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Research Article

Vertical gradient of nutrients in two dimictic lakes – influence of phototrophic sulfur bacteria on nutrient balance

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Abstract. Vertical profiles of soluble and particulate nutrients were analyzed at the end of summer stratification in two dimictic lakes located in northeast Germany. In addition, irradiance and plankton biomass were determined. The concentrations of particulate organic carbon and phytoplankton biomass in the epilimnion were higher in Lake Tiefer than in Lake Dudinghausen, even though the apparent trophic status of Lake Tiefer was higher than Lake Dudinghausen. In Lake Dudinghausen, phototrophic sulfur bacteria accumulated in the hypolimnion between 8 and 10 m, whereas in Lake Tiefer low light penetration prevented the development of phototrophic bacteria in those horizons in which sulfide might be present. Because both lakes have anoxic hypolimnia, we assumed that in both cases phosphorus was released from the sediment into the hypolimnion. In Lake Tiefer, redox conditions and the presence of nitrate and nitrite limited the water depth range in which P-release occurred. In Lake Dudinghausen, part of the released soluble reactive phosphorus was incorporated into the phototrophic sulfur bacteria biomass and thus transformed to particulate phosphorus. As much as 70% of the particulate phosphorus in the hypolimnion was found in the phototrophic sulfur bacterial layer, with 15–20% of this particulate phosphorus consisting of polyphosphate storage compounds. The low ratio of soluble reactive phosphorus to particulate phosphorus in the hypolimnion was, therefore, attributed to phototrophic sulfur bacteria. The phototrophic sulfur bacteria appear to act as an internal nutrient filter and convert soluble reactive phosphorus into particulate phosphorus.

Key words. Phosphorus; polyphosphate; phototrophic sulfur bacteria; phytoplankton; zooplankton.

Introduction

Vertical distribution and transformation of phosphorus in lakes is the net result of P-loading from the catchment area, sedimentation of particulate matter, chemical precipitation, P-regeneration and P-accumulation in plankton, and P-release from the sediment. In stratified lakes, gradients of temperature, light and oxygen influence the vertical flux of nutrients (Kufel and Kalinowska, 1997; Kleeberg and Schubert, 2000) as well as the vertical distribution of plankton communities (Reynolds, 1992). Often, strong gradients of nutrients are found in the metalimnion.

In addition to gradients of dissolved nutrients, a large accumulation of particulate nutrients may also occur in the metalimnion (Rodrigo et al., 2000). This accumulation is caused by reduced settling velocity, resulting in the mass development of autotrophic and heterotrophic plankton (Finlay et al., 1996; Gervais, 1998). Whether phototrophic sulfur bacteria are part of this plankton community in the metalimnion depends mainly on the availability of sulfur and light. If conditions are favorable, these bacteria can form dense layers with an intense pink, brown or green color, not only in the metalimnion, but reaching also into the hypolimnion. In most cases, Chromatiaceae dominate

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such populations and can contribute up to 90% of metalimnetic bacterial biomass (Guerrero et al., 1985).

The irradiance requirements of phytoplankton and phototrophic bacteria are quite different with respect to spectral composition and intensity (Vila et al., 1996a). The few investigations on irradiance dependency of phototrophic bacteria have shown that their light saturation points of photosynthesis (E_k) are very low compared with those of phytoplankton (Schanz et al., 1998). On the other hand, because light availability in situ is often quite low, many investigators have demonstrated that phototrophic bacterial communities are light-limited regardless of their low E_k (Camacho and Vicente, 1998; Sanchez et al., 1998; Rodrigo et al., 2000).

Phototrophic sulfur bacteria are able to synthesise a wide variety of storage compounds: sulfur, glycogen, poly-β-hydroxybutyrate and polyphosphate (Mas and Van Gemerden, 1995). Polyphosphate storage by anaerobic phototrophic sulfur bacteria was first described in the 1960s (Hughes et al., 1963; Zaitseva et al., 1965), but little is known about its ecological relevance. Polyphosphate storage compounds, serving as energy and phosphate reservoirs, are essential for growth of many microorganisms (Kornberg, 1995). Unfortunately, because field data on phototrophic sulfur bacteria exist only for a meromictic lake (Baneras and Garcia-Gil, 1996), the influence of these organisms on the P-cycle and P-retention in dimictic lakes cannot be representatively estimated. Züllig (1985) has suggested, however, that the occurrence of phototrophic sulfur bacteria is typical in mesotrophic stratified lakes. This begs the question as to whether or not the presence of these bacteria might stabilize phytoplankton productivity of such lakes by enhanced P-assimilation during the formation of polyphosphates. We investigated the vertical distribution of dissolved and particulate nutrients with respect to plankton distribution and light availability in two dimictic lakes. This paper focuses on quantifying the phosphate accumulation in phototrophic sulfur bacteria. The influence of the phototrophic sulfur community on net P-release is discussed with respect to the P-balance of dimictic lakes with anoxic hypolimnia.

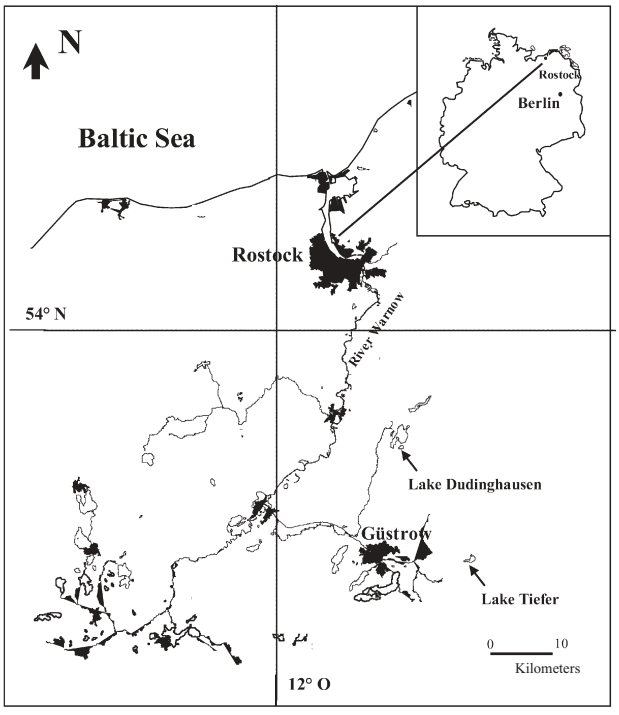

Figure 1. Geographic location of Lake Dudinghausen and Lake Tiefer in northeast Germany.

Table 1. Morphological and limnological characteristics of Lakes Dudinghausen and Tiefer in northeast Germany.

	Lake Tiefer	Lake Dudinghausen
Surface area (ha)	15.9	18.8
Volume (m^3)	1.69 10^6	1.29 10^6
Mean depth (m)	10.7	6.9
Maximum depth (m)	30.6	15.2
Total phosphorus (µg l^{-1})	21–97	32–80
Secchi depth (m)	0.7–2.2	0.6–3.6
Chlorophyll a (µg l^{-1})	2–13	2–6
Trophic classification 1999/2000/2001 (OECD 1982)	**low eutrophic**	**mesotrophic**

Materials and methods

Study sites

Lake Dudinghausen and Lake Tiefer are located in Mecklenburg-Vorpommern (northern Germany), approximately 40 km southeast of Rostock (Fig. 1). Relevant morphometric and water quality parameters are presented in Table 1. Thermal summer stratification starts in both lakes in May and ends in November. Based on measurements in 1999 and 2000, the lakes would be classified as mesotrophic (Lake Dudinghausen) and low eutrophic (Lake Tiefer) according to OECD (1982) criteria.

Sampling and analysis

Seasonal variations in phosphorus concentrations were determined in both lakes during 1998–2001. The vertical profiles were analyzed at the deepest points of the lakes in late summer (August-October 2000–2001). Water temperature, pH, dissolved oxygen, redox potential and turbidity were measured in 1-m increments with a Torphil Model 406 multiprobe (Remember eG, Germany).

Light intensity in the water column was measured with a spectroradiometer (SR-9901, Macam Ltd., Scotland) equipped with a submersible spherical sensor of 0.8 cm diameter. Attenuation spectra [$K_{o(\lambda)}$] were calcu-

lated by linear regression of the ln-transformed underwater spectra taken at 8 depths, as described in Schubert et al. (1995). Subsurface irradiance was calculated by means of continuous global irradiance measurements taken at Zingst (measurement station of the University Rostock) in 2000, using the spreadsheets published by Walsby (1997) for reflection correction. The irradiance at each depth was calculated from the measured attenuation spectra and the subsurface irradiance. Relative chlorophyll *a* (Chl *a*) specific absorption was calculated by means of underwater irradiance spectra and absorbance spectra as shown in Figure 5, assuming a Chl *a* specific absorption coefficient of 0.015 at 675 nm (Haardt and Maske, 1987) for standardization. The relative efficiency of absorption was calculated as the irradiance available at a specified depth divided by Chl *a* specifically absorbed irradiance.

Water samples for analysis of dissolved and suspended nutrients were taken at 0.5, 2, 4, 6, 8, 10, 15, 20, 25 and 29 m depth using a 2.6-L water sampler (Limnos AS, Turkur, Finland). The sampling depths, especially below the thermocline, were chosen according to the results of the multiprobe field measurements (redox potential, turbidity). Samples were stored at 4 °C in the dark until analysis, which was performed within 3–5 h. After membrane filtration (0.45 µm), samples for ammonium determination were fixed immediately; samples for determination of other dissolved nutrients were frozen. Nitrate, nitrite and ammonium were analyzed according to the method of Rohde and Nehring (1979). Soluble reactive phosphorus (SRP) was analyzed by applying the molybdenum blue method in a flow-through system according to Malcom-Lawes and Koon (1990). Total dissolved phosphorus (TDP) was measured as SRP after acidic hydrolysis under UV irradiation, using the molybdenum blue method of Nakamura et al. (1980). Dissolved organic phosphorus (DOP) was defined as the difference between TDP and SRP.

Particulate organic carbon (POC) was analysed with a Vario EL C/N Analyser (Heraeus GmbH, Germany) according to the method for sediment analysis of Verardo et al. (1990), which is also valid for POC in water samples (Schumann et al., 2001). Particulate phosphorus (PP) was determined as the HCl-soluble residues after heating at 500 °C (Andersen, 1976).

The extraction method of Psenner et al. (1984) was used to analyse the different forms of phosphorus in suspended matter as described by Selig et al. (2002). Fractions were defined as follows: H_2O-TP as available phosphorus, BD-SRP as reducible soluble phosphorus, NaOH-SRP as metal and sorptive bound phosphorus, NaOH-NRP as organic-bound phosphorus, and HCl-SRP as carbonate-bound phosphorus.

Phospholipids (Plipid) were extracted in chloroform/methanol as described in Findlay et al. (1989). Determination of polyphosphate (polyP) followed the procedure of Fitzgerald and Nelson (1966). Alkaline phosphatase activity (APA) was measured using 4-methylumbelliferyl phosphate as a fluorogenic substrate as described by Hoppe (1993).

Phytoplankton (including cyanobacteria) were analysed according to Utermöhl (1958). Abundant species were counted to at least 100 units. To determine the biovolume of dominant species, the dimensions of 10 specimens were measured and the volume of the closest geometrical figure was calculated (Edler, 1979). Biovolume was transformed into fresh-weight biomass assuming a mean density of 1 g cm^{-3}. Zooplankton abundance and biomass were determined as described in Heerkloss et al. (1991). Identification of the phototrophic sulfur bacteria species and cyanobacteria, as well as counting and size measurements, were conducted with an Olympus IX 70 fluorescence microscope as described by Rodrigo et al. (1999) and Schumann et al. (2001). For calculation of sulfur bacteria biovolume, all magenta-colored sulfur bacteria in 40 to 60 view fields (total of 6.25 to 9.375 mm^3) of a single sample were counted at 400-fold magnification. Ten specimens of every taxon were measured using 900-fold magnification (60 × 10 × 1.5), and biovolume calculated for the closest geometrical shape. DAPI (4′, 6-diamidino-2-phenylindoldihydrochloride) staining was performed according to Porter and Feig (1980). With this staining, polyphosphate granules fluoresce yellow at 526 nm under UV excitation (Allan and Miller, 1980; Kjelstad et al., 1991). In addition to microscopic analyses, bacteriochlorophyll *a* concentrations (BChl *a*) were used as a second, indirect estimate of phototrophic sulfur bacterial biomass. Pigments were extracted using acetone (24 h, 4 °C in the dark) and analysed by HPLC (Barlow et al., 1997). The system was calibrated using a commercially available BChl *a* standard (Sigma P B5906).

Carbon equivalents of the biomass of plankton organisms were calculated according to Edler (1979 for phytoplankton) and Heerkloss (1996 for zooplankton), whereas the calculations published by Vadstein et al. (1993) were used for calculation of phosphorus equivalents.

Results

Oxygen, inorganic nitrogen and phosphorus

Both lakes were stratified from May to October in the years 1998–2001. In September 2000, the thermocline began at depths of 5 m and 6 m in Lake Tiefer and Lake Dudinghausen, respectively (Fig. 2). The entire hypolimnion was anoxic in both lakes during September. The redox potential decreased with increasing depth in the hypolimnia of both lakes. While the highest turbidity was measured in the upper 4 m of Lake Tiefer, the turbidity in Lake Dudinghausen was higher in the hypolimnion, reaching a maximum at 8 m.

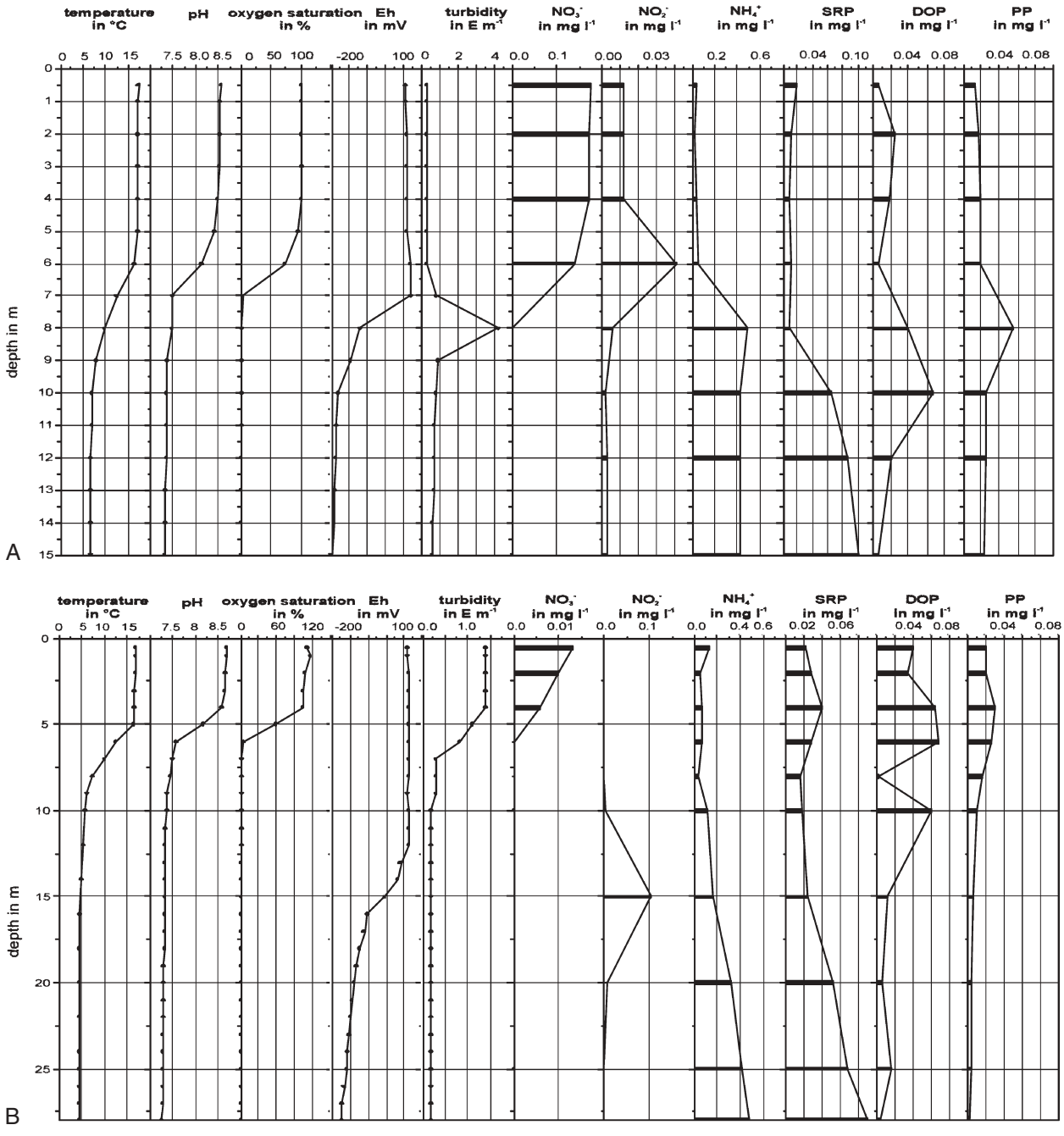

Figure 2. Vertical distribution of temperature, oxygen saturation, pH, Eh, dissolved inorganic nitrogen and P-compounds in Lake Dudinghausen (A) and Lake Tiefer (B) in September 2000.

Nitrate was detected in both lakes, but only in the epilimnion (Fig. 2). Although neither nitrate nor nitrite was found in the hypolimnion of Lake Dudinghausen, high nitrite concentrations were recorded at 15 m depth in Lake Tiefer. The ammonium concentration increased with increasing depth in both lakes. The SRP was also higher at greater depths in the hypolimnia of both lakes under anoxic conditions. In September 2000, the increase had already started at 10 m in Lake Dudinghausen, whereas in Lake Tiefer the SRP did not begin to increase until 15 m. DOP was the dominant P-pool in the epilimnion of Lake Tiefer. In Lake Dudinghausen, high DOP concentrations were recorded in the upper hypolimnion (between 8 and 10 m in September 2000; see Fig. 2), decreasing to low concentrations, comparable to subsurface concentrations, in the bottom layer.

In Lake Tiefer, PP decreased with increasing depth. In the upper hypolimnion, PP concentrations were compara-

ble to those in the epilimnion, but the composition of PP was different (Fig. 3). In Lake Dudinghausen, the PP-concentration at 8 m was twice as high as in the epilimnion. Most of the PP was organic P of this water depth of Lake Dudinghausen (NaOH-NRP, Fig. 3). The sequential P-fractionation reveals a change in the binding forms of PP with changing depth. Considerable changes took place in the BD- and NaOH-extractable P, as both fractions increased with increasing depth in both lakes. In addition to these two chemical P-fractions, the polyP and Plipid concentrations were also higher at 8 m than in the epilimnion in Lake Dudinghausen (Fig. 3).

APA activity, determined in the upper 10 m, decreased dramatically below the thermocline in both lakes (Table 2). A particularly low specific APA per POC was observed at a depth of 8 m in Lake Dudinghausen.

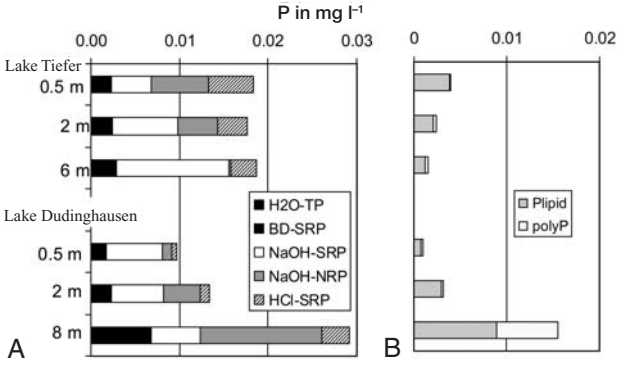

Figure 3. P fractionation (A), polyP and Plipid concentration (B) in different water layers of Lake Dudinghausen and Lake Tiefer in September 2000.

Underwater light climate

Both spectral composition and the intensity of underwater irradiance changed dramatically with increasing depth in both lakes (see Fig. 4 for attenuation spectra). Whereas spectral composition changed almost identically in both lakes, attenuation was much higher in Lake Tiefer. As a result, 1% PAR depth (i.e., the depth to which 1% of the subsurface photosynthetically active radiation can penetrate) in Lake Dudinghausen was located at 8.5 m; the corresponding depth in Lake Tiefer was 5.2 m. Figure 4

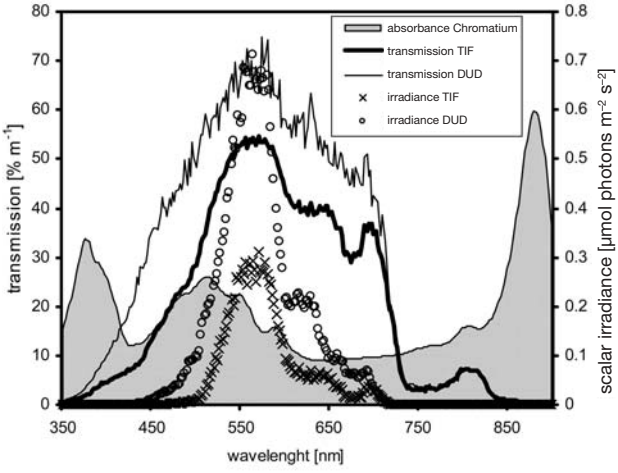

Figure 4. Attenuation spectra, absorbance spectra of *Chromatium* spp., and irradiance spectra in the metalimnia of Lake Dudinghausen and Lake Tiefer (September 2000). Symbols used: thick line – $K_{o(\lambda)}$ Lake Tiefer, thin line – $K_{o(\lambda)}$ Lake Dudinghausen, shaded area – absorbance (relative units) *Chromatium* spp., crosses – irradiance at metalimnion depth (5 m) in Lake Tiefer, open circles – irradiance at metalimnion depth (6 m) in Lake Dudinghausen.

Table 2. Vertical distribution of POC, phytoplankton biovolume (Phy-BV), zooplankton biovolume (Zoo-BV) and C/P equivalent content of plankton in Lakes Dudinghausen and Tiefer in northeast Germany; n.d. – no data.

depth in m	**APA** µmol $h^{-1}l^{-1}$	**POC** mg l^{-1}	**Phy**-biomass mg l^{-1}	**POC-Phy** mg l^{-1}	**PP-Phy** mg l^{-1}	**Zoo**-freshmass mg l^{-1}	**POC-Zoo** mg l^{-1}	**PP-Zoo** mg l^{-1}
Lake Dudinghausen								
0.5	16.0 ±0.9	0.53	3.08	0.377	0.003	1.02	0.077	0.002
2	15.9 ± 1.5	0.91	2.96	0.371	0.003	0.37	0.028	0.001
4	16.2 ± 0.5	0.96	4.36	0.551	0.004	0.03	0.002	0.0001
6	7.6 ± 0.1	0.49	n.d.	n.d.	n.d.	0.83	0.063	0.002
8	5.2 ± 0.2	2.60	n.d.	n.d.	n.d.	0.15	0.012	0.0004
10	3.0 ± 0.3	0.53	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
14	n.d.	0.46	n.d.	n.d.	n.d.	0.02	0.001	0.00004
Lake Tiefer								
0.5	11.3 ± 1.3	2.68	2.40	0.283	0.002	0.01	0.001	0.00003
2	11.7 ± 0.6	2.58	8.73	1.089	0.0085	0.04	0.003	0.0001
4	14.7 ± 3.5	2.45	9.60	1.070	0.0083	0.06	0.005	0.0002
6	9.9 ± 0.4	1.26	1.61	0.177	0.0013	0.03	0.002	0.00008
8	3.8 ± 0.1	0.46	n.d.	n.d.	n.d.	0.01	0.001	0.0001
10	3.4 ± 0.1	0.20	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
20	n.d.	0.63	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
29	n.d.	0.70	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.

compares the absorbance spectra of *Chromatium* spp. (as an example of purple phototrophic sulfur bacteria) with the underwater irradiance spectra in the metalimnia of both lakes. There were no significant spectral differences between the two lakes, but underwater irradiance availability in Lake Tiefer was clearly lower, even though the metalimnion – the uppermost limit for the occurrence of phototrophic sulfur bacteria – was approximately 1 m deeper in Lake Dudinghausen. This effect becomes more obvious when plotted against depth (Fig. 5). The decrease in irradiance availability was much steeper in Lake Tiefer; the 1-m difference in metalimnion depth did not compensate for the difference in attenuation. The same picture emerges if related to absorbed photons (also Fig. 5). The only remarkable difference with respect to spectral composition of the underwater light is that the Chl *a* specific absorbance efficiency of Lake Tiefer was greater than that of Lake Dudinghausen at depths below about 5 m. Using nearside continuous measurements of surface irradiance, doses of available irradiance at metalimnion depths were calculated. The results are as follows: at 5 m (metalimnion), an average dose of 0.18 µmol photons m^{-2} d^{-1} (May-October/Chl *a* concentrations were stable between 4–10 µg l^{-1} in the epilimnion during the summer period) were present in Lake Tiefer, whereas in Lake Dudinghausen 0.47 µmol photons m^{-2} d^{-1} were present at 6 m. At the depth at which anoxic conditions start in Lake Tiefer (15 m), only 0.0002 µmol photons m^{-2} d^{-1} were available, whereas at a similar depth in Lake Dudinghausen (8 m depth in September 2000) 0.19 µmol photons m^{-2} d^{-1} were available. In September 2001, when the phototrophic sulfur bacteria were located at 9 m depth, an average of 0.12 µmol photons m^{-2} d^{-1} was determined.

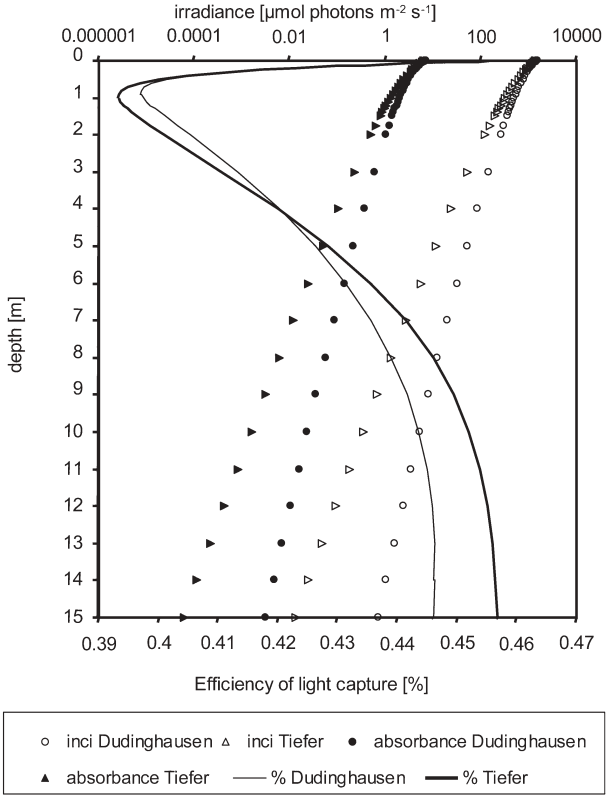

Figure 5. Depth-dependency of incident and Chl *a* specifically absorbed irradiance (also in % of incident irradiance – right Y-axis) of Lake Tiefer and Lake Dudinghausen in September 2000. Symbols used: open circles – incident irradiance in Lake Dudinghausen; filled circles – Chl *a* specifically absorbed irradiance in Lake Dudinghausen; open triangles – incident irradiance in Lake Tiefer; filled triangles – Chl *a* specifically absorbed irradiance in Lake Tiefer; thin line – efficiency of light capture in Lake Dudinghausen; thick line – efficiency of light capture in Lake Tiefer.

POC and plankton analysis

In Lake Tiefer, the POC concentration in the epilimnion was higher than in the hypolimnion (Table 2). In Lake Dudinghausen, the maximum POC concentration was at 8 m (Table 2), the same depth at which PP also peaked (Fig. 2A).

The highest phytoplankton biomass in Lake Tiefer was found between 2 and 4 m. These values were four to five times higher than in the subsurface layer or in the upper part of the hypolimnion (Table 2). In the epilimnion, the phytoplankton contribution to the POC pool varied from 10 to 45%; in the PP pool, the values ranged from 9 to 45%. In Lake Dudinghausen, the phytoplankton contributed 40–70% of the epilimnetic POC pool and 18–25% of the epilimnetic PP pool.

The zooplankton biomass was higher in Lake Dudinghausen. In contrast to Lake Tiefer, the vertical distribution of zooplankton biomass varied strongly in Lake Dudinghausen (Table 2). The percentage of zooplankton in the POC amounted to 1% in Lake Tiefer and varied between 0.5 and 14% in Lake Dudinghausen. The contribution of the zooplankton to the PP pool was generally below 5% in both lakes.

Phototrophic sulfur bacteria were only found in the hypolimnion of Lake Dudinghausen in the late summer period from August to October; however, the highest concentration of phototrophic sulfur bacteria was found at a depth of 8 m in September 2000 and at depths of 9 and 10 m in 2001. The pigment concentration varied from 12 to 36 µg BChl *a* l^{-1}. Microscopic investigations determined the biomass maximum to be 7.6 mm^3 l^{-1}, probably dominated by *Chromatium* spp. (Fig. 6A), in September 2000. DAPI-stained samples exhibited polyphosphate granules in the bacterial cells (Fig. 6B).

Phosphorus balance

Table 3 provides an overview of the distribution of the different phosphorus fractions in both lakes. The water volume ratios between the epilimnion and hypolimnion

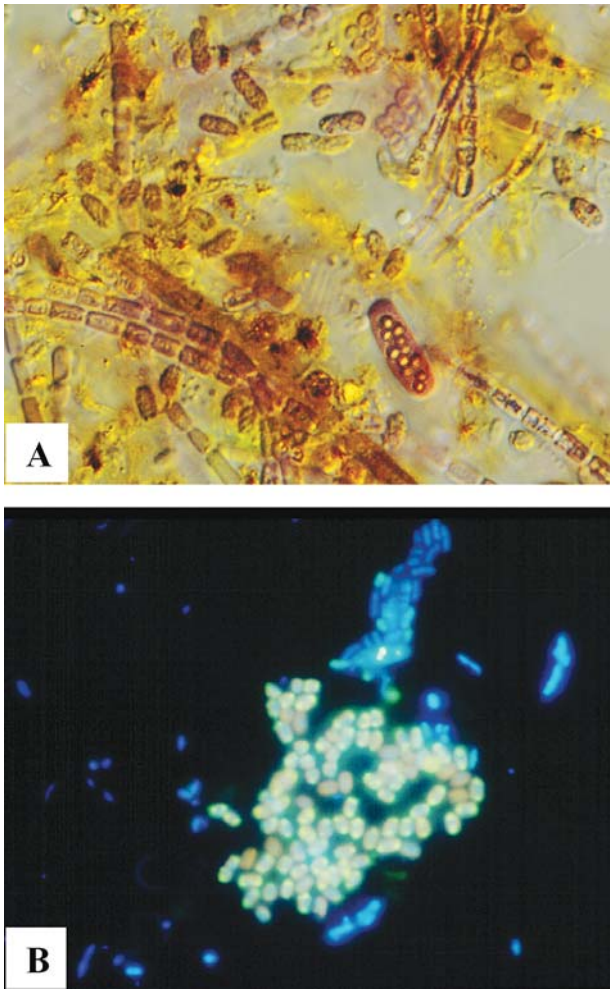

Figure 6. Epifluorescence photomicrograph (A) and a DAPI-stained photomicrograph (B) of a sample from the phototrophic sulfur bacterial layer (8-m water depth from September 2000) in Lake Dudinghausen.

Table 3. Water volumes and content of SRP, DOP and PP in the epilimnion and hypolimnion in Lakes Dudinghausen and Tiefer in northeast Germany.

	Volume in m^3	September 2000		
		PP in kg	DOP in kg	SRP in kg
Lake Dudinghausen	1286874	26.8	26.7	33.7
Epilimnion	826394	13.2	13.2	11.8
Hypolimnion	460479	13.6	13.5	21.9
Sulfur bacteria layer	168290*, 115478**	9.4	6.4	1.7
*2000, **2001				
Lake Tiefer	1692310	56.4	48.1	72.5
Epilimnion	692310	16.4	31.8	20.5
Hypolimnion	1000000	40	16.3	52

are different in Lake Tiefer and Lake Dudinghausen. In Lake Dudinghausen, the amounts of PP and DOP were similar in the epilimnion and hypolimnion, whereas the SRP content was almost twice as high in the hypolimnion, compared to the epilimnion, in 2000. Some 60–70% of the hypolimnetic PP of Lake Dudinghausen was found in the 1-m layer, in which the maximum phototrophic sulfur bacterial concentration occurred. For comparison, only 30–47% of the DOP and 8–20% of the SRP of the whole hypolimnion were found at this depth. In Lake Tiefer, the SRP and PP dominated the phosphorus fractions of the hypolimnion.

Discussion

SRP-release and polyP accumulation in phototrophic sulfur bacteria

SRP was released from the anoxic sediments in both lakes (Selig and Schlungbaum, 2003). In addition, SRP release was limited by redox conditions in the whole hypolimnion of Lake Tiefer. Higher SRP concentrations were only found in water layers lacking nitrite and nitrate. In Lake Dudinghausen, SRP release into the epilimnion was not affected by the presence of nitrate in the hypolimnion. Nonetheless, the increase in SRP stopped at 8 m in Lake Dudinghausen; here, the released SRP was transformed into particulate organic phosphorus by phototrophic sulfur bacteria. At 8–10 m, where the redox potential was less than –190 mV, and thus, detectable concentrations of sulfide might be expected (Kölling, 2000), the quantity and quality of light conditions were sufficient for anoxic phototrophic bacteria to develop. In this layer, phosphorus – transported by sedimentation from the epilimnion and released from the sediment – accumulated as particulate phosphorus. In addition to the PP, the highest polyP and Plipid concentrations were analyzed in the layer in Lake Dudinghausen containing the phototrophic sulfur bacteria. Fluorescence microscopy analysis (DAPI-staining of bacterial samples) confirmed that polyP was stored in the bacteria. The low POC : PP molecular ratio (18 : 1) also indicated significant accumulation of phosphate in reserve granula.

Until now, the importance of polyP storage by bacteria on the P-balance in dimicitc lakes has been described only for heterotrophic bacteria at the sediment-water interface (Davelaar, 1993; Gächter et al., 1988; Hupfer et al., 1995). For example, large amounts of stored polyP were reported under oxic conditions, with their release after a change to anaerobic conditions (Boström, 1988). In contrast to heterotrophic bacteria, phototrophic sulfur bacteria can store polyP under anaerobic conditions as well, when large quantities of SRP are released into dimictic lakes.

The lowest SRP concentrations in the lakes we investigated (0.008 to 0.010 mg P l^{-1}) might not have been low

enough for effective phosphate limitation to occur; however, bacteria are able to induce APA to obtain phosphate from organic molecules under P-depleted conditions. Baneras et al. (1999) described a higher alkaline phosphatase activity in the water layer with phototrophic sulfur bacteria in two small lakes in northern Spain. In the case investigated here, the APA concentration was not elevated in the bacteria layer of Lake Dudinghausen; in fact, APA activity was lower than in the epilimnion. On the other hand, Folt et al. (1989) and Rodrigo et al. (2000) demonstrated that phototrophic sulfur bacteria are able to perform vertical migrations, allowing them to exploit the phosphate pool of the lower hypolimnion under conditions of severe growth limitation; so it is likely that phosphate availability does not limit the growth of the phototrophic sulfur bacterial community in Lake Dudinghausen.

In other P-rich aquatic systems – wastewater treatment plants and the sediment-water interface of eutrophic lakes – accumulation of polyP in microorganisms has been observed after physiological stress (Gächter and Meyer, 1993; Röske and Schönborn, 1994). Feuillade et al. (1995) observed an increase in polyP concentrations in *Chlorella* spp. during recovery after P starvation. If such an observation of stimulation of nutrient storage after temporary limitation may be generalized, the enhanced polyP accumulation could be explained by temporary light limitation. Although a temporary sulfide limitation cannot be ruled out, the measured attenuation spectra measured suggest light limitation was more likely, especially in late summer.

Light conditions and spectral composition of PAR below the thermocline

Purple phototrophic sulfur bacteria like *Chromatium* spp., the dominant taxon in Lake Dudinghausen, strongly absorb the 440 to 580 nm waveband from the PAR (Vila et al., 1996b; see Fig. 5). The small spectral differences in attenuation of both lakes fail to explain the absence of phototrophic sulfur bacteria in Lake Tiefer because the Chl *a* specific absorbance efficiency was even higher below approximately 5 m in this lake; however, the occurrence of phototrophs does not depend on the efficiency of light capture alone, but on the available light energy. Because of higher attenuation, the light intensity in Lake Tiefer was lower at the thermocline than it was in Lake Dudinghausen. Although data about the quantum requirement for zero net growth (E_c) of phototrophic bacteria are scarce, based on the data of Schanz et al. (1998), the E_k seems to be located at around 0.13 mol photons m^{-2} d^{-1}. Assuming that the E_k reflects the average acclimation status of phototrophic organisms (Henley et al., 1993; Sagert and Schubert, 2000), this value could be used as a substitute for the minimum quantum requirement for zero net growth data, if we remember that E_k can be up to 10 times greater than E_c. Accordingly, the data from Lakes Dudinghausen and Tiefer allow the conclusion that phototrophic sulfur bacteria cannot be expected in Lake Tiefer because of light limitation. Already at metalimnion depth, light availability would be very close to the minimum quantum requirement. At the depth where sulfide can be present (redox potential less – 190 mV), the light availability was three orders of magnitude below the assumed minimum value, regardless of the uncertainties caused by the assumptions made for this calculation. It is, therefore, unlikely that positive net growth can occur at such a low energetic level. In contrast, the irradiance values in the phototrophic bacterial layer in Lake Dudinghausen quite closely match the E_k values published by Schanz et al. (1998), supporting the aforementioned hypothesis as well as the assumption that E_k reflects average light availability (Sagert and Schubert, 2000).

P distribution and P balance in the dimictic lakes

A direct comparison between the two lakes with regard to climatic and hydrological conditions can be made if their slightly different epilimnion/hypolimnion quotients are taken into account. In both lakes, regardless of differences in their trophic status, PP was not the dominant P-pool in the epilimnion. While phytoplankton dominated the PP-pool in Lake Tiefer, the contribution of the phytoplankton community to the epilimnion PP pool in Lake Dudinghausen was less than to the POC pool. The percentage contribution of the zooplankton to the PP pool was negligible in both lakes. The presence of DOP, the second P-fraction, in both lakes was somewhat unexpected. This fraction constituted 41 to 45% of the total P in the water column and was quantitatively important in all water layers in both lakes.

The dominant P-fraction in the hypolimnia of both lakes was SRP, regardless of the presence of phototrophic sulfur bacteria in Lake Dudinghausen; however, the phototrophic sulfur bacteria accumulated the dissolved nutrients at the top of the hypolimnion and may thus have acted as a "nutrient filter". Because the phototrophic sulfur bacteria incorporated the SRP released from the sediment under conditions of anoxia, 20–30% of the released SRP was transformed into PP and accumulated in bacterial biomass. Based on this observation, we conclude that the mean accumulation rate of SRP in the hypolimnion was lower in the lake with phototrophic sulfur bacteria (Fig. 7). Approximately 15–20% of the PP in the phototrophic sulfur bacterial layer was identified as storage compounds.

Little is known about polyP metabolism under anoxic conditions in the hypolimnion; therefore, conclusions drawn about the impact of degradation and mineralization processes of the phototrophic sulfur bacterial biomass on the P-balance are speculative at this point in time. A part of this bacterial biomass might be incorporated into

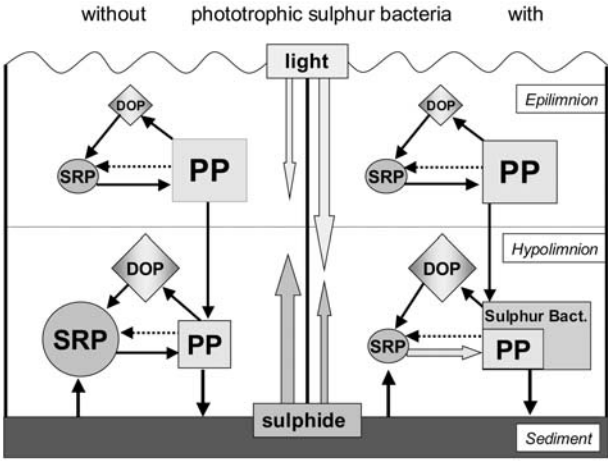

Figure 7. Schematic presentation of the P-balance in stratified, dimictic lakes with and without phototrophic sulfur bacteria.

zooplankton biomass by grazing (Gophen et al., 1974; Overmann et al., 1999). The degradation of phototrophic sulfur bacterial biomass can take place under either anoxic or oxic conditions. When light availability limits the growth of phototrophic sulfur bacteria, biomass may settle out of the water column under anoxic conditions. With the complete mixing of the water column in autumn, sulfide probably becomes the limiting factor for the continuous growth of sulfur bacteria. The hypolimnion, and presumably the sediment-water interface, become oxic, and the bacteria biomass is mineralized. The mineralized P might have been stably bound on the oxic sediment-water interface or released again into the water column. In addition to the mineralization, phototrophic sulfur bacteria may also serve as a carbon source for aerobic heterotrophic bacterioplankton under oxic conditions. For example, Overmann et al. (1996) described the importance of an upwelling of purple sulfur bacteria for heterotrophic bacterioplankton in the meromictic saline Lake Mahoney in British Columbia, Canada.

In Lake Dudinghausen, accumulated P might be quickly released through the mineralization of bacterial biomass, so that the accumulation of P in the phototrophic sulfur bacterial biomass might be only a temporary effect. It seems likely that phototrophic sulfur bacteria in Lake Dudinghausen can limit SRP-transport from the hypolimnion to the epilimnion during late summer. Selig and Schlungbaum (2003) calculated the benthic P-release by different mathematical methods for both lakes. The theoretical molecular diffusion rate from the sediment to the hypolimnion was higher in Lake Dudinghausen than in Lake Tiefer, but the mean accumulation rate of SRP in the hypolimnion was lower. By including PP from the phototrophic sulfur bacterial layer in the calculation, the accumulation rate of P in Lake Dudinghausen was in the same order of magnitude as calculated by Selig and Schlungbaum (2003), suggesting that the occurrence of phototrophic sulfur bacteria might be a factor in decreasing the apparent trophic status of Lake Dudinghausen.

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