Grazing by protists and seasonal changes in the size structure of protozooplankton and phytoplankton in a temperate nearshore environment (western Gulf of St. Lawrence, Canada)

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ABSTRACT: Seasonal changes in the biomass and size structure of phytoplankton and protozooplankton and the grazing effect of protozooplankton on phytoplankton were assessed in a temperate nearshore environment (Baie des Chaleurs, western Gulf of St. Lawrence, Canada). The size structure of protozooplankton and phytoplankton assemblages showed relatively similar seasonal changes. During spring, dominance by large (>20 µm) heterotrophic or mixotrophic ciliates and flagellates coincided with high biomass of chain-forming diatoms, suggesting that protists were important grazers of the spring diatom bloom. During summer, concentrations and biomass of ciliates and flagellates >20 µm were lower than they were during spring and, consequently, the relative contribution of protists <20 µm increased. The potential grazing impact of ciliates <20 µm on single celled cyanobacteria was similar to or even higher than that of nanoflagellates. Grazing experiments conducted in situ using the serial dilution technique and pre-screening water on 64 µm showed that 33 to 42% of the chl a biomass and 54 to >100% of phytoplankton production were consumed daily by protozooplankton. Thus, when predation was reduced, protozooplankton were able to control phytoplankton biomass and production. Predation by copepods, which showed high concentrations and were dominated by omnivorous Oithonidae during summer, may have played a central role in the seasonal changes in the size structure and biomass of protozooplankton. The observations and results from path analysis suggest the existence of 2 different food webs corresponding to bloom and summer conditions, respectively.

KEY WORDS: Size structure · Protozooplankton · Ciliates · Flagellates · Grazing · Phytoplankton · Temperate nearshore waters

INTRODUCTION

There is increasing evidence that the microbial food web is a fundamental and almost permanent feature of most oligotrophic and eutrophic marine waters. Some authors now refer to this food web as a maintenance system (Riegman et al. 1993). In the microbial food web, mixotrophic and heterotrophic nanoflagellates and ciliates <20 µm are major predators of bacteria and cyanobacteria (Bernard & Rassoulzadegan 1990, Capriulo 1991). These protozoan grazers are in turn the prey of ciliates >20 µm and dinoflagellates, which

also consume nanoplanktonic and sometimes microplanktonic algae (Capriulo 1990, Pierce & Turner 1992). While most metazoan zooplankton are relatively inefficient at capturing bacterial-sized prey, some are able to feed on protozooplankton (Stoecker & Capuzzo 1990, Sanders & Wickham 1993). The latter thus can act as a link between the smallest members of the microbial food web and larger organisms. In addition, it is generally considered that the high growth rates of protozooplankton should enable them to control assemblages of bacterioplankton and small phytoplankton, at least when temperature does not limit their growth and when predation by metazoa is low (Havens 1993, Riegman et al. 1993).

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Although some protozoan grazers can feed on phytoplankton larger than themselves (e.g. Hansen 1992), it has been demonstrated that protozoa have specific minimum, optimum, and maximum prey sizes, which are generally smaller than themselves (Rassoulzadegan et al. 1988, Lessard 1991). In temperate marine waters, seasonal shifts in the size distributions of autotrophic organisms generally occur throughout the year, between dominance by large cells during the spring and autumn phytoplankton blooms and dominance by ultraplankton during summer and winter (e.g. Durban et al. 1975, Claereboudt et al. 1994). These shifts are likely to influence the size distributions of protozooplankton assemblages and, hence, the overall structure and functioning of the microbial food web. However, there is little information available on changes in the size structure of heterotrophic ciliate and flagellate assemblages in temperate waters. Until recently, most of the data reported on protozooplankton were for organisms in the 20 to 200 μm size range (microzooplankton). For example, the biology of tintinnids (typically 40 to 200 μm) has been widely studied, whereas the distribution and grazing significance of the more delicate aloricate ciliates, particularly forms < 20 μm, remain poorly investigated in marine waters (Pierce & Turner 1992).

Since protozooplankton, especially ciliates, are known to be more abundant in nearshore waters than in the open sea (Campbell & Carpenter 1986, Pierce & Turner 1992), it follows that their role in food webs of nearshore marine waters is expected to be particularly important. However, although the significance of herbivorous protists to the fate of primary production is now widely recognised (Sherr & Sherr 1994), their grazing effects on natural plankton assemblages of temperate nearshore waters are still poorly known. Many studies on the grazing of herbivorous protists have been conducted under controlled laboratory conditions, often using monospecific cultures, so that the amount of data available on specific clearance rates of various protist species is increasing (e.g. Bernard & Rassoulzadegan 1990, Verity 1991, Hansen 1992). In contrast, field studies have generally used the serial dilution method of Landry & Hassett (1982) to determine the grazing activities of microzooplankton assemblages (e.g. Paranjape 1990, Weisse & Scheffel-Möser 1990, McManus & Ederington 1992). In these studies, in addition to herbivorous protozoa, the latter included small metazoan plankters such as copepod nauplii and planktonic larvae of benthic organisms (e.g. bivalves and crustaceans). Since these metazoan organisms can act as competitors and predators of herbivorous protists, it follows that grazing rates determined in such studies generally reflect complex herbivorous and carnivorous interactions (Verity et al. 1993, Sime-Ngando et al. 1995).

The aims of the present study were (1) to examine seasonal changes in the size distributions of ciliate and flagellate assemblages in relation to the size distribution of their potential food (i.e. phytoplankton) and the potential effects of their predators (i.e. mesozooplankton) and (2) to assess the intensity of grazing by protozooplankton on phytoplankton, in the absence of metazoan competitors or predators, using in situ incubations of water passed through a 64 μm mesh. In the present study, special emphasis was put on the smallest forms of ciliates and flagellates.

MATERIALS AND METHODS

Sampling. The study was conducted in the Baie des Chaleurs (western Gulf of St. Lawrence, Canada) from May to October in 1991 and 1992. The sampling station was located at Gascons, on the Québec shore of the bay (48° 10.60' N, 64° 54.80' W). The station (22 m depth, ca 1.8 km offshore) was occupied weekly, always during high tide to minimise variability due to tides. When primary production measurements were performed, additional samples were taken at dawn for determination of chlorophyll a (chl a) and nutrient concentrations, and additional CTD profiles were carried out, independent of the tidal cycle, in order to follow the recommendation of JGOFS (1988) for conducting of 24 h dawn-to-dawn in situ primary production incubations.

Water was collected using an 8 l Niskin bottle at 4 depths (1, 5, 9 and 21 m) in 1991 and 5 depths (1, 5, 9, 15 and 21 m) in 1992. All water samples were prefiltered through a 200 μm Nitex mesh and stored in isothermal containers until they were processed. Processing occurred within 2 h of collection. Temperature and salinity profiles were recorded with a CTD instrument (SBE 19, Seabird Electronics Inc., Bellevue, WA, USA). During summer 1992, several oblique tows were taken from bottom to surface, using a 64 μm net, to determine the composition of mesozooplankton assemblages.

Treatment of water samples. Water samples (250 ml) for determination of chl a and phaeopigment (Phaeo) were size-fractionated using 3-compartment Nuclepore® Swin-Lok 47 mm filter towers, with a 20 μm Nitex filter at the top, a 5 μm Nitex filter in the middle, and a Whatman GF/F filter at the bottom. Chl a retention properties of glass-fiber GF/F filters are similar to those of 0.2 μm membrane filters (Chavez et al. 1995). Filtration provided 3 size fractions, i.e. 0.2-5 μm, 5-20 μm, and 20-200 μm. However, since seasonal changes in the size fractions 5-20 μm and 20-200 μm were similar and the size fraction 5-20 μm was generally <10% of the total

chl *a* biomass, these 2 size fractions were pooled and are referred to in this paper as the >5 µm fraction. Filtration was under vacuum pressure <100 mm Hg. Filters for chl *a* and Phaeo were kept at −20°C until analysis. They were subsequently extracted in 90% acetone during 24 h in the dark at 0°C, and pigment concentrations were determined with a Turner Fluorometer 112 using the fluorometric method of Yentsch & Menzel (1963) and following the procedures given by Parsons et al. (1984).

During the 1992 sampling season, several primary production measurements were conducted *in situ*, from dawn to dawn, together with the grazing experiments. Nalgene 250 ml, acid-cleaned tissue culture flasks were filled with seawater from each sampling depth and inoculated with 10 µCi $H^{14}CO_3^-$. At each of the 5 sampling depths, 2 dark and 2 light bottles were incubated horizontally for 24 h (dawn to dawn) using a plexiglass frame. Processing of the samples followed the procedure described in Tamigneaux et al. (1995).

Water for determination of nitrate, nitrite, and phosphate was filtered on Whatman GF/F filters before analysis. To avoid possible contamination, water for determination of silicate was not filtered. All nutrient samples were frozen (−20°C) in 4-ml cryovials. Analyses were on a Alpkem AutoAnalyzer (Alpkem Inc., Wilsonville, OR, USA), following the colorimetric methods of Parsons et al. (1984).

Grazing experiments. In order to estimate the grazing of phytoplankton by protozooplankton, 7 experiments were performed between May and August 1992, using the dilution method of Landry & Hassett (1982). Diluted samples were incubated at 5 m depth at the sampling site, over 24 h from dawn to dawn. The day before each experiment, 35 l of seawater from 5 m depth was brought to the laboratory and filtered, first through a Whatman GF/F filter and then through a Gelman 0.2 µm 'minicapsule' filter. The particle-free water was kept at 10°C in darkness until further processing. On the day of the experiments, 5 dilution mixtures (2 l) were prepared with freshly sampled water from 5 m depth and the particle-free seawater, at ratios of 100:0, 90:10, 80:20, 60:40, 20:80 and 0:100 (particle-free control; Li 1990). Each treatment (i.e. each dilution mixture and undiluted control) was gently poured through an immersed 64 µm mesh in order to eliminate the smallest metazoa, e.g. copepod nauplii and copepodites. Finally, acid-cleaned, glass reagent bottles (1 l) were filled to the brim with each treatment and sealed carefully in order to exclude air bubbles. At the beginning and end of each experiment, samples for chl *a* (250 ml) and protozooplankton enumeration (100 ml) were taken from each dilution treatment. Only chl *a* was determined for the particle-free control.

Nutrient concentrations were determined for the undiluted sample at the start and end of the 24 h incubation in order to identify possible nutrient limitation over the 24 h time span.

The apparent growth rate (*k*) of chl *a* in individual bottles was calculated using the exponential model given by Landry & Hassett (1982):

$$P_t = P_0 \exp[k - g]t$$

where P_0 and P_t are chl *a* concentrations at the beginning and end of the experiment, respectively; *k* and *g* are instantaneous coefficients (d^{-1}) of phytoplankton growth and grazing mortality, respectively; *t* is time (d). Coefficients *k* and *g* were determined from linear regression of the apparent growth rate of phytoplankton ($1/t \ln[P_t/P_0]$) on the fraction of undiluted seawater. The negative slope of the line is the daily grazing coefficient (*g*) and the *y*-intercept is the phytoplankton growth rate (*k*). The null hypothesis (H_0) $g = 0$ was *t*-tested. Results from the serial dilution experiments were used to calculate the numbers of doublings per day of phytoplankton (Antia 1991):

$$\text{Doublings d}^{-1} = \frac{k}{\ln 2}$$

These were compared with estimates derived from chl *a* and primary production, calculated using the following formula (Antia 1991):

$$\text{Doublings d}^{-1} = \frac{\text{Primary production (µg C l}^{-1}\text{ d}^{-1})}{[\text{chl } a \text{ (µg l}^{-1}) \times 40(\text{µg C µg}^{-1}\text{ chl } a)]}$$

Laboratory measurements. In 1992, samples (100 ml) for enumeration of ultraphytoplankton (<5 µm) were fixed with paraformaldehyde (0.2% final conc.) and stored for at least 1 h in the dark at 5°C before further processing. Phototrophic eucaryotes <5 µm and cyanobacteria were enumerated under the epifluorescence microscope and their carbon biomasses were estimated following the procedure described in Tamigneaux et al. (1995). In 1991 and 1992, samples (250 ml) for enumeration of phytoplankton >5 µm and protozooplankton were preserved with 1% (vol:vol) acidic Lugol's solution. Within 6 mo, 100 ml subsamples were sedimented over 30 h before enumeration under the inverted microscope (Lund et al. 1958). Magnification was chosen according to cell size, i.e. 600× for cells less than 20 µm, 300× for cells 20–50 µm, and 100× for larger organisms. In 1992, samples for the determination of mesozooplankton (500 ml) were preserved with buffered 5% formalin.

Lugol's iodine is recognised as a good fixative for accurately estimating ciliate biomass (Jerome et al. 1993). However, there are limitations to the use of this preservative for protozooplankton, i.e. identification of protists to species level is difficult and discrimination be-

tween heterotrophic and autotrophic forms is not always possible. Distinguishing between strict phototrophs, mixotrophs, and strict heterotrophs using epifluorescence microscopy would have been valuable. However, while the presence of chlorophyll fluorescence in flagellates may indicate phototrophy, it is not proof (see Stoecker 1991 on chloroplast enslavement). In addition, the trophic status of most dinoflagellates (Larsen & Sournia 1991) and nanoflagellates (Capriulo et al. 1991, Hall et al. 1993) is under revision, since many phytoflagellates now appear to be mixotrophic (Jones 1994). The trophic status of the identified dinoflagellate genera was set according to Larsen & Sournia (1991). Among planktonic ciliates, *Myrionecta rubrum*, which is easily identified, is the only species known to be strictly autotrophic, whereas others are considered mixotrophic or heterotrophic (Pierce & Turner 1992).

During the seasonal survey of 1991, protozooplankton were only identified into broad taxonomic groups, i.e. aloricate choreotrich ciliates, choreotrich ciliates with a lorica (tintinnids), and microflagellates. Cell dimensions were not measured. In 1992, protozooplankton were identified into species, genera, or broader taxonomic groups and cell dimensions were measured and cells separated into size classes (<20, 20–50 and >50 µm) for better estimating their trophic roles (Rassoulzadegan et al. 1988, Bernard & Rassoulzadegan 1990). The term 'choreotrichs' refers herein to the subclass Choreotrichia in the phylum Ciliophora and it includes orders Choreotrichida and Oligotrichida (Montagnes & Lynn 1991).

In order to determine protozoan cell volumes, the length and width of 50 cells of each taxonomic group were measured at 600× using a calibrated ocular micrometer. Cell volumes were determined by applying standard geometric shapes to cells. Carbon biomasses were calculated using a conversion factor of 0.15 pg C μm^{-3} for flagellates (Sheldon et al. 1986) and 0.19 pg C μm^{-3} for ciliates (Putt & Stoecker 1989).

Path analysis. Path analysis was used to elucidate the importance of direct links between protist abundances and concentrations of their potential food particles. Path analysis allows us to state hypotheses on causal relationships among variables and to assess the consequences of the assumed relationships using coefficients of linear regression and correlation computed among the variables (Sokal & Rohlf 1981, Legendre & Legendre 1984). Also, the percentages of non causal relationships (spurious correlations) associated with path coefficients can be computed. Path analysis was carried out using the R package for multivariate data analysis (Legendre & Vaudor 1991). Path coefficients are interpreted by examining their signs and relative magnitudes, rather than using their individual statistical significance (Sokal & Rohlf 1981).

RESULTS

Oceanographic conditions

Oceanographic conditions at the sampling station were discussed in detail by Tamigneaux et al. (1995). During the sampling seasons of 1991 and 1992, water temperature ranged from 0°C at the beginning of May to 15–20°C at the end of July and in August. Salinity ranged from 31 PSU at the beginning of May to 26 PSU at the end of June, when freshwater runoff was strong. The water column was generally stratified from the end of May to the end of August and the surface mixed layer seldom extended deeper than 5 to 10 m during this period.

For both years, the spring phytoplankton bloom was observed in May and June (Figs. 1 & 2). At this time, phytoplankton >5 µm accounted for >60% of the total chl *a* biomass, which reached up to 20 mg m^{-3} The bloom was dominated by *Chaetoceros* spp. and *Thalassiosira* spp. in chains, with individual cells ≤30 µm. At the end of the bloom, chains of cells started to aggregate and there was massive sinkage of resulting flocculates to the bottom (observed by scientific SCUBA divers at the sampling site). During the summers of the 2 years, phytoplankton <5 µm accounted for >90% of the total chl *a* biomass, which was generally <2.5 mg m^{-3} (Figs. 1 & 2). Concentrations of cyanobacteria reached their highest values during summer (Fig. 2b) and no colonial forms were observed throughout the season. Autumn blooms of phytoplankton (September and October) were somewhat less pronounced, with chl *a* concentrations <7 mg m^{-3}. The autumn phytoplankton bloom in October 1992 was dominated by thecate dinoflagellates *Ceratium* sp.

Dynamics of protozoa in 1991

Throughout the 1991 sampling season, protozoa were numerically dominated by flagellates 2 to 200 µm, with concentrations ranging from 2.2×10^3 to 5.6×10^6 cells l^{-1} (mean 3.7×10^5) (Fig. 1c). Over the 22 m water column (areal concentrations), flagellated cells always accounted for ca 95% of the protozoan assemblage, the remaining 5% being ciliates (tintinnids and aloricate ciliates). During summer, the flagellate assemblage was almost entirely dominated by nanoflagellates. In the ciliate assemblage, aloricate choreotrichs were always dominant, with concentrations ranging from 1.2×10^2 to 2.8×10^4 cells l^{-1} (average 4.5×10^3) (Fig. 1d). Tintinnids ($<10^2$ to 1.9×10^3 cells l^{-1}, average 5.2×10^2) made up, on average, 19% of the ciliates, and were thus 4 times less abundant than the aloricate choreotrichs (Fig 1e). Throughout the sea-

son, concentrations of both microflagellates and aloricate choreotrichs were always higher in the upper half of the water column (0 to 10 m) than below (Fig. 1).

During the second fortnight of June, abundances of aloricate choreotrichs and tintinnids peaked in the upper part of the water column (Fig. 1d, e). The rapid increase in the concentration of ciliates corresponded to a peak concentration of chl a in the >5 µm fraction, i.e. the spring diatom bloom (Fig. 1a, b). Following this peak, ciliates remained at low concentrations throughout the summer. Ciliate numbers started to increase again in autumn, when large phytoplankton resumed a dominant role. Flagellate numbers (Fig 1c) increased at the end of June after the ciliate peak, continued to rise through the end of July, and returned to low concentrations at the beginning of August, when a second peak of tintinnids occurred (Fig. 1e). From the end of the spring bloom, flagellates were numerically dominated by cells <10 µm.

Seasonal dynamics and size distributions of protozoa in 1992

Since chl a biomasses and concentrations of protozoa were generally maximal near the surface during 1991 (Fig. 1), only protozoa from the 5 m samples were enumerated in 1992. Within the flagellate assemblage, cells <20 µm (primarily cells 5 to 15 µm in length) were always numerically dominant and accounted for 50 (May) to ≥80% (June to September) of the protist assemblage (Fig 2a). They reached their highest concentrations during summer (Fig. 2b), when the bulk of the chl a biomass was <5 µm (Fig. 2e). Concentrations of nanoflagellates displayed a seasonal trend relatively similar to that of cyanobacteria (Fig. 2b) but not of chl a <5 µm (Fig. 2d). In 1992, the abundances of nanoflagellates varied throughout the summer, with high concentrations being observed 3 times. The 3 peaks closely followed or corresponded to peak concentrations of cyanobacteria (Fig. 2b). Contribution of flagellates >20 µm to the protist assemblage was always <30%

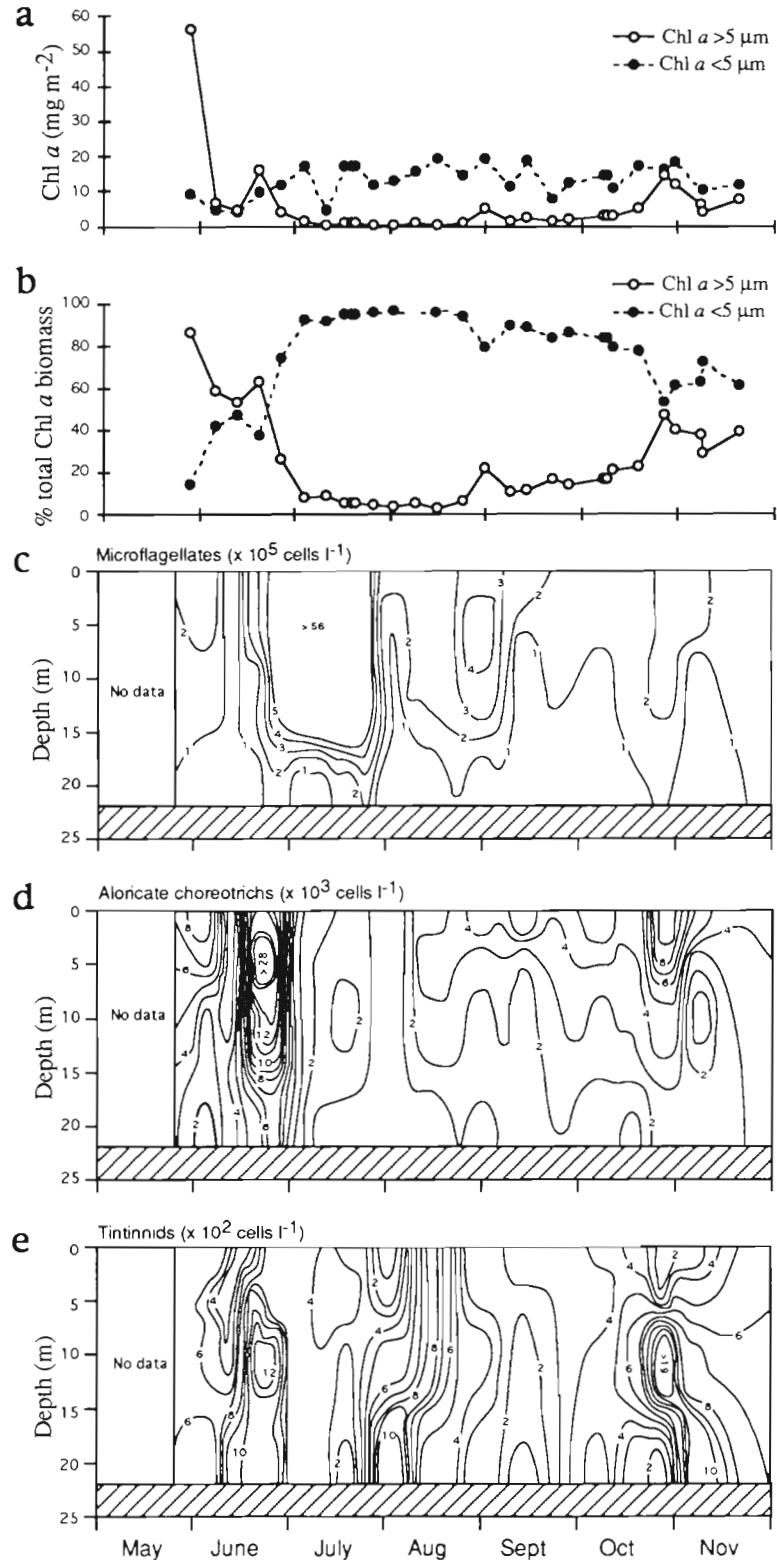Fig. 1. Seasonal changes in 1991 in (a) chl a concentrations of phytoplankton >5 and <5 µm, (b) relative contributions (%) of the 2 size fractions to the total biomass of phytoplankton (values were integrated over the 22 m water column), and depths of isopleths for the concentrations of (c) microflagellates, (d) aloricate choreotrichs, and (e) tintinnids
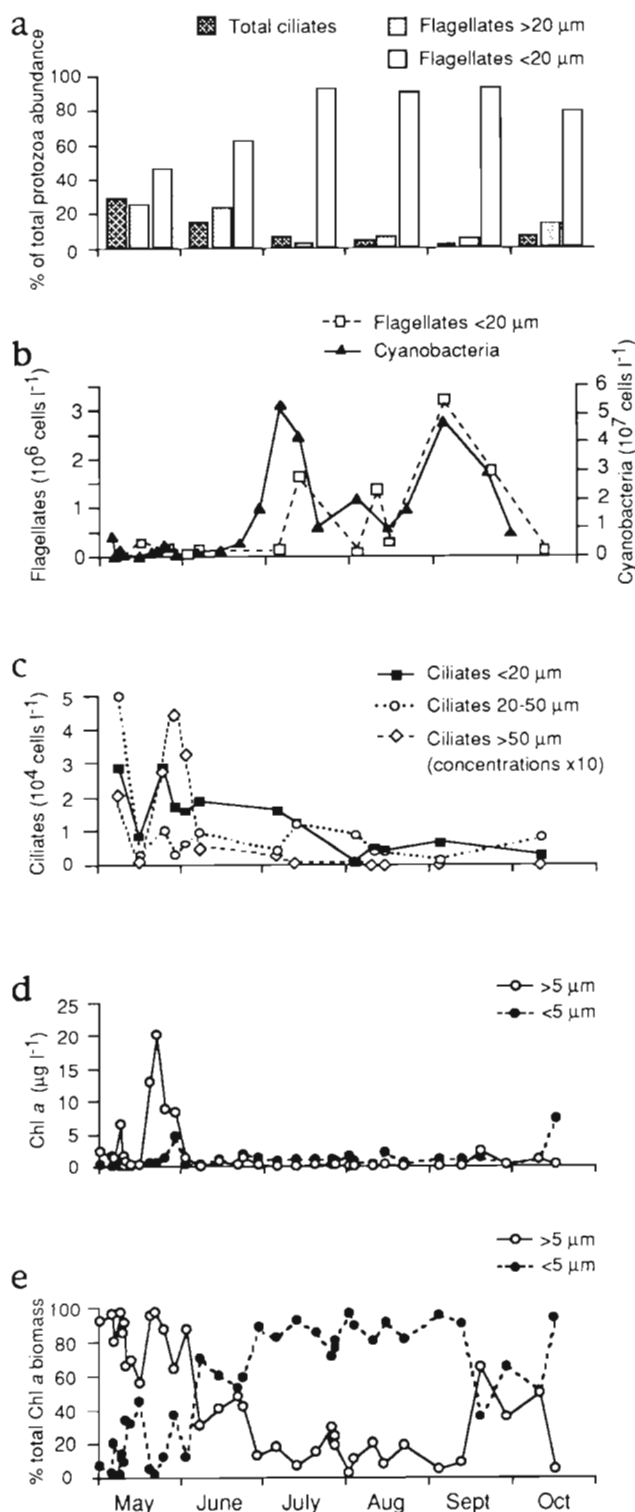
Fig. 2. Seasonal changes at 5 m depth in 1992 in (a) relative contributions (%) of ciliates and flagellates to total protist abundance, (b) concentrations of flagellates <20 μm and cyanobacteria, (c) concentrations of ciliates in 3 size fractions, (d) concentrations of chl *a* in fractions >5 and <5 μm, and (e) relative contributions (%) of the 2 size fractions to the total biomass of phytoplankton

(Fig. 2a). Flagellates <20 μm reached their highest concentrations during May and June, with values >4.0 × 10^4 cells l^{-1}.

Within the ciliate assemblage, concentrations of cells tended to decrease from small (<20 μm) to large (>50 μm) organisms. The overall abundance of ciliates was highest during May and at the beginning of June (Fig. 2c), i.e. the period when the phytoplankton biomass was dominated by the >5 μm fraction (Fig. 2e). Following the spring diatom bloom, the abundance of ciliates remained at low level until October. Ciliates >50 μm were only observed from May through July and their concentrations were relatively low (Fig. 2c). During the spring, ciliates >50 μm accounted for up to 17% of the ciliate assemblage whereas, after June, their contribution was generally <1%. Ciliates >50 μm were primarily *Laboea strobila*, *Helicostomella* sp., and a large (60 to 80 μm diam.) *Strobilidium* sp. With the exception of high values during the bloom of chain forming diatoms, in May, abundances of ciliates in the 20–50 μm size range remained at ca 5 × 10^3 cells l^{-1} during the whole sampling season (Fig. 2c), their contribution to the ciliate assemblage ranging from 29 to 59%. As was the case for the other size fractions, the abundance of ciliates <20 μm was higher during spring than summer and, on the whole, they numerically dominated the ciliate assemblage.

Carbon biomass of protozoa in 1992

The total carbon biomass of protozoa reached its highest values during spring (>500 μg C l^{-1}), after which there was a decrease to low values (<100 μg C l^{-1}) during summer and autumn (Fig 3a). During spring and at the beginning of summer, ciliates dominated the biomass of the protozoan assemblage; whereas, at the end of summer and in autumn, flagellates were the major contributors.

The biomass of the total ciliate assemblage (Fig. 3a) was highest during spring (>400 μg C l^{-1}) and remained at lower values during summer and autumn (<100 μg C l^{-1}), this decline being observed for cells both <20 and >20 μm (not shown). In contrast to cell numbers, where the major contribution was that of ciliates <20 μm (Fig. 2c), ciliates >20 μm always dominated the biomass of the ciliate assemblage (Fig 3c). Ciliates 50 to 200 μm were dominant during May and June, whereas cells 20–50 μm dominated the ciliate biomass from July through October

Cells >20 μm (Fig. 3b) generally dominated the carbon biomass of flagellates, their proportion decreasing from May through October. In contrast, the contribution of flagellates <20 μm to the total flagellate biomass increased during summer, reaching 46 μg C l^{-1} in Sep-
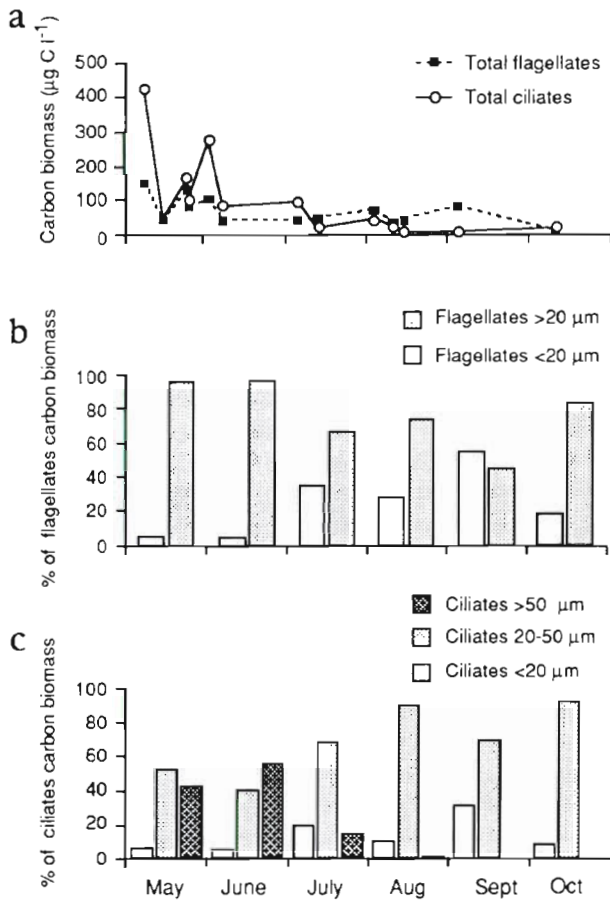
Fig. 3. Seasonal changes at 5 m depth in 1992 in (a) carbon biomass of the ciliate and flagellate assemblages, (b) relative contributions (%) of flagellates >20 and <20 μm to the flagellate biomass, and (c) relative contributions (%) of ciliates >20, 20–50 and <20 μm to the ciliate biomass

Fig. 4. Seasonal changes in carbon biomasses of heterotrophic dinoflagellates >20 μm at 5 m depth in 1992

tember. Within the flagellates >20 μm, there were some large naked and thecate dinoflagellates belonging to heterotrophic or mixotrophic genera. These represented high carbon biomasses, with standing stocks up to 270 μg C l^{-1} during the spring diatom bloom in May (Fig. 4). At this time of the year, *Gyrodinium spirale* and *Gymnodinium* spp. were the most abundant dinoflagellates, with concentrations up to 1×10^4 and 3.5×10^4 cells l^{-1}, respectively. From June through October, the concentrations and carbon biomasses of heterotrophic dinoflagellates >20 μm steadily decreased to low values (Fig. 4).

Trophic relationships between protists and phytoplankton size fractions

In 1991, there were significant positive linear correlations between the abundances of both aloricate choreotrichs and tintinnids and chl *a* in the >5 μm frac-

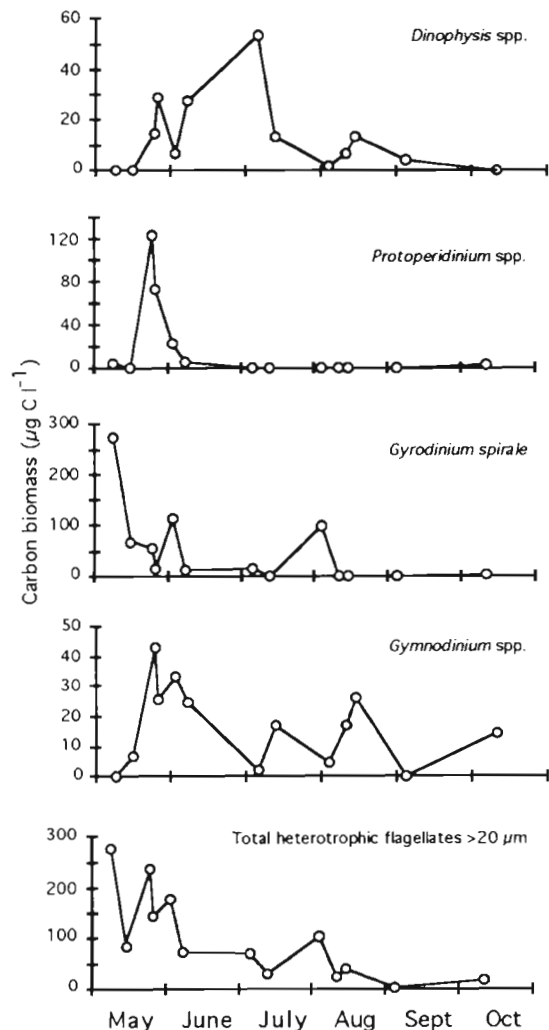tion (Table 1a). Correlations between microflagellate abundances and size-fractionated chl *a* were not statistically significant. Using data from 1992, correlations were computed between the abundance and biomass of each size fraction of ciliates and flagellates and the chl *a* in 2 size fractions (Table 1b). Concentrations and biomasses of ciliates (except those >50 μm) were significantly correlated with chl *a* in the >5 μm fraction, but not with that in the <5 μm fraction. Biomass of flagellates >20 μm was also significantly correlated with chl *a* in the >5 μm fraction. Finally, the concentrations and biomasses of protists were not significantly correlated with the biomasses of single celled cyanobacteria (Table 1b).

Path analysis was used to further elucidate seasonal changes in the interactions among temperature, protist abundances, and the concentrations of their potential food particles. Fig. 5a shows the model of maximum

Table 1 (a) Linear correlation coefficients between abundances of ciliates (aloricate choreotrichs and tintinnids) and flagellates and chl a biomass in 2 size fractions in 1991 (n = 15, values integrated over the 22 m water column). (b) Correlations betweeen the cell concentrations and carbon biomass of ciliates and flagellates and the size-fractionated chl a and carbon biomass of cyanobacteria in 1992 (n = 13). $^*p < 0.05$; ns: not significant ($p > 0.05$); nm: not measured

| | | Size-fractionated chl a | | Cyanobacteria |
		<5 µm	>5 µm	carbon biomass
(a) 1991				
Aloricate choreotrichs		−0.30 ns	0.71 *	nm
Tintinnids		−0.52 ns	0.59 *	nm
Flagellates		0.28 ns	−0.23 ns	nm
(b) 1992				
Ciliates <20 µm	Concentration	−0.15 ns	0.80 *	−0.24 ns
	Carbon biomass	−0.19 ns	0.71 *	−0.26 ns
Ciliates 20–50 µm	Concentration	−0.33 ns	0.59 *	−0.27 ns
	Carbon biomass	−0.47 ns	0.69 *	−0.25 ns
Ciliates >50 µm	Concentration	0.14 ns	0.49 ns	−0.47 ns
	Carbon biomass	−0.33 ns	0.53 ns	−0.43 ns
Flagellates <20 µm	Concentration	0.07 ns	−0.34 ns	−0.09 ns
	Carbon biomass	0.11 ns	−0.31 ns	−0.10 ns
Flagellates >20 µm	Concentration	0.22 ns	0.22 ns	0.06 ns
	Carbon biomass	−0.13 ns	0.80 *	−0.45 ns

hypothesised causal interrelationships among variables. Separate calculations were done for the spring phytoplankton bloom (28 May to 24 June), summer (1 July to 20 August) and late summer-autumn (27 August to 15 November), using the large 1991 data set. Only statistically significant path coefficients (p <0.05) are shown in Fig. 5b–d, where the percentage of noncausal relationships (spurious correlations) are also given.

Results from path analysis were different for the 3 seasons (Fig. 5b–d). Overall, relationships between protists and phytoplankton were stronger and more complex during the spring diatom bloom than during summer and autumn. In spring (Fig. 5b), the abundance of microflagellates was strongly and positively related to the concentration of chl a <5 µm (path coefficient = 0.74), whereas it was negatively related to chl a >5 µm (−0.35). During spring, the highest path coefficient (1.02) was from chl a <5 µm to aloricate ciliates and the abundance of tintinnids was positively related to chl a >5 µm (0.88). In summer (Fig. 5c), there was a positive path from chl a <5 µm to aloricate ciliates (0.82) and to flagellates (0.93). In late summer-autumn (Fig. 5d), there was a positive path from chl a <5 µm to flagellates (0.57) and aloricate ciliates (0.79). However, 67 % of the path from chl a <5 µm to aloricate ciliates was non causal (spurious) There was no significant path from chl a >5 µm to protists.

The paths from temperature to protists were never significant during the sampled periods (Fig 5b–d). In contrast, there were strong and positive paths from temperature to chl a <5 µm during spring (0.86), sum-

mer (0.70), and late summer-autumn (0.50). In summer, there was a significant path from temperature to chl a >5 µm (0.71).

Grazing experiments in 1992

Initial conditions for the grazing experiments are given in Table 2. Assemblages of protozooplankton <64 µm in the incubation bottles were always numerically dominated by flagellates <20 µm and, with 2 exceptions in May, ciliates >50 µm were virtually absent from the bottles during summer. Generally, the abundance of ciliates <20 µm was higher than or similar to that of ciliates 20–50 µm. Initial chl a concentrations in control bottles filled with the particle free seawater were <0.05 µg l^{-1} and no increase occurred during the course of incubations.

Results from the 06 May, 23 May and 11 August incubations cannot be used, because the slopes (g) of the regressions were not significantly different from zero (Table 3). For the remaining dates, growth rates of phytoplankton estimated from grazing experiments were high, i.e. values decreased from 1.58 doublings d^{-1} at the end of spring to 0.61 at the end of summer. During the incubations, protozoan grazers removed between 29 and 42 % of the initial phytoplankton standing stock per day. Between 54 and 125 % of phytoplankton potential production was grazed daily (Table 3). The grazing coefficients were relatively homogeneous throughout the sampling season, with values ca 0.5 d^{-1}.
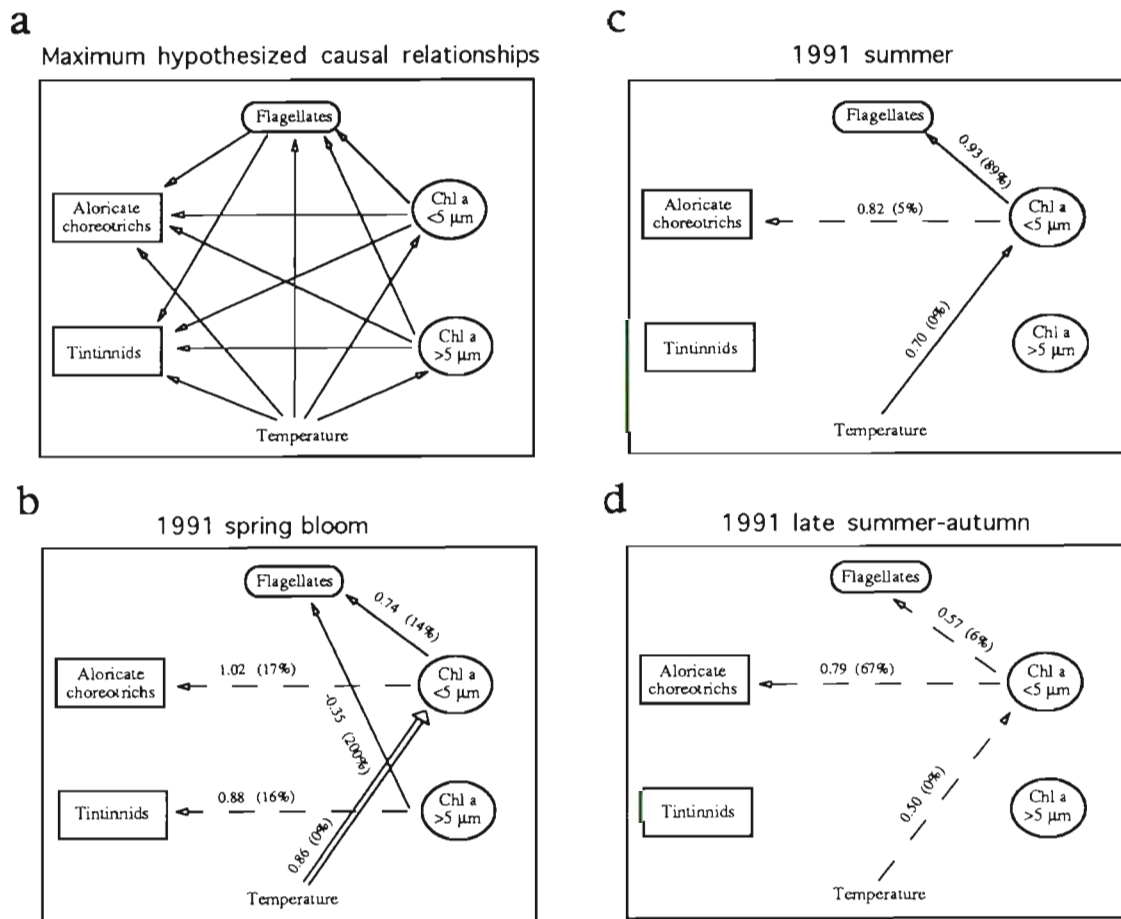
Fig. 5. Schematic representation of path analyses showing (a) the model of maximum hypothesized causal relationships among temperature, protists, and phytoplankton and the path coefficients computed using 1991 data from the (b) spring bloom, (c) summer, and (d) late summer-autumn. ($\Longrightarrow$) $p < 0.001$; ($\longrightarrow$) $0.001 < p < 0.01$; ($-- \bullet$) $0.01 < p < 0.05$. Only significant ($p < 0.05$) path coefficients are shown. The intensity of the path and the percentage of non-causal relationship (spurious correlation) are given

Since flagellates and ciliates <20 µm are considered to be bacterivorous, and therefore should not significantly graze phytoplankton, the average clearance rates of protists were calculated by dividing the daily loss of phytoplankton biomass in the incubation bottle by the corresponding abundance of protists >20 µm. Clearance rates estimated for protists >20 µm were between 0.4 and 8 µl ind.$^{-1}$ h^{-1}, so that, on average, the phytoplankton carbon grazed by these protists was 11 µg C l^{-1} d^{-1} (Table 4). Phytoplankton growth rates

Table 2. Conditions in undiluted controls at the start of grazing experiments and ambient seawater temperature recorded at 5 m depth before the incubations started

Date (1992)	Ambient temperature (°C)	Initial algal biomass (µg chl a l^{-1})	Initial ciliate biomass (µg C l^{-1})	Choreotrich ciliates			Flagellates	
				<20 µm (cells l^{-1})	20–50 µm (cells l^{-1})	>50 µm (cells l^{-1})	<20 µm (cells l^{-1})	>20 µm (cells l^{-1})
6 May	1.5	2.85	130	2.6×10^4	1.7×10^4	1.4×10^3	2.2×10^5	2.8×10^3
23 May	6.5	2.76	112	2.8×10^4	1.0×10^4	2.4×10^3	1.6×10^5	4.9×10^4
31 May	7.8	0.45	263	1.6×10^4	5.6×10^3	$<10^2$	7.3×10^4	3.4×10^4
29 Jul	14.7	2.05	17	5.2×10^3	2.0×10^3	$<10^2$	4.7×10^5	3.8×10^3
7 Aug	12.3	0.35	24	5.4×10^3	3.7×10^3	$<10^2$	1.4×10^6	1.8×10^4
11 Aug	12.6	1.62	16	4.5×10^3	4.2×10^3	$<10^2$	3.0×10^5	2.5×10^4
13 Aug	11.0	0.53	8	2.1×10^3	2.1×10^3	$<10^2$	1.6×10^5	$<10^2$

Table 3. Grazing of phytoplankton chl a by protozooplankton: results of dilution experiments. k: phytoplankton growth coefficient; g: grazing coefficient; phytoplankton doublings = k /ln 2; potential phytoplankton production = $(P_0 e^k) - P_0$; actual phytoplankton production = $(P_0 e^{(k-g)}) - P_0$; % initial chl a removed = (potential production – actual production)/P_0; % potential production removed = (potential production – actual production)/potential production; $^*p < 0.05$; ns: not significant (p > 0.05)

Date (1992)	k (d^{-1})	Phytoplankton doublings (d^{-1})	% initial chl a produced (d^{-1})	g (d^{-1})	% initial chl a removed (d^{-1})	% potential phytoplankton production removed (d^{-1})
6 May	ns	ns	ns	ns	ns	ns
23 May	ns	ns	ns	ns	ns	ns
31 May	1.09*	1.58	198	0.54*	42	63
29 Jul	0.77*	1.11	115	0.34*	29	54
7 Aug	0.65*	0.94	92	0.40*	33	69
11 Aug	ns	ns	ns	ns	ns	ns
13 Aug	0.41*	0.61	51	0.55*	42	125

Table 4. Estimated clearance rate of protists >20 µm and phytoplankton carbon grazed daily. Estimated hourly clearance rate = (incubated water cleared d^{-1})/(24 h × protist concentration); incubated water cleared d^{-1} = % P_0 grazed d^{-1}; phytoplankton carbon grazed = % P_0 grazed d^{-1} × P_0 × 40 µg C^{-1} µg chl a

Date (1992)	Clearance rate (µl ind^{-1} h^{-1})	Carbon grazed (µg C d^{-1})
31 May	0.41	8
29 Jul	2.07	24
7 Aug	0.63	5
13 Aug	7.88	9

computed using results from the ^{14}C primary production experiments were between 0.4 and 1.5 doublings d^{-1}, whereas growth rates estimated from the results of dilution experiments were between 0.9 and 1.6 doublings d^{-1}.

DISCUSSION

Methodological considerations

Before discussing the results, potential bias of the serial dilution method must be addressed. For example, nutrient recycling may be low in the most dilute treatments, so that nutrient limitation may affect phytoplankton growth. In the present study, macronutrient concentrations in all incubation bottles were always above detection at the end of the incubation. Furthermore, the fact that growth rates of phytoplankton in the incubation experiments (Table 3) were relatively high and typical of temperate waters (see Goldman et al. 1979 for comparison) indicates that no nutrient limitation occurred during the course of the incubations.

Changes in assemblages may occur at the prefiltration step (Lynn & Montagnes 1991). The effect of reverse filtration with a 64 µm mesh on the ciliate assemblage was examined and no significant decrease or change was found, i.e. ciliate concentrations in the undiluted bottles were always within the range of concentrations observed during the seasonal survey (Figs. 1 & 2). Finally, since phytoplankton never reached high biomasses in the experiments, the plots of apparent growth rate versus fraction of undiluted seawater never exhibited non-linearity, which would have been indicative of saturated feeding responses.

Protozooplankton abundance and biomass

Concentrations of protozooplankton in the present study (Figs. 1 & 2) are similar to those generally reported for temperate nearshore and estuarine waters. Concentrations of flagellates as high as 10^6 cells l^{-1} are not exceptional in nearshore areas and in the open ocean (Painting et al. 1992, Vaqué et al. 1992). Values of 10^3 to 10^4 cells l^{-1} for aloricate choreotrichs and $<10^2$ to 10^3 cells l^{-1} for tintinnids are generally observed in nearshore waters (Dolan & Coats 1990, Pierce & Turner 1992, Buskey 1993).

In contrast to cell concentrations, estimates of carbon biomasses for protozooplankton (Figs. 3 & 4) are quite high when compared with values from the literature. Biomasses of heterotrophic flagellates generally range between <0.1 and 50 µg C l^{-1} (Lessard 1991). However, biomasses of heterotrophic flagellates >100 µg C l^{-1} have sometimes been reported in temperate nearshore waters during diatom blooms (Hansen 1991, Bralewska & Witek 1995). Flagellate biomasses >100 µg C l^{-1} observed in May (Figs. 3 & 4) are therefore among the highest reported and can be attributed to the high abundances of large heterotrophic dinoflagellates like *Gyrodinium spirale* (Fig. 4), whereas the values from

summer and autumn are less unusual. During summer and autumn, carbon biomasses of the ciliate assemblage were similar to those in other nearshore and estuarine systems. For example, ciliate biomasses of 8 to 16 µg C l^{-1} were reported in Chesapeake Bay, USA (Dolan & Coats 1990), 1 to 56 µg C l^{-1} in Kiel Bight, Germany (Smetacek 1981), and 0.2 to 52 µg C l^{-1} in the St. Lawrence Estuary (Sime-Ngando et al. 1995). Values >150 µg C l^{-1} observed during May and June are unusual and related to the high concentrations of large ciliates such as tintinnids.

Size structure of phytoplankton and protozooplankton and controlling factors

The concentration and biomass of protozooplankton generally followed the seasonal dynamics of phytoplankton. Peaks in protozooplankton abundance and biomass precisely matched the spring and autumn blooms of phytoplankton >5 µm, whereas abundances and biomasses of protozoa decreased during summer, when phytoplankton were dominated by the <5 µm fraction (Figs. 1 to 4). The high correlation coefficients between the biomasses of protozooplankton and phytoplankton >5 µm (Table 1) may be an indication that the 2 variables responded similarly to the same environmental factor (e.g. temperature), or that dynamics of protozooplankton, on the whole, responded to the abundance of large phytoplankton, or that the 2 were controlled by mesozooplankton grazing. These 3 alternatives are, in turn, discussed here.

(1) The possibility that correlations between the 2 groups reflected the influence of temperature was tested using partial correlations. When controlling for the effect of temperature in 1991, the abundances of aloricate choreotrichs and tintinnids remained significantly correlated to chl a >5 µm ($r = 0.82$, $p < 0.01$ and $r = 0.63$, $p < 0.05$, respectively). In 1992, the only partial correlation significantly different from zero was that between ciliates <20 µm and chl a >5 µm ($r = 0.74$, $p < 0.01$). Accordingly, path analysis showed no significant causal relationship between temperature and protist abundance or chl a >5 µm in 1991 (Fig. 5). Thus, temperature changes alone cannot completely explain the parallel variations of large phytoplankton and protozooplankton, which were observed during the 2 sampling seasons.

(2) In nearshore waters of the Kattegat, heterotrophic dinoflagellates >20 µm are abundant during periods dominated by large phytoplankton, upon which they feed (e.g. spring diatom bloom and autumn bloom of autotrophic dinoflagellates), whereas smaller forms are abundant during periods dominated by nanophytoplankton (Hansen 1991). Similarly, in the Baie des

Chaleurs, heterotrophic dinoflagellates, like *Dinophysis* spp., *Protoperidinium* spp., *Gyrodinium* spp., and *Gymnodinium* spp., which are grazers of large diatoms (Jacobson & Anderson 1986, Hansen 1991, Neuer & Cowles 1994), were particularly abundant during the spring diatom bloom (Fig. 4). Given that feeding rates of dinoflagellates are comparable to those of ciliates (Capriulo 1991), large flagellates can sometimes be major grazers of diatom blooms (Lessard 1991). Consequently, high concentrations and biomasses of large herbivorous flagellates in the bay during spring may have been related to the high biomasses of their food items, i.e. diatoms. This is supported by the strong correlation between the carbon biomass of flagellates >20 µm and chl a >5 µm in 1992 (Table 1). However, since large flagellates and the dominating nanoflagellates were not enumerated separately in 1991, path analysis did not show any significant link between large phytoplankton and large flagellates during the spring bloom (Fig. 5b). These results stress the potential importance of large flagellates in the fate of the spring phytoplankton bloom in the nearshore waters of the Baie des Chaleurs.

A trophic relationship between ciliates and phytoplankton >5 µm (Table 1, Fig. 5b), the latter being primarily chain-forming diatoms of the spring bloom, would be more unusual, since laboratory experiments have shown that naked oligotrichous ciliates and tintinnids feed almost exclusively on nanoplankton (Rassoulzadegan et al. 1988, Bernard & Rassoulzadegan 1990). However, in the Baie des Chaleurs, ciliates did respond numerically to the spring phytoplankton bloom in both 1991 and 1992 (Figs. 1 & 2). The sizes of individual cells of the chain-forming bloom diatoms (primarily *Chaetoceros* spp. and *Thalassiosira* spp.) were <30 µm, i.e. in the size range potentially available to ciliates. Results from a study in the northwest Atlantic support the view that ciliates can efficiently feed on chain-forming diatoms and control their growth (Paranjape 1990). In addition, in the St. Lawrence Estuary, microscopic observations showed ciliates with up to 10 ingested cells of bloom forming diatoms such as *Skeletonema* spp., *Chaetoceros* spp. and *Thalassiosira* spp. (Sime-Ngando et al. 1995). Given that high abundance and biomass of ciliates were observed during the spring and the ciliates and diatoms were similar to those observed by Sime-Ngando et al. (1995), it is concluded that herbivorous ciliates likely played an important role as consumers of the spring diatom bloom in the nearshore waters of the Baie des Chaleurs.

The carbon standing stock of heterotrophic ciliates varied from 10 to 530% of that of phytoplankton (these values can be derived from Figs. 2d & 3a). Values well above 100% during spring and at the begin-

ning of summer could only be supported by rapid phytoplankton growth. Maximum doubling rates of phytoplankton during spring were >1 d^{-1} (Table 3), so that the high production of phytoplankton could support the high biomass of the ciliate assemblage dominated by large forms. During summer, the standing stock of heterotrophic ciliates was $<100\%$ of the phytoplankton standing stock. In addition, nanoflagellates and ultraplanktonic eucaryotes were abundant, and their sizes (3 to 20 µm) were within the optimal range for tintinnids and aloricate ciliates >50 µm (Rassoulzadegan et al. 1988). Therefore, the relative disappearance of tintinnids and aloricate ciliates >50 µm during summer was probably not the result of food limitation. A recent study by Wiackowski et al. (1994) showed that, when total ciliate abundance is depressed by zooplankton, the relative frequency of larger ciliates decreases. Selective feeding by mesozooplankton on large heterotrophic flagellates has also been reported in a study by Weisse (1991). Thus, increased grazing by mesozooplankton on protozoa may have accounted for the shift from larger to smaller forms of ciliates and flagellates.

(3) A caveat to the above discussion on factors that possibly controlled the biomass and size structure of protozooplankton is the absence, in the present study, of data on the grazing of mesozooplankton on the protozoan assemblage. In 1991 and 1992, ratios of Phaeo/ (chl a + Phaeo) in the >20 µm fraction (not shown), which can be considered a rough index of faecal pellet concentrations and mesozooplankton grazing (Welschmeyer et al. 1984, Bustillos-Guzman et al. 1996), provide indication that production of faecal pellets by mesozooplankton was higher during summer than spring. Seasonal changes in these ratios are consistent with published data on seasonal variations of mesozooplankton abundance in the Baie des Chaleurs (Fig. 6) (Lacroix & Filteau 1970, 1971). Moreover, Y. Lagadeuc, M. Boulé & J. Dodson (unpubl.) report that, at a nearshore station close to our sampling site, copepods reached particularly high concentrations in August and September 1991, with adults and nauplii often exceeding 2×10^4 ind. m^{-3} and *Oithona similis* and *Pseudocalanus* sp. dominating the copepod assemblage. These results are in agreement with qualitative samples taken at our station in July and August 1992, which showed that *Oithona similis* accounted for $44 \pm 21\%$ of adult copepods and $53 \pm 22\%$ of copepodites (not shown). Oithonidae are considered to be omnivorous, with a preference for motile food such as dinoflagellates (Paffenhöfer 1993)

Since (1) in temperate nearshore water, a greater percentage of the carbon ingested by copepods is generally supplied by ciliates when phytoplankton stocks are low or dominated by small cells (Fessenden & Cowles 1994), as was the case in the present study from June until August (Figs. 1a & 2d), (2) there are indications that concentrations and grazing activities of copepods reached their highest values during summer, and (3) small omnivorous Oithonidae dominated the copepod assemblage, we hypothesise that copepods exerted stronger grazing pressure on protozooplankton during summer than spring. The same was observed in the adjacent Gulf of St. Lawrence, where heterotrophic ciliates and dinoflagellates make up a high fraction of the diet of *Calanus finmarchicus* during summer, despite the low concentrations of these protists (Ohman & Runge 1994). Similar changes in the seasonal dynamics of copepods were also proposed by Hansen (1991) to explain the peak of large heterotrophic dinoflagellates during the spring phytoplankton bloom and their absence during a late summer bloom, in nearshore waters of the Kattegat.

Grazing effect of protozooplankton on phytoplankton

Concentrations of single celled cyanobacteria in nearshore waters of the Baie des Chaleurs were high during summer (Fig. 2b). It is generally considered that nanoflagellates (Campbell & Carpenter 1986) and ciliates <20 µm (Rassoulzadegan et al. 1988) are the main grazers of these autotrophic procaryotes. However, since the carbon biomass of cyanobacteria is low compared to that of autotrophic eucaryotes (Li 1995, Tamigneaux et al. 1995) and the efficiency of chl a extraction from cyanobacteria is low (Joint & Pomeroy 1986), changes of chl a concentration in dilution experiments most probably reflect changes in the abundance of autotrophic nanoflagellates and dinofla-
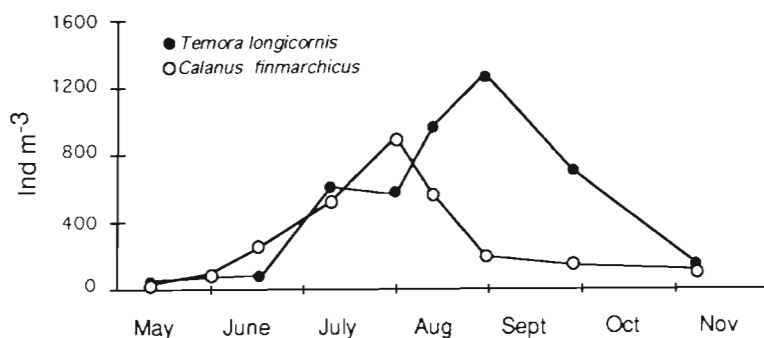

Fig. 6. Seasonal changes in the concentrations of 2 frequent and abundant copepod species in the 0 to 40 m layer of the Baie des Chaleurs (redrawn after Lacroix & Filteau 1970, 1971)

gellates and of diatoms instead of changes in the abundance of cyanobacteria. Consequently, grazing coefficients calculated from dilution experiments are considered to mainly reflect grazing by herbivorous ciliates and flagellates >20 µm on autotrophic eucaryotes. Clearance rates estimated for the assemblage of protozooplankton >20 µm were between 0.4 and 8 µl ind.$^{-1}$ h^{-1} (Table 4). These estimates are at the lower end of values generally reported for tintinnids (up to 85 µl ind.$^{-1}$ h^{-1}; Capriulo 1991), but within the range of clearance rates reported for ciliates <50 µm, 0.36 to 6.38 µl ind.$^{-1}$ h^{-1} (Bernard & Rassoulzadegan 1990), for aloricate ciliates, 0 to 12 µl ind.$^{-1}$ h^{-1} (Rublee & Gallegos 1989), and for dinoflagellates, 0.5 to 28 µl ind.$^{-1}$ h^{-1} (Lessard & Swift 1985). This supports the idea that aloricate ciliates and flagellates >20 µm, which were abundant in our experiments, played an important role as consumers of phytoplankton in nearshore waters.

There are few reports of grazing experiments that concern protozooplankton only, since most studies also included microzooplankton. In such experiments, small metazoa are allowed to compete with and/or prey on protozoan grazers. Our results are in agreement with those of a comparable study by Gifford (1988) in nearshore waters off Nova Scotia, for which water had been filtered through a 100 µm mesh to remove copepod nauplii. It was found that 21 to 50 % of the initial standing stock and 40 to 100 % of the potential production were grazed daily by protozoa (in the Baie des Chaleurs, corresponding values were 29 to 42 % and 54 to 125 %, respectively; Table 3). More generally, grazing rates in the present study were in the upper range of values reported for microzooplankton grazing (Pierce & Turner 1992), indicating that, when predation is reduced, protozooplankton are able to consume all the primary production and even more. Indeed, in spite of relatively high primary production (Table 5), chl a biomass remained low and relatively stable during the summer months (Figs. 1a & 2d). However, since water in experiments was prefiltered

through a 64 µm mesh, chain-forming diatoms may have been fragmented into smaller components, thus making some additional phytoplankton biomass available to ciliates. Grazing rates from May were therefore possibly overestimated. Keeping in mind these caveats, grazing by herbivorous ciliates and flagellates could account for the low and steady chl a biomass observed during summer.

Estimated effect of grazing on cyanobacteria

Maximum numbers of nanoflagellates were observed from the end of the spring bloom to the end of summer (Figs. 1 & 2) as is generally the case in temperate estuaries (Coffin & Sharp 1987, Dolan & Coats 1990). In contrast to nanoflagellates, ciliates <20 µm were more abundant from May to July than at the end of summer (Fig. 2c). However, because of the low contribution of large forms during summer, the relative contribution of the <20 µm fraction to the carbon biomass of ciliates was highest from July to September (Fig 3c). Nanoflagellates and ciliates <20 µm are considered to be consumers of ultraplanktonic and bacteria-sized particles (Reid et al. 1991), and, indeed, maximum concentrations of autotrophic ultraplankton <5 µm (i.e. cyanobacteria and eucaryotes) were observed from July through September (Figs. 1a & 2b, d). That the biomass of phytoplankton <5 µm remained very constant during summer suggests a close coupling with their consumers.

To estimate the potential grazing of nanoflagellates on cyanobacteria, a clearance rate of 15 nl ind.$^{-1}$ h^{-1} was assumed for nanoflagellates grazing on these bacteria-sized algae (Caron et al. 1991). Because we could not reliably distinguish autotrophic from heterotrophic/mixotrophic protists, we made the conservative assumption that only 50 % of the nanoflagellates were potential grazers of cyanobacteria. This is highly conservative since, in the Gulf of St. Lawrence, epifluorescence microscopy showed that mixotrophs may sometimes account for 94 % of the flagellate assemblage, the remainder being heterotrophs (C. Lovejoy pers. comm.). Using the carbon biomass of cyanobacteria, it was calculated that flagellates <20 µm could consume 2 to 24 % (mean = 6 %) of the standing stock of cyanobacteria. These results lead to the conclusion that the effect of nanoflagellates on cyanobacteria was probably weak. However, recent studies indicate that, in nearshore waters, ingestion rates of bacteria-sized food items by pigmented nanoflagellates are similar to those of

Table 5. Comparison of phytoplankton growth rates estimated from the results of *in situ* grazing experiments and from *in situ* incubations using the ^{14}C primary production method. ^{14}C doublings per day = primary production/(chl a × 30 µg C µg chl a^{-1}). ns: not significant (p > 0.05)

Date (1992)	Phytoplankton chl a biomass (µg chl a l^{-1})	^{14}C primary production (µg C l^{-1} d^{-1})	^{14}C doublings d^{-1}	Grazing expt doublings d^{-1}
6 May	2.85	37	0.43	ns
23 May	2.80	116	1.38	ns
31 May	1.53	52	1.13	1.58
29 Jul	1.78	23	0.4	1.11
7 Aug	0.40	18	1.5	0.94

nanoflagellates that lack chloroplasts (Epstein & Shiaris 1992, Hall et al. 1993). In the present study, if all nanoflagellates were considered to be bacterivorous, they could play an important role in regulating cyanobacterial numbers but they would probably not fully control them.

Small ciliates are the next candidates as grazers of cyanobacteria, since it has been reported that picoplankton may constitute up to 72% of the diet of ciliates <30 µm (Rassoulzadegan et al. 1988) and individual clearance rates of ciliates are higher than those of nanoflagellates. In addition, small oligotrichs may have significantly higher clearance rates for cyanobacteria compared to those for bacteria, these ciliates meeting all of their carbon requirements by feeding exclusively on picoplankton (Simek et al. 1996). Assuming a clearance rate of 0.5 µl ind.$^{-1}$ h^{-1} for ciliates <20 µm grazing on cyanobacteria (Bernard & Rassoulzadegan 1990), it is estimated that these small ciliates could have consumed an average 0.41 µg C l^{-1} d^{-1} (0.01 to 1.61 µg C l^{-1} d^{-1}), which corresponds to 17% (1 to 35%) of the cyanobacteria standing stock. It therefore appears that the potential grazing effect of ciliates <20 µm on cyanobacteria is at least equivalent to that of nanoflagellates (if 100% of the nanoflagellates were grazers) or even higher if it is assumed that 50% of the nanoflagellates are grazers.

Spring and summer food webs in temperate nearshore waters

Data in the present study support the view that, in temperate nearshore waters, there is a close link between seasonal changes in the size structures of phytoplankton and protozooplankton assemblages. Our observations and the results of path analyses combined with findings from the literature suggest the existence of 2 different food webs corresponding to bloom and summer conditions, respectively (Fig. 7). These 2 webs are based on the sizes of autotrophic cells and of herbivorous protozoa, on the changes in mesozooplankton grazing pressure and, hence, on resulting changes in trophic pathways mediated by protozooplankton. This model is somewhat similar to that of Rivkin et al. (1996), who documented, in sedimenting particulate matter of the Gulf of St. Lawrence, replacement of phytoplankton material (spring bloom) by an equivalent amount of faecal pellets (summer).

During the spring bloom (Fig. 7a), the phytoplankton assemblage mainly consisted of chain-forming diatoms (>5 µm). Because of their low abundances at this time, mesozooplankton did not exert significant grazing pressure on phytoplankton or protozooplankton and production of faecal pellets was low. Given the weak control by mesozooplankton, large-sized herbivorous
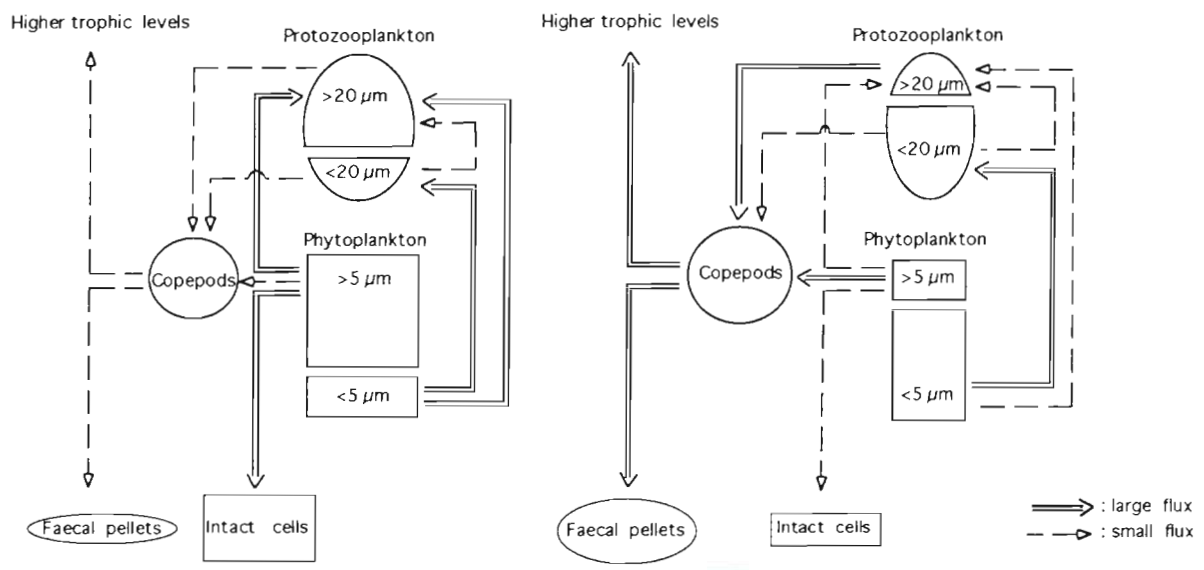
Fig. 7. Conceptual model showing trophic interactions among mesozooplankton (copepods), protozooplankton (<20 and >20 µm), and phytoplankton (<5 and >5 µm), for 2 typical seasonal situations in temperate nearshore waters: (a) during the spring phytoplankton bloom, biomasses of large phytoplankton and large protozooplankton are high and concentrations of copepods are low. (b) During summer, phytoplankton and protozooplankton assemblages are dominated by small cells and copepod concentrations reach their seasonal maximum. Areas of geometric figures reflect concentrations and widths of arrows refer to absolute fluxes

protists (>20 µm) feeding on spring bloom phytoplankton developed very high biomasses. However, protozoan grazers did not succeed in controlling the phytoplankton growth and a significant part of the algal biomass sedimented to the bottom at the end of the bloom. During the decline of the bloom, growth of phytoplankton <5 µm was enhanced by the warming of the water (Fig. 5b), these small algae were then exploited (but not controlled) by <20 µm ciliates and flagellates. This view is supported by the results of path analysis, which showed a strong trophic path from phytoplankton >5 µm to large tintinnids and from phytoplankton <5 µm to small aloricate ciliates and flagellates (Fig. 5b). Thus, only a small part of the primary production was channelled towards higher trophic levels of the pelagic food web, directly to copepods or through protozooplankton.

During summer (Fig. 7b), in contrast with the spring diatom bloom, the phytoplankton assemblage was dominated by ultraplankton (<5 µm). Their biomass slightly increased at the end of the bloom, after which it remained relatively constant. At the same time, the abundance of omnivorous copepods and production of faecal pellets were highest. High grazing pressure by omnivorous copepods maintained the biomass of protozooplankton at lower levels than during the spring bloom. The weak correlation coefficients (Table 1) and the absence of significant paths from large phytoplankton to protists during summer (Fig. 5c, d) are consistent with a control of protist abundance by mesozooplankton. Protozoa, especially ciliates, actively grazed on phytoplankton and kept a close coupling with small primary producers. The relative contributions of small-sized ciliates and flagellates (<20 µm) strongly increased in the protozooplankton assemblage during summer, because of the small size of autotrophic organisms available and of selective grazing of larger protozoa by copepods. During summer, protozoa, especially ciliates, were therefore an important link in transferring primary production to carnivorous mesozooplankton and, thus, to large metazoans (e.g. fish larvae).

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This article was submitted to the editor