (± 0.5 °C). Glass incubation bottles (outer diameter = 4.0 cm; height = 8.5 cm; volume = 75 ml) are inserted into bored holes in the temperature-regulated aluminium block. Irradiation was provided by 12 V halogen lamps (Philips Model 6434 FR, 20 W, 12 V, 18°) mounted under each glass vessel. The light could be adjusted individually in each glass bottle using perforated metal sieves of different mesh sizes, and/or neutral-density filters (Schott, NG-series) mounted in a tray held between the halogen lamps and the incubation bottles. A laboratory Quantum Scalar Meter, equipped with a 1.9-cm diameter solid Teflon sphere irradiance collector (Model QSL-100, Biospherical Instruments Inc., San Diego, CA, U.S.A.) was used to determine the photon flux density experienced by the plankton during the incubations.

Two replicate aliquots were incubated at each of nine levels of incident irradiation (including dark) ranging from 10 to 1700 $\mu mol\ m^{-2}\ s^{-1}$. The samples were shaken throughout the 3-h incubation and maintained at ambient lake temperatures. After 3 h, samples (45–55 ml) were taken from each glass bottle and immediately filtered on to pre-combusted GF/F filters, rinsed and stored at –20 °C until analysis.

Allocation of photoassimilated carbon into lipid and other major metabolic fractions

The frozen, labelled, particulate matter on each GF/F filter was macerated to a fine pulp with glass pestles that fit into conical centrifuge tubes. The initial extraction was made using high pressure liquid chromatography (HPLC)-grade dichloromethane : methanol (2 : 1 v/v). After grinding, the suspension was vortexed for 1 min and then extracted for a further 10 min at 4 °C. The contents of the centrifuge tube were filtered through a Whatman glass fibre (GF/F) filter. The filter was washed with 1.5 ml of the dichloromethane : methanol solution. The combined filtrate was washed with a 0.88% KCL solution comprising 20% of the total volume (Folch, Lees & Sloane-Stanley, 1957) and again vortexed for 1 min. This was followed by centrifugation at 5500–6000 r.p.m. for 10 min at 4 °C. Duplicate aliquots of the dichloromethane fraction (lipid) were removed using an Eppendorf pipette and dried in a scintillation vial. Scintillation cocktail (3.5 ml Instagell II containing 5% carbosorb) was added and the ^{14}C in the sample was quantified using a Canberra-Packard Tri-Carb liquid scintillation analyser (Model, 1900 CA). Similarly, aliquots of the upper, aqueous, methanol layer fraction were also removed in duplicate with an Eppendorf pipette, dried in a scintillation vial, and treated as

above. This modified method (Folch *et al.*1957) extracts lipids quantitatively (Christie, 1982; Wainman & Lean, 1990, 1992; H. Rai & N. Dockal, unpublished information).

Further fractionation was carried out using the subcellular macromolecular fractionation procedure of Li, Glover & Morris, (1980), Hitchcock, (1983) and McConville, Mitchell & Wetherbee, (1985). The rest of the material with the extracted pellet was again filtered through a GF/F filter. The filter with suspended material was resuspended in 4 ml of 5% trichloroacetic acid (TCA) at 95 °C for 30 min. The suspension was filtered through another pre-combusted GF/F filter and 4 ml of 5% TCA was added to wash the filter. Duplicate 1-ml aliquots of the filtrate were then dried in scintillation vials. The dry residue was suspended in 1 ml of distilled water before the scintillation fluid was added. TCA-insoluble material, which collected on the filter, was placed directly into a vial and scintillation fluid (Filter Count™ complete LSC-Cocktail, Packard Instrument Co., Downers Grove, IL, U.S.A.) was added. Quench corrections were made automatically using an external standard. An isotopic determination factor of 1.06 was used as suggested by Steemann Nielsen (1952). ^{14}C uptake rates were calculated according to Rai (1982). No separate measures of total carbon assimilation were made during this study, since previous investigations have consistently shown that the extraction techniques employed here result in a complete recovery of radiolabel (Palmissano & Sullivan, 1985; Rai, 1993). Data reported herein for total uptake are the sum of the different extracted fractions. All radioactivities were subsequently corrected for the initial or time zero values.

Photosynthetic rates from each of the two replicates at nine light levels were pooled to construct photosynthetic vs. light intensity (*P–I*) curves. The response of each macromolecular fraction in each sample to light was calculated by fitting the relation between incorporation rate and light intensity to the following function (Platt, Gallegos & Harrison, 1980):

$$P^B = P_S^{B*} (I\text{–}e^{-a})^* e^{-b}$$

where $a = \alpha I/P_S^B$, and $b = \beta I/P_S^B$. P^B (μg C (μg Chl-*a*)$^{-1}$ h^{-1}) is the specific photosynthetic rate at irradiance *I*. The slope of the light-limited portion of each *P–I* curve is designated α. The parameter α is an index of light utilization efficiency. In the present study, α has units of (μg C) (μg^{-1} Chl-*a*)$^{-1}$ (μmol m^{-2} s^{-1})$^{-1}$. P_S^B is the maximum potential photosynthetic rate and has units of μg C μg^{-1} Chl-*a* h^{-1}. I_k is the light saturation parameter (Talling, 1957), defined as P_{max}^B/α with units of μmol m^{-2} s^{-1}. The parameter β is the negative slope at high irradiance and is the index of photoinhibition at higher irradiance (same units as α). P_{max}^B, the maximum photosynthetic rate at high saturation, was determined from P_S^B with the following equation:

$$P_{max}^B = P_S^{B*} (\alpha/(\alpha + \beta))^* (\beta/(\alpha + \beta))^{\beta/\alpha}.$$

All parameters reported here are chlorophyll-*a* specific values, but superscript B has been eliminated from the parameters α, β and P_{max} to coincide with general usage. LIP_{max}, α-LIP, and I_k-LIP; LMW_{max}, α-LMW, and I_k-LMW; POL_{max}, α-POL, and I_k-POL; PRT_{max}, α-PRT, and I_k-PRT are the lipid (LIP), low molecular weight metabolites (LMW), polysaccharide (POL), and protein (PRT) analogues of the *P–I* parameters, respectively.

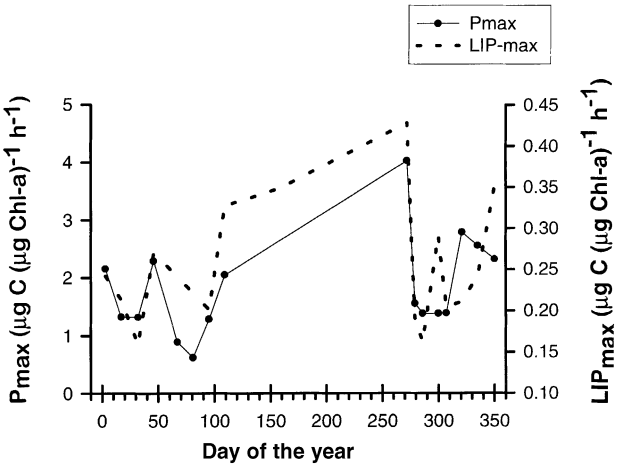

Fig. 1 Seasonal patterns in the chlorophyll *a*-specific rates of carbon (C) fixation (P_{max}) and its biomolecular analogue for lipid (LIP_{max}) for Lake Schöhsee in 1995.

Water chemistry

Chlorophyll-*a* values reported here were corrected for phaeopigments (Marker, Nusch & Rai, 1980). Total P, soluble reactive P (SRP), NO_3-, NO_2-, Kjeldahl N, soluble reactive silica (SRS) and total silica, were measured according to Grasshoff, Ehrhard & Kremling, (1983). Inorganic carbon (Table 1) was determined using an infrared CO_2 analyser (Tocomaster model 915B, Beckman). Cell concentrations of seston were determined immediately from field-collected

Table 2 Chlorophyll-specific rate of C fixation (P_{max}) and C fixation into lipid (LIP_{max}), their photosynthetic parameters*, ratios and lipid fraction of carbon fixation (LIP-FCF)

Date	Days of the year	α	α-LIP	P_{max}	LIP_{max}	I_k	I_k-LIP	α/α-LIP	I_k/I_k-LIP	LIP-FCF
03.01	3	0.0198	0.0174	2.1556	0.242	109	14	1.14	7.8	11.2
17.01	17	0.0187	0.0047	1.3234	0.2133	71	45	3.98	1.6	16.1
01.02	32	0.0123	0.0037	1.3212	0.1594	107	43	3.32	2.5	12.1
15.02	46	0.0149	0.0032	2.2893	0.2709	154	85	4.66	1.8	11.8
08.03	67	0.1276		0.8927						
22.03	81	0.109		0.6178						
05.04	95	0.0159	0.0067	1.2857	0.2016	81	30	2.37	2.7	15.7
19.04	109	0.0225	0.0075	2.0476	0.3274	91	44	3.00	2.1	16.0
08.06	158	0.0227	0.0028	2.2965	0.3488	101	125	8.11	0.8	15.2
29.09	272	0.056	0.0079	4.0302	0.428	72	54	7.09	1.3	10.6
04.10	279	0.0134	0.0015	1.5525	0.1912	116	127	8.93	0.9	12.3
11.10	286	0.0221	0.0035	1.381	0.1623	62	46	6.31	1.3	11.8
25.10	300	0.0135	0.0029	1.3811	0.2907	102	100	4.66	1.0	21.0
01.11	307	0.0151	0.0036	1.3883	0.205	92	57	4.19	1.6	14.8
15.11	321	0.0137	0.0029	2.787	0.2106	203	73	4.72	2.8	7.6
29.11	335	0.0199	0.0174	2.5536	0.242	128	14	1.14	9.2	9.5
14.12	350	0.0211	0.0117	2.3205	0.3514	110	30	1.80	3.7	15.1

*The lipid equivalents to the photosynthetic parameters were determined in the same fashion using the amount of C fixed into the lipid extract.
For photosynthetic parameters definition see text.

Table 3 The mean percentage of ^{14}C incorporated into lipids for natural phytoplankton communities (LIP-FCF, lipid fraction of carbon fixation)

Study sites	References	LIP-FCF (%)
Schöhsee	Present study	16.5
Lake Michigan	Fahnenstiel *et al*. (1989)	24.7
Lake Huron	Fahnenstiel *et al*. (1989)	21.3
Lake Ontario	Cuhel *et al*. (1984)	38
Jack Lake	Wainman & Lean (1992)	15.6
Anstruther Lake	Wainman & Lean (1992)	18.2
Bay Lake	Wainman & Lean (1992)	14.9
Lake Hylke and Lake Orn	Jensen, 1985	15–28
Lake Pavian	Amblard & Bourdier (1990)	14–24
Eastern Mediterranean	H. Rai & N. Dockel (unpublished data)	23.7
In other regions of the world		
New Zealand Coastal Systems Eastern Canadian Arctic Coastal Oceanic regions of Caribbean Sea and Western Atlantic Ocean	Upwelling (Priscu & Priscu, 1984); (Li & Platt, 1982); (Morris, Smith & Glover, 1981).	13–26

samples using an electronic particle counter (CASY 1; Cell Analyser System, Schärfe System, Reutlingen, Germany).

Results

The chlorophyll-specific rate of total C fixation (P_{max}) varied between 1.29 and 4.03 (μg C (μg Chl-*a*)$^{-1}$ h^{-1}) (mean = 2.04). The chlorophyll-specific rate of C fixation into lipid (LIP_{max}) ranged from 0.159 to 0.428 (μg C (μg Chl-*a*)$^{-1}$ h^{-1}) (mean = 0.242). Major peaks in LIP_{max} and P_{max} occurred in early spring and winter when the phytoplankton community was dominated by diatoms and cryptophytes and thus would be expected to have a relatively large amount of allocation to lipid (Fig. 1 and Table 2). LIP_{max} was apparently not related to any of the static indicators of nutrient status or to physical variables like water temperature and daylength (Table 3).

The ratio of α to α-LIP is a measure of the relative rate of C fixation to lipid C fixation. α-LIP was higher at lower temperatures, whereas α did not show definite peaks throughout the period of study. These trends in α and α-LIP values caused the ratios to remain high throughout the study period, however, lower

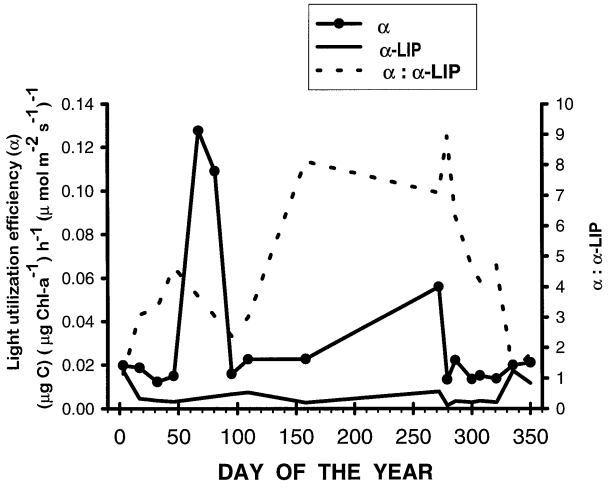

Fig. 2 Seasonal patterns in the light utilization efficiency (α), its lipid analogue (α-LIP), and α : α-LIP ratio for Lake Schöhsee in 1995.

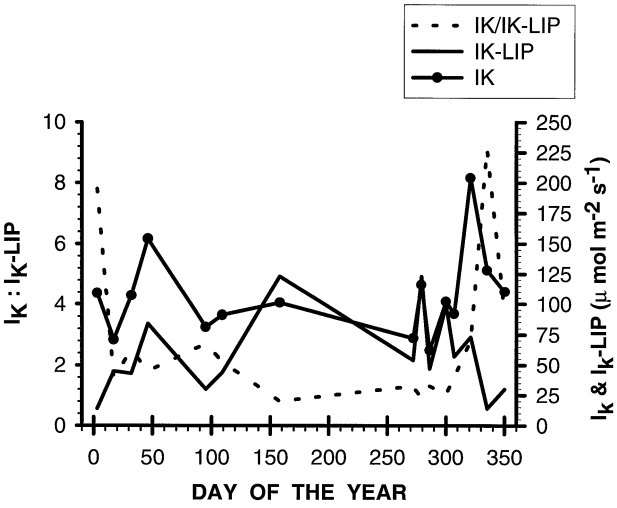

Fig. 3 Seasonal patterns in light intensity at the intercept of α and P_{max} (I_k), its lipid analogue (I_k-LIP), and the ratio I_k : I_k-LIP for Lake Schöhsee in 1995.

α : α-LIP ratios were observed during the low water temperature periods (Fig. 2 and Table 2).

The light saturation ratio I_k : I_k-LIP in Schöhsee increased during the winter and early spring when water temperatures were very low. I_k was always higher than I_k-LIP indicating C fixation into lipid was light saturated before C fixation throughout the period of study (Fig. 3 and Table 2).

The LIP-FCF generally shows distinct peaks during winter and early spring (Fig. 4). The seasonal mean LIP-FCF values varied between 7.6 and 21.0 (mean 13.4). LIP-FCF was apparently not related to any of the static indicators of nutrient status. There were

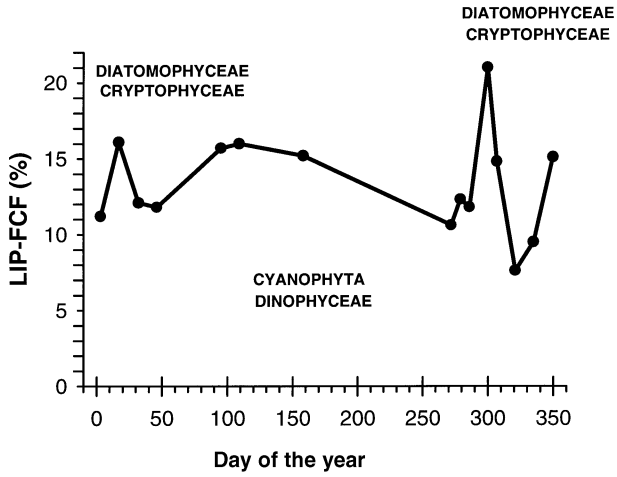

Fig. 4 Seasonal pattern in the relative allocation of ^{14}C–CO_2–C into the lipid fraction (LIP-FCF) of Lake Schöhsee phytoplankton in 1995.

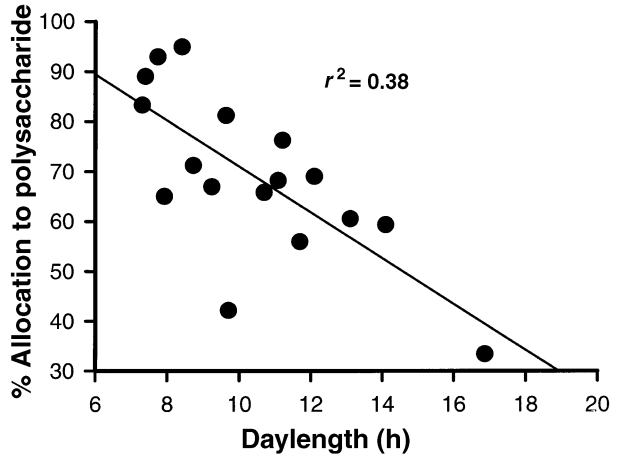

Fig. 5 Seasonal pattern in the relative allocation of ^{14}C–CO_2–C into the polysaccharide fraction (POL-FCF) of Lake Schöhsee phytoplankton as a function of daylength in 1995.

no significant correlations between the three lipid parameters, P_{max}, LIP_{max} and LIP-FCF and any measure of the availability of nutrients (Si, N, P, inorganic C) or elemental ratios (C/N, N/P, C/P). Furthermore, the three lipid parameters were not correlated to temperature or day length.

The POL-FCF (Fig. 5) varied between 33.4 and 94.9 (mean = 69.1). POL-FCF generally decreased with increasing daylength (h) and was significantly correlated with increasing daylength (r^2 = 0.38, n = 17). The PRT-FCF (Fig. 6) values varied between 6.6 and 25.2 (mean = 15.47). PRT-FCF generally increased with increasing water temperature and was significantly correlated with water temperature (r^2 = 0.27, n = 17).

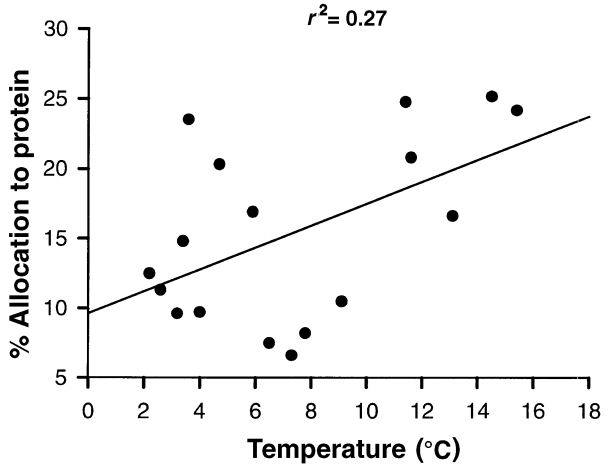

Fig. 6 Seasonal pattern in the relative incorporation of $^{14}C_2$–CO_2–C into the protein fraction (PRT-FCF) of Lake Schöhsee phytoplankton as a function of water temperature (°C) in 1995.

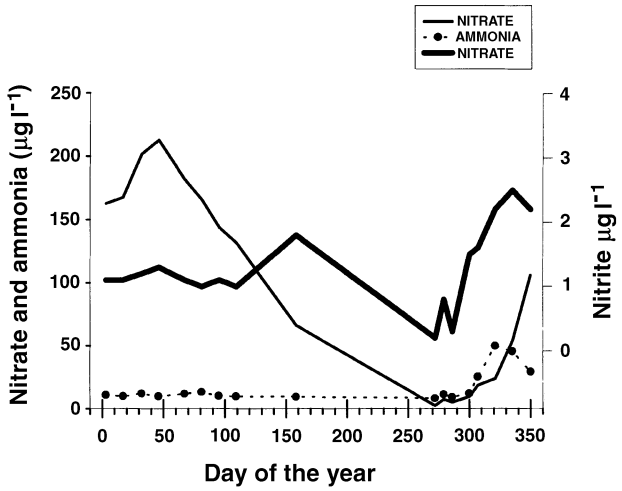

Fig. 7 Nitrate, nitrite and ammonia concentrations (µg l^{-1}) in Lake Schöhsee during 1995.

Discussion

Allocation of recently fixed carbon in freshwater ecosystems is related to a number of physical variables; for example, allocation to protein has been related to temperature (Cuhel & Lean, 1987a), allocation to carbohydrate has been related to daylength (Cuhel & Lean, 1987b), while allocation to lipid has been related to daylength and temperature (Wainman & Lean, 1992). In Lake Schöhsee, the allocation of carbon to the protein and polysaccharide macromolecular fractions appears to be related to physical variables (Fig. 5). Protein allocation is related to temperature (Fig. 6) by the relationship: Percent protein (% PRT) = 9.7 + 0.78*temperature (°C), $n = 17$, $r^2 = 0.27$. This value for the slope is greater than that from other lakes, which range from 0.5 to 0.7, whereas the Y-intercept is similar to Lake Ontario and several small headwater lakes (Wainman & Lean, 1996).

Carbohydrate allocation was related to daylength (Fig. 5) by the relationship: percentage carbohydrate (% POL) = 112–4.31*daylength (h), $n = 17$, $r^2 = 0.38$. The slope of –4.32 (SE = 1.41) is about the same as the slope of –3.5 that has been found for a number of other meso- to oligo-trophic waters (Wainman & Lean, 1996). The Y–intercept of 112.0 (SE = 15.1) is anomalously high compared to the intercept from other lakes of 73.0. However, there are no data in the region of the Y–intercept for Lake Schöhsee and therefore this parameter estimate is prone to error.

In terms of algal biomass, the lipid-rich diatoms and cryptophytes were much more abundant in the spring than in autumn, which seems to coincide with the high α-LIP observed at this time. This suggests that a shift in community structure favouring dominance of diatoms and cryptophytes, such as was observed in Lake Schöhsee in the spring, creates the conditions that favour an algal assemblage that is geared to maximize lipid production as seen by the high α-LIP values at this time. The latter is probably unrelated to nutrients but more related to the pronounced change in community structure compared to the summer when cyanophyta and dianophyceae dominate.

The LIP-FCF in Lake Schöhsee varied between 7.6 and 21.0 (mean = 13.39). The pattern of LIP-FCF was similar to the patterns of the photosynthetic parameter α. α-LIP was inversely proportional to the α : α-LIP ratio which indicates a decrease in the efficiency of C fixation into lipid compared to total C fixation, whenever the ratio of α : α-LIP increased, and vice versa.

The light saturation parameters (I_k and I_k-LIP) suggest that C fixation into lipid was saturated at far lower light intensities than total C fixation throughout the study period (Fig. 3). This suggests C fixation into lipid requires generally less light and was therefore more light efficient compared to C fixation. These results contrast with the data from Wainman & Lean (1992) who found that more light is required for saturation and that carbon fixation into lipid is less light efficient compared to overall C fixation.

Very few existing data for freshwater LIP-FCF are available. Jensen (1985) reported that the LIP-FCF varied between 15 and 20% in a temperate, eutrophic freshwater lake. Amblard & Bourdier (1990) reported

the average LIP-FCF ranged between 14 and 24% for Lake Pavian (subarctica) and suggested, that the difference in LIP-FCF is due to a decrease in temperature and light with depth. Wainman & Lean (1992) reported that the mean seasonal LIP-FCF for three freshwater lakes studied was 15.8%. The mean LIP-FCF in lakes Ontario (Cuhel & Lean, 1987a), Michigan and Huron (Fahnenstiel *et al.*, 1989) are 21.3, 24.7, and 20.7%, respectively, (Table 4).

Madariaga & Joint (1992) have suggested that the ratio of allocation to protein and LMW (low molecular weight metabolites) is a reasonable indicator of nutrient depletion in algae. To test whether this ratio was related to nutrient deficiency, the data were divided into two groups. The first group had low nutrients (nitrate less than 50 $\mu g\, l^{-1}$ or SRS less than 50 $\mu g\, l^{-1}$) and the second high nutrients (greater than 50 $\mu g\, l^{-1}$ nitrate or SRS greater than 50 $\mu g\, l^{-1}$). The protein : LMW material was higher in the low-nutrient group (mean 3.0, SD = 2.2, $n = 11$) than in the high-nutrient group (mean 1.8, SD = 1.6, $n = 6$) but the difference was not significant.

It is clear from work undertaken on lipid synthesis in algal cultures that only certain species of algae, under specific nutrient conditions, will produce large amounts of lipid. The algae species which are prone to increase lipid synthesis are the diatoms, cryptophytes and chrysophytes and then only when either silica or nitrogen or both nitrogen and silica are in short supply. None of the static indicators of nutrient status (dissolved nitrate, SRS, C/N or N/P) is apparently related, at least by simple correlation analysis, to the allocation of carbon to lipid in Lake Schöhsee, which is similar to the observations of Wainman & Lean (1992).

The problem with looking for correlation between any single variable, such as silica, and allocation of photosynthate to lipid is that a number of other factors, like abundance of the appropriate algal species, is very important. Finally, as with many biological systems there is bound to be a timelag between cause and effect. Algae, like other organisms, take time to adapt to their surroundings. So if nutrients decline, we should expect the algae to respond right away, possibly by changing the relative allocation of carbon amongst the main macromolecular pools. Also, it is reasonable to expect these complex communities to take a little time to adapt to the new conditions; for example, nitrite and nitrate levels reached an all-time seasonal low between days 270–290 (Fig. 7). Shortly after this, LIP_{max} and the LIP-FCF increased sharply (Figs 1 and 4, respectively), while, simultaneously, cell numbers declined sharply ($\top$ day 300). Thus, it is clear that simple correlation analyses may fail to identify biological cause and effect if timelags are not taken into account.

Examination of seasonal trends in photosynthetic parameters in natural phytoplankton communities can yield important insights into carbon fluxes to the main macromolecular end-products of photosynthesis. With this information we can begin to correlate the quantity and photosynthetic efficiency of carbon fixation into these end-products with the seasonal changes in phytoplankton community structure. This information, in conjunction with physico-chemical data, permits generalizations about the underlying forces driving carbon through pelagic foodwebs. This is crucial if we are to understand the effects of climate forcing, nutrient cycling, or the effects of anthropogenic stressors on aquatic ecosystems.

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