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Dynamics in bacterioplankton production in a shallow, temperate lake (Lake Neusiedl, Austria): evidence for dependence on macrophyte production rather than on phytoplankton

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ABSTRACT: The seasonal dynamics in bacterioplankton abundance and production were studied at 3 characteristic stations (open water, large pond within the reed belt and within the reed *Phragmites australis*) in the shallow Lake Neusiedl, Austria, and related to phytoplankton primary production and dissolved organic carbon (DOC). DOC concentrations ranged from about 1 to 2.5 mmol l^{-1} with humic DOC contributing between 40% during the winter and 55% during the summer. Phytoplankton production was highest in the pond within the reed belt, where the attenuation was lowest, reaching 110 mg C m^{-2} d^{-1} during a distinct phytoplankton bloom in August. Bacterial abundance ranged from 2×10^6 cells ml^{-1} during winter to about 10×10^6 cells ml^{-1} during summer. Bacterial production calculated by thymidine (TdR) and leucine (leu) incorporation, respectively, were in good agreement at the stations in the reed belt, but bacterial production based on leu incorporation was significantly lower than bacterial production based on TdR incorporation at the open water station. Based on a bacterial growth yield of 16% determined in an earlier study, bacterioplankton carbon demand was always at least 1 order of magnitude higher than carbon production of phytoplankton, indicating that bacterioplankton metabolism in Lake Neusiedl is heavily dependent on non-phytoplankton sources of DOC. The bacterial carbon demand (ranging from 225 to 870 mg C m^{-2} d^{-1} depending on the sampling site and substrate used) could be matched by the production of the reed *P. australis* amounting to 750 to 4510 mg C m^{-2} d^{-1}. Since there is no major allochthonous organic matter input from other sources, this macrophyte production is obviously channeled to the pelagic food web via the bacterioplankton.

KEY WORDS: Shallow lake · Bacteria · Phytoplankton · Reed · *Phragmites* · Dissolved organic matter · Humic substances

INTRODUCTION

Bacterioplankton are closely linked to phytoplankton via the release of dissolved organic carbon (DOC) from phytoplankton either directly via extracellular release or indirectly via viral lysis or grazing of herbivorous zooplankton (Lampert 1978, Azam et al. 1983, Peduzzi & Herndl 1992). Recently it has also been shown that grazing activity of protists contributes significantly to the production of DOC (Tranvik 1994, Strom et al. 1997). This close trophic dependence of bacterioplankton activity on phytoplankton has been shown in numerous studies for marine and large lotic systems (Chróst 1986, Riemann & Søndergaard 1986, Vadstein et al. 1989, Weisse et al. 1990, Caron 1994, Ducklow et al. 1995). DOC supply for bacterioplankton growth in small lotic and lentic systems is mediated to

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a large extent by non-phytoplankton or allochthonous sources (Servais & Garnier 1990, Cotner & Wetzel 1992, Hudson et al. 1992, Berger et al. 1995, Fiebig 1995, Mann & Wetzel 1996). With decreasing volume of the water body, the autochthonous DOC production from non-phytoplankton sources becomes increasingly important for the system's metabolism (Wetzel 1992). Findlay et al. (1992), for example, estimated that in the Hudson Estuary, the amount of allochthonous carbon input needed to support bacterial productivity is 3 to 6 times the net carbon fixation by phytoplankton. Coveney & Wetzel (1995) arrived at a similar conclusion in their detailed study of Lawrence Lake. The net heterotrophic pelagic system was fueled by periphytic and macrophytic production.

The shallow Lake Neusiedl (mean depth 1.1 m), Austria, is characterized by a large reed belt of *Phragmites australis* covering about 55% of the total lake surface (Löffler 1979), and low transparency of the water column due to wind-induced resuspension limiting the availability of light for phytoplankton production (Dokulil 1994). Nevertheless, the pelagic food web is well-developed with large standing stocks of protists and mesozooplankton (Herzig 1979). Thus, one might assume that bacteria, and not phytoplankton, provide the base for the pelagic food web. Heterotrophic bacteria, in turn, might utilize the DOC becoming available from the reed *P. australis* and its periphytes. Water exchange between the reed belt and the open areas is crucial to support bacterial activity in the open water and, indirectly, the pelagic food web. Measuring the DOC dynamics in the open waters and within the reed belt might provide insight into the dynamic exchange processes. Large differences in the concentration of DOC and bacterioplankton activity between the reed belt and the open waters indicate low exchange, while a more even distribution of DOC and bacterial production might indicate rapid exchange processes. Another explanation for an evenly distributed DOC concentration and bacterial activity, however, could be that wind-induced mixing provides DOC from porewater of the sediment to the open water bacteria. Wind-induced resuspension is rather limited in the reed belt. If the pelagic production (phytoplankton and bacteria) is driven by wind-induced input of nutrients from the porewater of the sediment, and not via exchange processes between the reed belt and the open water, the pattern in phytoplankton and bacterial production would not covary between the 2 sites.

In order to decipher the connectivity between the reed belt and the open waters of the shallow, eutrophic Lake Neusiedl, we compared the DOC concentrations and the microbial community and activity at 3 different sites (the open waters, the waters within the reed belt and an open pond in the reed belt) over a year. At all 3 stations, bacterial carbon demand exceeded phytoplankton production by more than 1 order of magnitude, indicating the importance of non-phytoplankton-derived DOC for bacterioplankton metabolism in Lake Neusiedl.

MATERIALS AND METHODS

Sampling location. A detailed description of the limnology of Lake Neusiedl is given in Löffler (1979). Briefly, Lake Neusiedl (47° 42' N, 16° 46' E) is the largest shallow, alkaline brown-water lake in central Europe (115 m a.s.l., surface area 321 km^2, maximum depth 1.8 m, mean depth 1.1 m, pH 8.5 to 9.1). About 55% of the lake is covered by the reed *Phragmites australis* (178 km^2); within this reed belt, there are numerous reedless ponds of variable size. The water level of the lake is controlled by precipitation (500 to 700 mm yr^{-1}) and evaporation. Frequent resuspension of the sediment caused by winds and currents results in a high concentration of suspended solids in the water column during the ice-free period (Secchi depth ≈0.2 m); during winter, when the lake is ice-covered for up to 3 mo, the concentration of the suspended solids declines due to the reduced water column turbulence. Reedless ponds within the reed belt, such as the Ruster Poschn (surface area 40 000 m^2) are more sheltered from wind-induced mixing and resuspension of the sediments and exhibit therefore a higher transparency (Secchi depth ≈0.6 m). Due to the shallow water column of Lake Neusiedl, water temperature can change rapidly in spring and fall.

Sampling. Water samples were taken with cleaned (1 N HCl, and rinsed 3 times with water from the sampling site) 5 l polycarbonate flasks at 3 characteristic stations, at weekly to 4-weekly intervals, from November 1996 to October 1997. Stn L (Lake) represents the open lake, and Stn RP (Ruster Poschn) is in the center of a reedless pond within the reed belt. At these 2 stations, samples were taken from 0.5 m depth. The other sampling station was located within the reed belt (Stn R). There, water samples were taken at 0.2 m depth (water column depth 0.5 m). During sampling, water temperature was measured in the surface (0.2 m depth) and bottom layer (0.2 m above bottom at the 2 open water stations) and the photosynthetic active radiation (PAR, 400 to 700 nm) at 10 cm depth intervals was measured with a LiCor 1000 radiometer. From these radiation measurements, attenuation coefficients were calculated for each station. The collected water was stored in an isolated box in the dark and brought back to the laboratory (Biological Station Illmitz) within 30 min.

Chlorophyll *a* and phaeophytin. We filtered 200 to 1500 ml of the water sample onto Whatman GF/F

(glass fiber) filters (47 mm filter diameter) and stored frozen (–20°C) in glass scintillation vials until analysis. Pigments were extracted from the filters in 10 ml of 90% acetone at 4°C overnight and the pigment concentration estimated spectrophotometrically with a Merck-Hitachi U 2000, using the equations given in Parsons et al. (1984). For the determination of the phaeopigments, the solution was acidified with 2 drops of 2 N HCl and the absorbance measured again (Parsons et al. 1984).

Phytoplankton primary production. Phytoplankton primary production was measured by the uptake of radiolabeled ^{14}C-sodium bicarbonate and its subsequent conversion into organic carbon. We poured 50 ml subsamples through a 62 µm mesh (to remove potential grazers) into HCl-rinsed BOD-flasks to which 1 µCi $Na(H^{14}CO_3)_2$ (Amersham) was added. Bottles were incubated in duplicates in an outdoor tank filled with lake water and exposed to 100, 50, 30, and 10% of surface solar radiation at *in situ* temperature from dawn to dusk with 2 dark controls (wrapped in aluminum foil). After incubation, the samples were filtered onto 0.45 µm pore-size cellulose nitrate filters (Millipore HAWP, 25 mm diameter) and rinsed with 10 ml of 0.45 µm filtered lake water. Thereafter, the filters were exposed to a fume of concentrated HCl for 2 h to remove inorganic carbon and placed in scintillation vials. Filters were dissolved in 1 ml ethyl acetate (Riedel de Haen), and 8 ml scintillation cocktail (Packard Insta plus gel) was added. After 10 h, radioactivity was measured using a Packard Tri-Carb 2000 scintillation counter with external standard ratio technique for sample quenching. For calculating primary production, the mean disintegrations per minute (dpm) of the dark controls was subtracted from the mean dpm of the light-exposed treatments at each radiation level. Estimates of phytoplankton primary production of the respective water column were obtained by integrating the measured daily production at the different radiation levels over the water column.

Enumeration of bacteria. Twenty ml subsamples were fixed with concentrated formalin (2% v/v, final conc.); depending on the bacterial abundance, 1 to 6 ml were used to enumerate bacterial abundance by DAPI staining and epifluorescence microscopy at 1250× magnification (Porter & Feig 1980). Bacterial abundance was enumerated on duplicate subsamples. At least 300 bacteria were counted per filter. Bacterial abundance was converted into bacterial carbon biomass assuming a carbon content of 20 fg C $cell^{-1}$ (Lee & Fuhrman 1987).

Measurement of bacterial activity. Bacterial activity was assessed by measuring the incorporation of [^{3}H]-thymidine (Amersham, SA = 85 Ci $mmol^{-1}$, 20 nM final conc.) into bacterial DNA (Fuhrman & Azam 1982) and [^{3}H]-leucine (Amersham, SA = 131 Ci $mmol^{-1}$, 10 nM final conc.) into bacteria (Simon & Azam 1989). Five ml subsamples were incubated in triplicates with 2 formalin-killed controls for 30 to 90 min (depending on the *in situ* temperature) at *in situ* temperature in the dark. After incubation, the subsamples were filtered onto cellulose nitrate filters (Millipore, HAWP 0.45 µm, 25 mm filter diameter) and rinsed 3 times with 5% chilled TCA for 5 min. Radioactivity of the filters was measured after adding 1 ml ethyl acetate (Riedel de Haen) and 8 ml scintillation cocktail (Packard Insta plus gel); after 10 h, radioactivity was measured as described above. Thymidine (TdR) incorporation into the bacterial cells was converted into bacterial cell production using the conversion factor of 1.8×10^{18} cells produced mol^{-1} TdR; for leucine (leu) a factor of 0.07×10^{18} cells mol^{-1} leu was used (Reitner et al. 1997a). These conversion factors were determined in dilution cultures with water collected from Stn RP over a seasonal cycle by measuring the TdR and leu incorporation into bacteria during their exponential growth in the absence of grazers.

Fractionation of DOC into humic and non-humic components. Only water collected at Stn L was fractionated into humic and non-humic DOC. Upon return to the laboratory, 1.5 l of the water collected at Stn L was filtered through combusted (450°C for 4 h) Whatman GF/F filters (100 mm diameter) mounted in a combusted glass filter holder; thereafter, the pH of the water was measured and 10 ml of the sample withdrawn, acidified and stored at –20°C for subsequent DOC analysis (described below). All the glassware used was thoroughly acid-washed (with 1 N HCl), rinsed with double-distilled water and combusted (450°C for 4 h).

From the 1.5 l of the Whatman GF/F-filtered water, 0.5 l was fractionated into a humic and a non-humic fraction of the DOC using macroporous Amberlite XAD-8 resin according to the method described in detail elsewhere (Aiken 1985, Moran & Hodson 1990, Reitner et al. 1997b). Briefly, the water was adjusted to pH 2 with 6 N HCl and poured through a column filled with Amberlite XAD-8 resin and subsequently eluted with 200 ml of 0.1 N NaOH; in this paper we define the humic fraction of the DOC as material retained by the macroporous resin Amberlite XAD-8 at pH 2 and subsequently eluted with 0.1 N NaOH. Previous studies have shown that the humic fraction isolated with Amberlite XAD-8 is similar to those isolated by other methods (Peuravuori et al. 1997). Both the DOC fraction not retained by the XAD-8 resin (considered as the non-humic fraction) and the fraction eluted from the XAD-8 resin were adjusted to pH 10 and poured through a cationic exchange column filled with Amberlite IR-118H. Water flow through the columns was

adjusted to a rate of ~40 ml min^{-1}. Subsequently, the humic and the non-humic fraction of the DOC were adjusted to the original pH with 6 N HCl and 2 N NaOH and brought up to the original volume with 0.2 µm filtered double-distilled water. Thereafter, samples for DOC analysis were taken from the humic and the non-humic fraction, as well as from the double-distilled water, acidified to pH 2 with 50 µl of 6 N HCl and stored frozen (–20°C) together with the acidified unfractionated DOC samples in combusted glass scintillation vials with Teflon-lined caps until DOC analysis. Acidification of the water before freezing the samples prevented precipitation and flocculation in the samples and therefore greatly improved the reproducibility of replicate DOC measurements (Reitner et al. 1997b).

Determination of DOC. The acidified samples were completely thawed and the DOC content was determined using a Shimadzu TOC-5000 after sparging the samples with CO_2-free air. Standards were prepared with potassium hydrogen phthalate (Kanto Chemical Co. Inc.); a platinum catalyst on quartz was used (Benner & Hedges 1993, Reitner et al. 1997b). Five replicate measurements were performed per sample. The overall analytical precision was always better than 3%.

Absorption of DOC. The absorbance characteristics of the DOC were measured against double-distilled water at 250 and 365 nm and, additionally, scans from 500 to 250 nm (data not shown) were made using a Beckman DU 640-I and a 5 cm quartz cuvette. The absorption ratio between 250:365 nm was calculated in order to determine possible differences and shifts in the molecular size spectrum of the DOC between the sampling stations and over the investigation period (Stewart & Wetzel 1980, 1981).

RESULTS

Temperature regime and PAR attenuation

Over the entire investigation period, water temperature varied between 0.1 and 23.7°C. The ice cover lasted from the end of December to February, reaching its maximum thickness (28 cm) in early February. During the ice cover, the near bottom waters of Stns L and RP were about 1°C warmer than the surface water layers underneath the ice. The entire water column of Stn R (within the reed belt) was frozen during January and February. The smallest seasonal variation of the attenuation coefficient (1.22 to 4.47 m^{-1}, mean ± SD = 2.24 ± 0.68 m^{-1}, n = 22) was found at Stn RP, which is largely protected from wind-induced sediment resuspension, while Stn L showed the largest variability (1.39 to 14.71 m^{-1}, mean ± SD = 4.96 ± 3.20 m^{-1}, n = 22). The attenuation coefficient for Stn R ranged from 1.80 to 10.13 m^{-1} (mean ± SD = 4.04 ± 2.45 m^{-1}, n = 19).

DOC dynamics

The absorption ratios, between 250:365 nm, of the unfractionated DOC for the different sampling sites were highest at Stn L (mean ± SD: 9.72 ± 1.07, range: 7.89 to 13.03, n = 22) and lowest at Stn R (mean ± SD: 6.87 ± 0.72, range: 5.84 to 8.27, n = 18) (Fig. 1A). The absorption ratios of Stn RP were between the ratios of the other 2 stations (mean ± SD: 8.30 ± 0.59, range: 7.13 to 9.54, n = 22) (Fig. 1A). All the site-specific absorption ratios were significantly different from each other (ANOVA, Bonferroni, $p < 0.0001$). There were no distinct seasonal dynamics detectable for Stns L and RP; however, at Stn R, the absorption ratio declined from spring to August (Fig. 1A).

Generally, DOC concentrations were lowest and flucuated only over a small range at Stn L (mean ± SD: 1.24 ± 0.17 mmol C l^{-1}, range: 1.03 to 1.64 mmol C l^{-1}, n = 22), and slightly higher with a larger seasonal variation at Stn RP (mean ± SD: 1.36 ± 0.35 mmol C l^{-1}, range: 0.96 to 2.47 mmol C l^{-1}, n = 22) (Fig. 1B). At Stn R, within the reed belt, DOC concentrations were siginificantly higher (ANOVA, Bonferroni, $p < 0.0001$, n = 10) than at the 2 other stations during the vegetation period of the reed *Phragmites australis* (mean ± SD: 1.50 ± 0.17 mmol C l^{-1}, range: 1.22 to 1.84 mmol C l^{-1}, n = 19) (Fig. 1B).

Fractionation of the bulk DOC from Stn L into the humic and non-humic fractions resulted in an average recovery of 95.6 ± 7.8% (range: 84.2 to 113.6, n = 21) of the unfractionated bulk DOC. The mean contribution of humic substances to the total DOC was 46.1 ± 5.4% (range: 36.9 to 55.4%, n = 21), or in terms of carbon, 0.57 ± 0.10 mmol C l^{-1} (range: 0.38 to 0.76 mmol C l^{-1}, n = 21) (Fig. 1C). Generally, DOC concentrations declined from winter to the end of August (Fig. 1C). The contribution of humic DOC to the bulk DOC was around 45% during the winter, increased in spring to a maximum of 55% and declined again in early fall (Fig. 1C). The concentration of the non-humic DOC ranged from 0.40 to 0.85 mmol C l^{-1} (mean ± SD: 0.61 ± 0.13 mmol C l^{-1}, n = 21).

Seasonal dynamics of phytoplankton biomass and production

Phytoplankton biomass measured as chlorophyll *a* (chl *a*) concentration ranged from 2.14 to 17.10 µg chl *a* l^{-1} at Stn RP (mean ± SD: 5.85 ± 3.47 µg chl *a* l^{-1}), at Stn R from 2.44 to 10.32 µg chl *a* l^{-1} (mean ± SD: 5.12 ± 1.97 µg

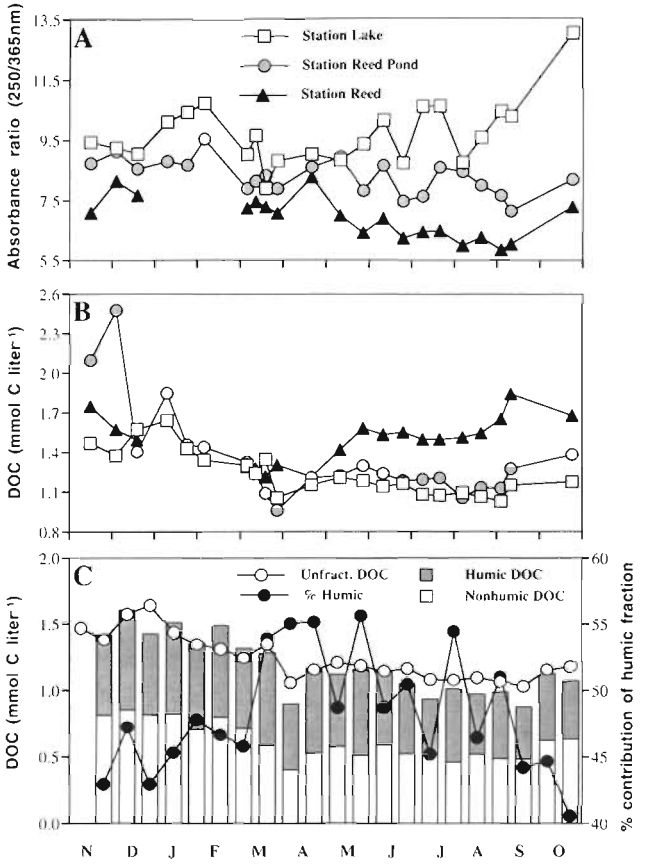

Fig. 1 DOC dynamics at the 3 sampling sites in the Lake Neusiedl over the study period from November 1996 through October 1997. (A) Absorption ratio of 250:365 nm wavelength; (B) total DOC concentration; and (C) distribution of humic and non-humic DOC and the % contribution of the humic DOC to the total DOC (right axis); for comparison, the total, unfractionated DOC concentration is also given

chl *a* l^{-1}) and was slightly higher at Stn L (mean ± SD: 7.28 ± 3.96 µg chl *a* l^{-1}, range: 2.42 to 16.57 µg chl *a* l^{-1}). From March until June, Stn L was characterized by generally high chl *a* concentrations, while at Stn RP highest chl *a* concentrations were reached in January and March (Fig. 2A). No site-specific significant differences in the chl *a* concentrations were detectable (ANOVA, Bonferroni, $p > 0.0589$). Concentrations of phaeopigments varied considerably over the seasonal cycle (Stn L: 0.77 ± 1.05 µg l^{-1}, Stn RP: 1.65 ± 1.35 µg l^{-1}, and Stn R: 1.15 ± 1.43 µg l^{-1}) (data not shown).

Mean primary production integrated over the water column was 16.59 ± 14.93 mg C m^{-2} d^{-1} for Stn L, 19.81 ± 25.54 mg C m^{-2} d^{-1} for Stn RP, and 1.94 ± 3.74 mg C m^{-2} d^{-1} for Stn R (Fig. 2B). Two pronounced peaks in phytoplankton production were found for the open water Stns L and RP: a first smaller peak after the ice melt in mid-March, and a large peak in mid-August. The latter peak in primary production was also detectable, although not as pronounced, at Stn R (Fig. 2B). This peak in activity is also reflected in the specific productivity (Fig. 2C), indicating that highly active cells at comparatively low concentrations were responsible for the high phytoplankton production rates in August. This high production coincided with a shift in the phytoplankton community. While the abundance of the cyanobacterium *Microcystis* sp. declined in August by about 1 order of magnitude, the abundance of Chrysophyceae remarkably increased (data not shown).

Seasonal dynamics of bacterial abundance and activity

Bacterial abundance (Fig. 3A) at the 3 stations showed similar fluctuations throughout the annual cycle, with lowest abundances at low water temperatures. At Stn L, 73% of the fluctuations of bacterial abundance could be explained by water temperature; at Stns RP and R, this was 59 and 57%, respectively. The highest mean bacterial abundance was observed at Stn R (mean ± SD = 59.4 ± 26.9 × 10^5 cells ml^{-1}, range: 21.1 to 116.0 × 10^5 ml^{-1}, n = 19). At Stn RP, the abundance varied from 22.4 to 132.4 × 10^5 ml^{-1} (mean ± SD = 56.6 ± 26.6 × 10^5 ml^{-1}, n = 22) and the lowest mean bacterial abundance (mean ± SD = 51.7 ± 16.9 × 10^5 cells ml^{-1}, range: 24.8 to 81.6 × 10^5 ml^{-1}, n = 22) was recorded for Stn L (Fig. 3A).

Bacterial production calculated from TdR incorporation measurements showed the expected, distinct seasonal pattern with low production during the winter (lowest production at Stn L: 5.3 µg C l^{-1} d^{-1}) and a rapid increase after the melting of the ice by the end of March, and consistently high rates during summer (between 100 and 300 µg C l^{-1} d^{-1} for all 3 stations) (Fig. 3B). Averaging bacterial production over the seasonal cycle estimated from TdR and leu incorporation measurements, respectively, indicated that bacterial production estimated by the 2 methods were not significantly different from each other for Stns RP and R (Fig. 3B,C) although the 2 measurements did not always exhibit the same tendency. Mean bacterial production estimated by TdR and leu incorporation was for Stn RP 126.0 ± 76.4 µg C l^{-1} d^{-1} and 110.0 ± 49.0 µg C l^{-1} d^{-1}, respectively, for Stn R 126.9 ± 68.9 µg C l^{-1} d^{-1} and 119.4 ± 80.8 µg C l^{-1} d^{-1}, and for Stn L, 102.5 ± 70.5 µg C l^{-1} d^{-1} and 46.7 ± 34.4 µg C l^{-1} d^{-1}, and therefore significantly lower for leu than for TdR (Wilcoxon, $p < 0.001$).

Bacterial growth rates determined from TdR and leu incorporations exhibited large variations over the seasons, ranging from 0.05 to 2.57 d^{-1} for all sampling sites; the mean annual bacterial growth rate based on TdR incorporation was 0.91 ± 0.51 d^{-1} for Stn L, 1.09 ±

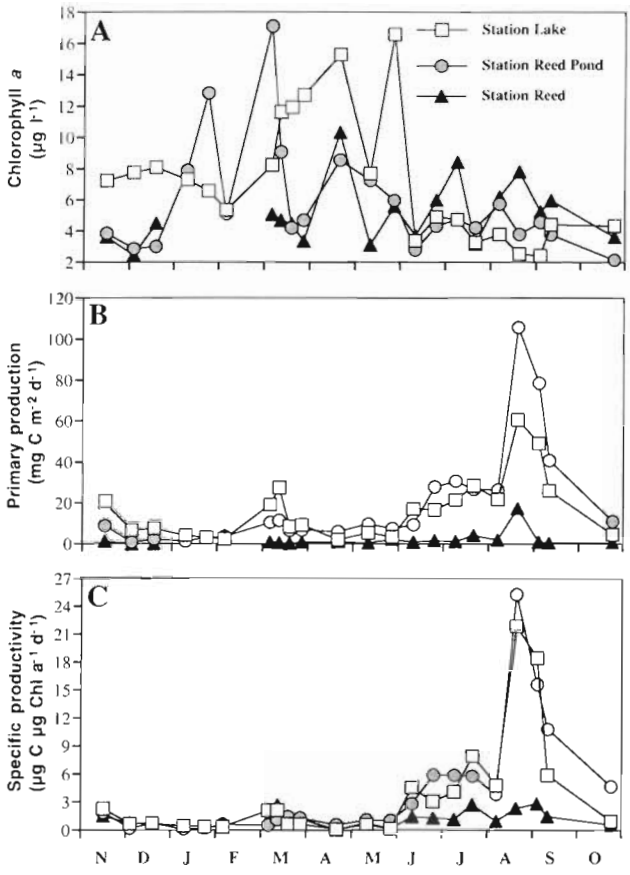

Fig. 2. Phytoplankton dynamics at the 3 stations in Lake Neusiedl over the investigation period. (A) Chlorophyll *a* concentrations; (B) phytoplankton production; and (C) phytoplankton specific productivity

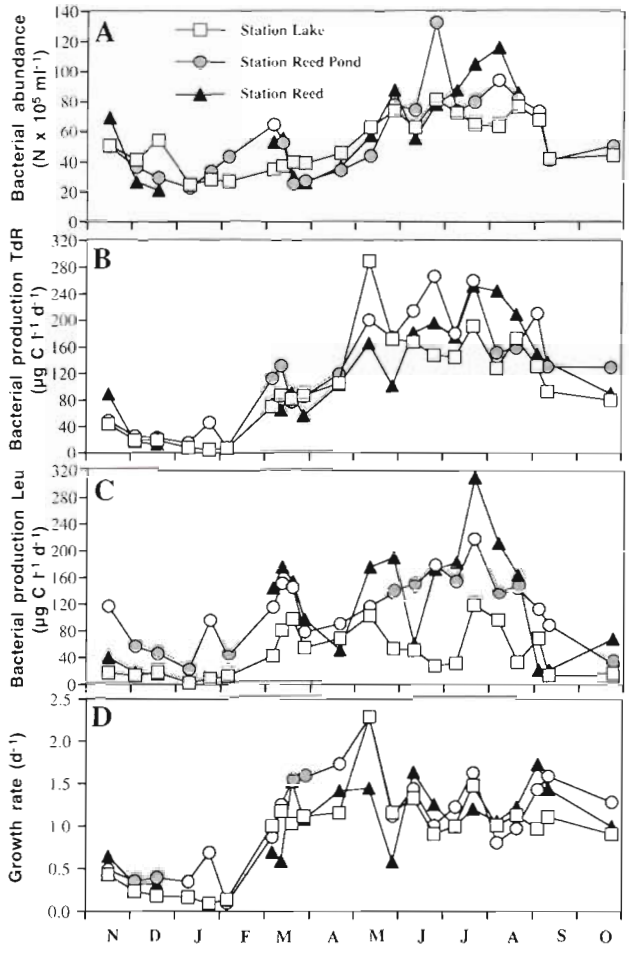

Fig. 3. Fluctuations in (A) bacterial abundance, bacterial production measured by (B) thymidine (TdR) and (C) leucine (Leu) incorporation, and (D) growth rates (bacterial production estimated from TdR incorporation/biomass) at the 3 sampling sites

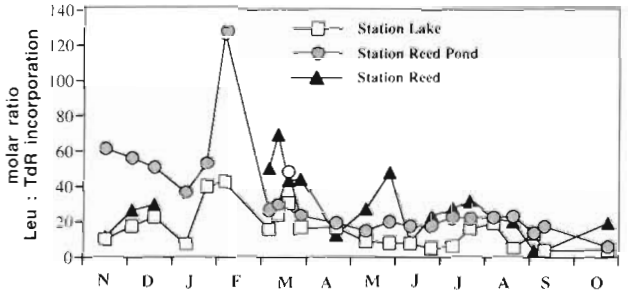

Fig. 4. Dynamics of the molar ratio of Leu:TdR incorporation at the 3 sampling sites over the investigation period

0.54 d^{-1} for Stn RP and 1.06 ± 0.42 d^{-1} for Stn R (Fig. 3D). Mean bacterial growth rates based on leu incorporation were for Stn L 0.45 ± 0.34 d^{-1}, for Stn RP 1.05 ± 0.51 d^{-1}, and for Stn R 0.99 ± 0.61 d^{-1}. Bacterial growth rates were significantly lower at Stn L for both leu and TdR as compared to the more sheltered Stns RP and R (Wilcoxon, $p < 0.001$, $n = 19$, for both).

The molar ratio of leu:TdR incorporation was 15.5 ± 10.8 (range: 3.8 to 42.6, $n = 22$) for Stn L, 33.2 ± 25.4 (range: 6.1 to 126.6, $n = 22$) for Stn RP and 27.5 ± 16.8 (range: 3.7 to 69.3, $n = 19$) for Stn R, and therefore significantly higher at Stns RP and R (Wilcoxon, $p < 0.001$, $n = 19$, for both) than at Stn L (Fig. 4). Generally, higher molar leu:TdR incorporation ratios were observed during winter and lower ratios during the summer months (Fig. 4).

DISCUSSION

The sampling stations used in this study comprised all the characteristic environments of the lake: the open water, the ponds within the reed belt and the water of the reed belt itself. They are characterized by different turbulence regimes. While mixing in the open water (Stn L) prevents stratification throughout the year, Stn RP is more protected from wind-induced

turbulence and, consequently, has a lower attentuation of light. Stn R inside the reed belt is characterized by a shallow water column (~30 cm) and reduced exchange of water with the open lake. Nevertheless, the development of the phytoplankton bloom in August was detectable at all 3 stations (Fig. 2), indicating that water exchange is sufficient between the open water and the reed belt to allow a concurrent development of plankton blooms in the different subsystems.

Major differences between the 3 subsystems were detectable for DOC. Stn R, within the reed belt, exhibited consistently higher DOC concentrations than the other 2 stations from April to October (Fig. 1B). Also, the absorption ratio of the DOC was significantly lower at Stn R, indicating a higher fraction of labile DOC than at the other 2 stations. Since the specific production of phytoplankton was lower at Stns RP and L than at Stn R, the higher fraction of labile DOC probably originates from the reed and its epiphytes. Epiphytes might release substantial amounts of extracellular release (Haines et al. 1987, Grimshaw et al. 1997). The higher fraction of labile DOC at Stn R, however, did not cause elevated bacterial abundance and growth rates as compared to Stns RP and L (Fig. 3).

Bacterial versus phytoplankton production

Mean annual bacterial production based on TdR incorporation for the 3 stations ranged from 38.1 mg C m^{-2} d^{-1} at Stn R to 139 mg C m^{-2} d^{-1} at Stn RP and based on leu from 36 to 121 mg C m^{-2} d^{-1}, respectively. Mean annual phytoplankton production ranged from 2 to 20 mg C m^{-2} d^{-1}, at Stns R and RP, respectively (Table 1). The bacterial growth yield was determined in a previous study (Reitner et al. 1997a) and in a preliminary experiment. While in Reitner et al. (1997a) the bacterial growth yield was estimated from the DOC decline in the ice-covered lake (no DOC input) and the concomitantly performed bacterial incorporation of thymidine over a period of 3 mo, in a subsequent exercise during the summer months, bacterial production and respiration were measured in 0.8 µm filtrates. Both independent estimates resulted in a bacterial growth yield of 16%. This bacterial growth yield is in the range of growth yields reported from a diverse range of freshwater and marine systems (del Giorgio & Cole 1998).

Using the growth yield of 16% determined for Lake Neusiedl (Reitner et al. 1997a), the mean annual bacterial carbon demand at the 3 different stations ranges

Table 1. Summary of bacterial carbon demand (bacterial C demand) at the 3 stations (L: Lake, RP: Reed Pond, R: Reed) of Lake Neusiedl, Austria, based on leucine (Leu) and thymidine (TdR) incorporation measurements, phytoplankton production and the ratio between bacterial carbon demand and phytoplankton production. Bacterial carbon demand was estimated using a bacterial yield of 16% (Reitner et al. 1997a). nd: not determined due to completely frozen water column. Rates are given in mg C m^{-2} d^{-1}

Date (d/mo/yr)	Bacterial C demand based on TdR			Bacterial C demand based on Leu			Primary production			Bacterial C demand TdR/ primary production			Bacterial C demand Leu/ primary production		
	L	RP	R	L	RP	R	L	RP	R	L	RP	R	L	RP	R
15/11/96	303	337	168	119	809	75	20.0	8.9	1.3	15	38	134	6	91	60
03/12/96	135	179	32	92	393	33	6.6	0.8	0.0	20	212		14	465	
18/12/96	137	160	25	122	318	30	7.5	2.4	0.0	18	66	946	16	131	1102
08/01/97	57	109	nd	16	156	nd	4.2	1.4	nd	14	79	nd	4	113	nd
22/01/97	36	320	nd	57	662	nd	3.2	3.1	nd	12	103	nd	18	214	nd
03/02/97	52	58	nd	86	285	nd	2.4	3.6	nd	22	16	nd	36	80	nd
05/03/97	486	779	139	296	801	272	19.1	10.4	0.9	25	75	148	16	77	288
11/03/97	604	912	123	560	1047	331	27.5	11.4	0.6	22	80	221	20	92	597
18/03/97	565	533	171	675	1005	290	8.5	6.6	0.2	66	81	851	79	152	1438
26/03/97	602	592	107	380	542	183	9.2	6.7	0.8	66	88	126	41	81	216
16/04/97	731	827	196	476	627	96	2.2	6.0	1.0	340	137	196	221	104	97
05/05/97	2000	1387	312	710	807	331	5.5	9.7	0.5	367	142	596	130	83	632
21/05/97	1189	1206	191	369	946	358	3.5	7.4	2.2	340	162	87	106	127	163
04/06/97	1160	1484	343	354	1015	113	17.0	9.4	0.9	68	158	369	21	108	122
18/06/97	1023	1843	370	192	1234	325	16.7	27.9	1.7	61	66	218	12	44	192
02/07/97	1002	1248	330	219	1065	344	21.5	30.7	1.3	47	41	264	10	35	275
14/07/97	1325	1797	475	820	1507	584	28.4	26.7	4.2	47	67	114	29	56	140
30/07/97	888	1050	461	667	928	398	21.7	26.4	1.9	41	40	245	31	35	212
12/08/97	1199	1099	395	231	995	309	60.6	105.7	17.3	20	10	23	4	9	18
26/08/97	909	1457	282	477	780	40	49.1	78.5	0.9	19	19	310	10	10	44
02/09/97	644	903	258	94	613	41	26.0	40.8	0.4	25	22	620	4	15	98
15/10/97	554	895	170	90	213	128	4.6	10.9	0.7	120	82	240	19	19	181
Mean	709	872	239	323	761	225	16.6	19.8	1.9	81	81	317	39	97	326
SD	488	529	130	238	339	153	14.9	25.5	3.7	110	52	256	51	94	375

from 240 to 872 mg C m^{-2} d^{-1} (based on TdR incorporation) and from 225 to 761 mg C m^{-2} d^{-1} (based on leu incorporation)(Table 1). Therefore, bacterial C demand is at least 1 order of magnitude higher (39 to 326 times) than the amount of carbon supplied by phytoplankton production. In our measurements of the primary production we did not include the dissolved production, i.e. the release of photosynthetically fixed DOC into the ambient water, which serves as a potential food source for bacterioplankton. Even high release rates of phytoplankton-derived DOC would not match the C demand by bacterioplankton. Release rates are usually reported to be in the range of 0 to 30% of the photosynthetically fixed carbon (Lignell 1990, Williams 1990, Malinsky-Rushansky & Legrand 1996) but can be as high as 70% under P-limited conditions (Kaltenböck & Herndl 1992, Obernosterer & Herndl 1995). Thus, in Lake Neusiedl phytoplankton-derived DOC is only of minor importance for bacterioplankton metabolism. Even at Stn L, which reflects the open water conditions of the lake, bacterial C demand was always higher than phytoplankton primary production, even under bloom conditions (at leads ~4 times, Table 1). Although there is some uncertainty in the bacterial production estimates since we used a single conversion factor for TdR and Leu, the general conclusion that bacterial C demand cannot be met by phytoplankton production is robust.

The C demand for bacterioplankton is therefore likely to be matched by the large-standing crop of *Phragmites australis* and its periphytes. The primary production of *P. australis* amounts to 1370–8200 mg C m^{-2} reed belt d^{-1} (Kvet & Husak 1978) and about 55% of the lake is covered by *P. australis* (Löffler 1979). Calculating the average primary production of *P. australis* for the entire lake, the primary production of the reed amounts to 750–4510 mg C m^{-2} d^{-1}. Therefore, the estimated bacterial C demand for bacterioplankton ranging from 225 to 870 mg C m^{-2} d^{-1} (depending on the stations and the substrate used) is only slightly lower than the primary production estimates for *P. australis* (Table 1). The high contribution of this cellulose-rich material to the DOC pool is evident from the data on the contribution of humic material to the bulk DOC (Fig. 1). In a previous study (Reitner et al. 1997a), it was shown that the contribution of humic DOC to bulk DOC declines during the winter. In the present study, the same tendency was found; low humic DOC contribution was detectable during the winter and increased towards the summer to values above 55% (Fig. 1).

Based on the bacterioplankton C demand calculated from TdR incorporation measurements, a bacterial growth yield of 16% (Reitner et al. 1997a) and the DOC concentrations at the 3 different stations, we arrived at a mean DOC turnover time of 86 d (range: 8 to 519 d, n = 22) for Stn L, 57 d (range: 8 to 328 d, n = 22) for Stn RP, and 42 d (range: 11 to 212 d, n = 19) for Stn R. Based on leu incorporation, mean turnover times were 150 d (range: 17 to 1325 d, n = 22), 36 d (range: 11 to 157, n = 22) and 56 d (range 9 to 182, n = 19) for Stns L, RP, and R, respectively. As expected, DOC turnover times were generally higher during the winter season and lower during the summer.

In summary, major differences between the 3 subsystems were found only in the concentration and quality of the DOC. Within the reed belt, DOC concentrations were higher with a higher fraction of the DOC being labile. Since there were no significant differences detectable in phytoplankton chl *a* concentrations and the specific phytoplankton production between the stations, we concluded that the elevated DOC concentrations within the reed belt originate from the reed and its periphytes. Overall, bacterioplankton metabolism in Lake Neusiedl is always at least 1 order of magnitude higher than phytoplankton production and, therefore, depends on non-phytoplankton-derived DOC, which is probably supplied by the reed *Phragmites australis* and its periphytes. Thus, the pelagic food web of Lake Neusiedl is based largely on reed (and its periphytes) production; its DOC production is exported into the open waters and supports bacterioplankton growth there. Thus, the basis of the well-developed pelagic food web in Lake Neusiedl is bacterioplankton which are preyed upon by protists and possibly also by mesozooplankton. Phytoplankton play only a minor role in suporting the pelagic food web. Future studies should focus on the DOC production of the reed *P. australis* and its periphytes.

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