Estonian Journal of Ecology, 2013, **62**, 4, 229–243 doi: 10.3176/eco.2013.4.01

Dominant cyanobacterial genera in Lake Peipsi (Estonia/Russia): effect of weather and nutrients in summer months

Reet Laugaste✉, Kristel Panksep, and Marina Haldna

Centre for Limnology, Estonian University of Life Sciences, 61101 Rannu, Tartumaa, Estonia
✉ Corresponding author, reet.laugaste@emu.ee

Received 24 July 2013, revised 7 October 2013, accepted 10 October 2013

Abstract. Hydrochemical and phytoplankton data from L. Peipsi (3555 km^2, mean depth 7.1 m) for July–September 1997–2011 (two lake basins) and for August 2003–2011 (three lake basins) were analysed. Our aim was to explain the impact of nutrient content and weather factors on the composition and species dominance of cyanobacteria. The share of cyanobacteria was on average 64% of the total biomass, maximum values amounted to 93% and 38 g m^{-3} in the areas of open water. Close to the lake shores these values reached 99% and 100 g m^{-3} in some cases. The most prevalent taxa affecting cyanobacterial biomass were *Gloeotrichia echinulata* in the littoral areas and *Microcystis* species in the open water. Principal component analysis placed all dominant genera (*Aphanizomenon*, *Anabaena*, *Gloeotrichia*, and *Microcystis*) separately from each other. Stepwise multiple analysis showed *G. echinulata* to be fairly independent of nutrients and related to the days with water temperature over 22 °C. The biomass of the genus *Microcystis* was found to have evidently the strongest positive connections with phosphorus and also with iron, as well as with all potentially toxic (vacuolated) forms of cyanobacteria. The other group of cyanobacteria, mainly small-celled colonial forms (*Aphanocapsa*, *Aphanothece*, *Cyanodictyon*, etc), constituted on average up to a fourth of the cyanobacterial biomass; its biomass showed a reasonably positive correlation with nitrogen and a negative correlation with water level. In summer, the succession of cyanobacterial genera in the lake started with *Anabaena*, then *Gloeotrichia* appeared (in the larger and deeper moderately eutrophic northern part of the lake), followed by *Microcystis* and *Aphanizomenon*.

Key words: cyanobacteria, succession, nutrients, water level, temperature.

INTRODUCTION

Considerable studies have been devoted to cyanobacterial blooms and to the mechanisms of their dominance in different bodies of water worldwide (Dokulil & Teubner, 2000; Salmaso, 2000; Oliver & Ganf, 2002, among others). Factors promoting blooms are well known and have been analysed thoroughly. However, according to Oliver & Ganf (2002), the distinct morphological, ecological, and physiological characteristics of individual species suggest that it is not possible to distinguish the role or principal factor promoting the dominance of particular species. Besides the nutrient levels and characteristics commonly assessed, many other factors (e.g., mixing regime, CO_2 availability, differences in light requirements and growth rate) influence the emergence of individual species. Tan et al. (2008) connected the domination of cyanobacteria with their preferred germination conditions, and stressed that temperature, resuspension, and bioturbation are driving factors in their recruitment.

Although a trend of dominance of cyanobacteria in the case of a low N to P ratio is evident, it is not possible to establish strict threshold values for it. Havens et al. (2003) noted that the ratios of mineral forms of N to soluble reactive P (DIN : SRP) in Lake Okeechobee (USA) reveal stronger seasonal and year-to-year variability than the total N to total P (TN : TP) ratios. They found the critical ratio of TN to TP for proliferation of cyanobacteria to be 22 and suggested a DIN : SRP of 10 as favourable for the growth of N_2-fixing forms of cyanobacteria. Ni et al. (2012) hypothesized that a combination of high water temperature (>24 °C) and a high TP value (>0.06 mg L^{-1}) together with the regulation of TN : TP ratios (less than 40) enhance the growth of cyanobacteria, in particular *Microcystis*, in the eutrophic Lake Qingshan (southeastern China). According to Scheffer (1998), *Microcystis* blooms are not common in shallow well-mixed lakes (evidently, he considered very shallow *Limnothrix*-type lakes).

The majority of the lakes described in the literature are either very shallow (mean depth about 3 m), or relatively deep and stratified. According to Havens et al. (2003), cyanobacteria blooms in moderately deep, stratified eutrophic lakes are characterized by the dominating Nostocales and, in contrast, shallow lakes are dominated by the family Oscillatoriaceae (*Limnothrix*-type) owing to their ability to use low amounts of light in turbid water. These authors noted that high turbidity, high sharing stress, and low water column stability are co-occurring phenomena in shallow lakes. Our research site, Lake Peipsi, does not fit either group of lakes. Being not stratified but deeper (mean depth 8.3 m in its largest part), its bottom sediments are not so easily accessible to the action of the wind, and water turbidity is not high (mean Secchi value for the ice-free period being 1.8 m in its largest part). Thus, cyanobacteria dominance in L. Peipsi is somewhat different from that observed in deeper stratified as well as very shallow lakes.

The availability of long-term data on cyanobacterial genera/species dominance in L. Peipsi (Estonia/Russia) enabled us to study the conditions preferred by the different genera present. Formerly published data (e.g., Laugaste et al., 2001, 2008) show that the principal genera – *Gloeotrichia*, *Anabaena*, *Aphanizomenon*, and *Microcystis* – can co-exist in the lake. However, commonly one or two of these tend to dominate over the others. According to the data accumulated for the last five decades (Laugaste et al., 2001), a bloom of cyanobacteria occurred in the lake in all years studied, even in cool summers. The most conspicuous forms, visible with the naked eye, were *Gloeotrichia echinulata* (J. S. Smith) P. Richter in L. Peipsi *sensu stricto* (*s.s.*) and bundles of *Aphanizomenon flos-aquae* Ralfs in L. Pihkva (the southernmost part of L. Peipsi *sensu lato* (*s.l.*)). These two algal species dominated either alternately or in different lake parts. Patches of bloom, consisting principally of *Anabaena* and *Microcystis* species, may occur even in the central part of L. Peipsi. Milky when decaying, these patches were particularly prominent in the southern part of L. Peipsi *s.s.*, in L. Lämmijärv, and L. Pihkva in August–September in the 1990s and 2000s. According to Tanner et al. (2005), in Lake Peipsi *s.s.*, the concentration of microcystins at a depth of 30–50 cm was 50 $\mu g\ L^{-1}$ in the open area and up to 1074 $\mu g\ L^{-1}$ in the nearshore area at the beginning of September 2002. Cyanobacterial blooms are more intensive in coastal areas. Thus, the biomass of *G. echinulata* on the surface can rise up to 800 $g\ m^{-3}$, as was the case in the Julys of 1965 and 1980 near the western shore and in 1997 in the northern, usually the least nutrient rich, part of the lake (Laugaste et al., 2001). *Aphanizomenon* has prevailed mainly in August–September; however, a maximum level of biomass was recorded in the late July of 1991: 250 $g\ m^{-3}$ in the pelagial zone of the central part of L. Pihkva. A very intensive bloom was observed in the same location in 1972, when *Gloeotrichia* raised the values of biomass even over 100 $g\ m^{-3}$ in July–August and *Aphanizomenon* did the same in September–October of that year (Laugaste et al., 2001).

In the present study we focused on the dominance of different cyanobacterial genera in summer, with special attention being paid to any possible connections with nutrients and weather conditions. We hypothesized that temperature would be one of the factors promoting recruitment of different genera; the prevalence of heterocystous forms might be linked with nitrogen limitation; domination of the genus *Microcystis* could be linked with the phosphorus content.

MATERIALS AND METHODS

Site description

Lake Peipsi (Peipus) proper or Peipsi *s.l.* is a large (area 3555 km^2) and shallow (mean depth 7.1 m), mainly unstratified lowland water body. Located on the Estonian–Russian border, L. Peipsi is the largest transboundary lake in Europe. Its volume of water is 25 km^3 at long-term mean water levels. The mean residence time of water is two years. Water level fluctuations in the lake are considerable with an average annual range of 1.15 m (Jaani et al., 2008). The catchment area of 47 800 km^2 is shared between Estonia (34%), Russia (58%), and Latvia (8%). Forest and semi-natural areas dominate in the lake drainage basin, agricultural areas cover around 14% of the basin (Jaani, 2001). The lake consists of three basins. The northern part, the largest and deepest, is L. Peipsi *s.s.*, the southernmost part is L. Pihkva (Pskov), which is connected with L. Peipsi *s.s.* by the narrow river-like L. Lämmijärv (Fig. 1). The largest inflows are the Velikaya River in Russia and the Emajõgi River in Estonia. The outflowing Narva River runs into the Gulf of Finland, part of the Baltic Sea. It should be also noted that the town of Narva draws drinking water from this river. The duration of ice cover is variable, from 2 weeks to 6 months. The water is the warmest (21–22 °C in open water on average) in July–August. The lake is well mixed by the wind and well aerated by

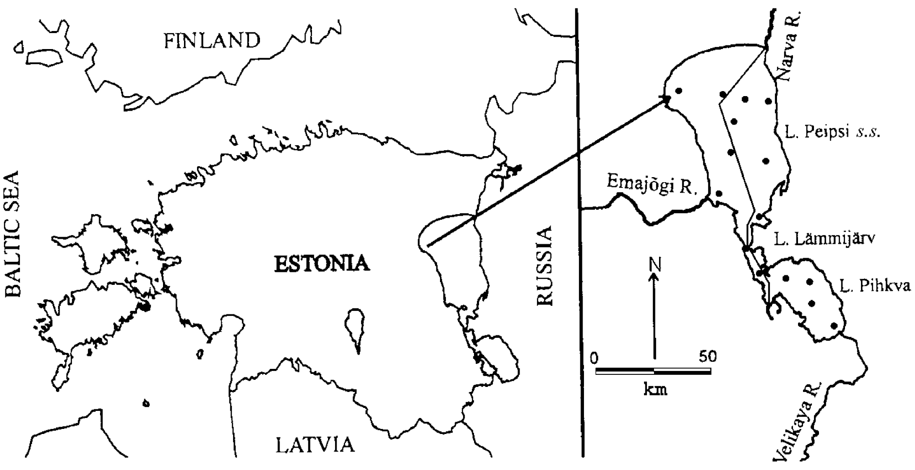

Fig. 1. Location of Lake Peipsi with sample sites. Peipsi *s.s.* – Peipsi *sensu stricto*.

waves and currents. There is no permanent stratification of temperature, oxygen content, or hydrochemical parameters in the ice-free period. Sometimes transient oxygen deficit occurs on the bottom of the lake. The water is alkaline, the mean pH in the ice-free period is 8.36 (Milius & Haldna, 2008). The main pollution source is the Velikaya River,which flows from the south into L. Pihkva (Haberman et al., 2010). The water volume of L. Pihkva is eight times less than that of L. Peipsi *s.s.* (Table 1). On the basis of the OECD (1982) classification, L. Peipsi *s.s.* is considered as an eutrophic water body, while L. Pihkva is hypertrophic at present. The increasing difference in total phosphorus (TP) concentrations between the northern and southern parts of the lake clearly shows that the input of P from the south is increasing (Kangur & Möls, 2008). A continuous and even accelerating deterioration of the quality of the lake water has occurred up to the present (Kangur & Möls, 2008).

Table 1. Some characteristics of different parts of L. Peipsi proper (Jaani, 2001). Asterisks (*) denote the geometrical mean values for August 2003–2011

Parameter	L. Peipsi *s.s.*	L. Lämmijärv	L. Pihkva
Area, km^2	2611	236	708
Mean depth, m	8.3	2.5	3.8
Maximum depth, m	12.9	15.3	5.3
Volume, km^3	21.79	0.60	2.68
Secchi depth, m*	1.6	0.8	0.6
TP, mg m^{-3}*	46	87	121
TN, mg m^{-3}*	657	1039	1136
Chl *a*, mg m^{-3}*	20.7	54.4	64.8

Sampling and processing

The material for 1997–2011 (June–September, once a month) was collected from six sampling sites only in L. Peipsi *s.s.* and L. Lämmijärv. The data for the whole lake (15 sampling sites) including L. Pihkva, located almost entirely in Russia, were available for August 2003–2011 (Fig. 1).

Well-mixed samples for phytoplankton analysis were obtained in parallel with hydrochemical data, by stirring the water in a large vessel. The water was collected at 1 m intervals through the entire water column using a two-litre Van Dorn sampler. Samples were preserved with Lugol's (acidified iodine) solution and processed applying the Utermöhl (1958) method. Hydrochemical samples (parameters in Table 2) were analysed in the Tartu Branch of the Estonian Environmental Research Centre. The data for water temperatures and water levels were obtained from the Institute of Meteorology and Hydrology at the Estonian Ministry of

Table 2. Analysed main parameters, data from July to September 1997–2011. Abbreviations: Phyto – phytoplankton; bm – biomass; Cy – cyanobacteria; Temp – temperature, everyday average for the warmest month, July, 1997–2011; COND – conductivity; DIN – sum of mineral forms of N (NO_2, NO_3, NH_4); Water level, everyday average for May–August, 1997–2011, 200 cm is equal to 30 m a.s.l.

Variable	Unit	*N*	Mean	SD	Minimum	Maximum
Phyto bm	G m^{-3}	280	12.23	8.68	1.42	47.69
Cy % in bm	%	280	60.84	18.39	5.25	97.27
Cy bm	g m^{-3}	280	7.9	7.16	0.56	38.13
Gloeotrichia bm	g m^{-3}	280	0.62	1.93	0	20.99
Anabaena bm	g m^{-3}	280	0.55	1.55	0	12.79
Aphanizomenon bm	g m^{-3}	280	0.86	1.76	0	14.12
Microcystis bm	g m^{-3}	280	2.85	5.17	0	35.47
Cy others bm	g m^{-3}	280	2.02	2.49	0	9.77
Temp	°C	434	20.4	2.1	16.3	25.9
pH		280	8.5	0.3	7.1	9.4
COND	µS cm^{-2}	280	269	28	104	394
HCO_3	meq L^{-1}	280	2.5	0.26	0.97	3.9
COD_{Cr}	mgO L^{1}	280	35.05	9.75	16	62
NH_4N	µg L^{-1}	280	34.27	31.25	5	350
DIN	µg L^{-1}	280	104	89	33	1203
PO_4P	µg L^{-1}	280	15.04	13.79	2	110
DIN : PO_4P		280	14.8	16.0	1.0	109.4
TN	µg L^{-1}	280	786	297	410	2100
TP	µg L^{-1}	280	65.8	42.2	15	360
TN : TP		280	14.45	7.07	1.94	58.67
SI	mg L^{-1}	247	1.32	1.05	0.1	6
FE	mg L^{-1}	251	0.24	0.26	0.02	2.3
CL	meq L^{-1}	280	0.18	0.03	0.04	0.28
SO_4	meq L^{-1}	280	0.26	0.06	0.06	0.37
Water level	cm	1722	209	37	108	304

Environment. The water levels for the months from May to August were analysed separately. The sum of the water temperatures taken daily for June and July, the sum of temperatures over 20 °C in these months, and the number of days with water temperature over 22 °C were also included. Besides the whole lake measurements, its individual basins were also analysed, particularly since *Gloeotrichia* and *Anabaena* preferred the northern eutrophic part, L. Peipsi *s.s.*, whereas *Aphanizomenon* and *Microcystis* favoured the southern hypertrophic part, L. Pihkva. It was found that as a result of this separation of data, the hydrochemical parameters became closer to normal distributions. The dominant four genera were analysed separately.

Log-transformed values for hydrochemical parameters and square root values for phytoplankton parameters were used for statistical analyses. To find out which chemical and weather factors had a statistically significant effect, the stepwise multiple regression model was used (significance level 0.001). For grouping cyanobacteria genera and the most effective environmental factors two first principal components of PCA were used. Spearman correlation analysis was applied to calculate the relationship between variables. Statistical analyses were run in R 2.14.0 (R Development Core Team, 2011).

RESULTS

As the results for the two southern lake basins, L. Lämmijärv and L. Pihkva, are comparable and markedly different from the northern basin, L. Peipsi *s.s.* (Table 1), hereafter they will be grouped together under the name L. Pihkva. In addition to the N_2-fixing heterocystous genera (*Gloeotrichia*, *Anabaena*, *Aphanizomenon*), a category consisting of potentially toxic cyanobacteria (all these three genera plus *Microcystis*) and another group of residual (others) small-celled and/or thin filamentous cyanobacteria (*Aphanocapsa*, *Aphanothece*, *Cyanodictyon*, *Radiocystis*, *Limnothrix*, etc.) were made. These three categories were analysed as individual groupings (Fig. 2). Some well-known potentially toxic cyanobacteria such as *Planktothrix agardhii* (Gomont) Anagnostidis & Komárek and *Woronichinia naegeliana* (Unger) Elenkin found intermittently in L. Peipsi (Laugaste et al., 1996) were not taken into account. It should be noted that the genus *Gloeotrichia* consisted of only one planktonic species in the lake (*G. echinulata*), while the genus *Aphanizomenon* was represented mainly by *A. flos-aquae* and small quantities *A. issatchenkoi* (Ussaczew) Proshk.-Lavr., *A. skujae* Komárk.-Legn. & Cronberg, and *A. gracile* Lemm. Two important species were present from the genus *Microcystis*: *M. viridis* (A. Braun) Lemm. and *M. wesenbergii* (Komárek) Komárek, while other *Microcystis* species (*M. botrys* Teiling, *M. aeruginosa* Kütz., *M. flos-aquae* (Wittr.) Kirchner etc.) occurred in smaller quantities. Approximately 10 *Anabaena* species were identified in the fresh material; however, establishing the dominant species when counting was difficult, as frequently only pieces of filaments without akinetes and heterocysts were found in the counting chamber. *Anabaena flos-aquae* G. S. West was the most common dominant in the lake

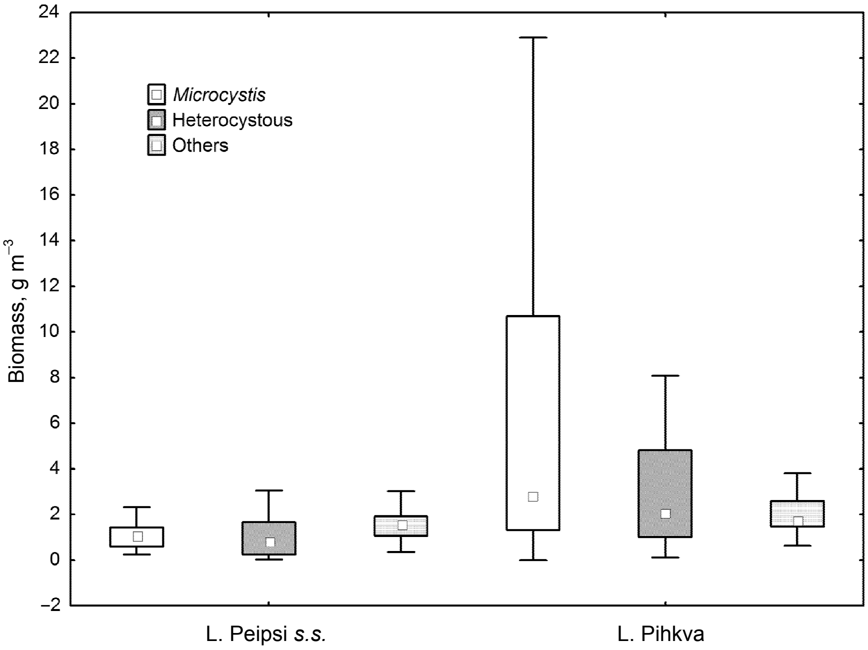

Fig. 2. Biomass of heterocystous forms, *Microcystis*, and the residual group (Others) of cyanobacteria (median, quartiles, and min–max range) in different parts of Lake Peipsi proper for August.

noted in earlier works (Laugaste et al., 2001), but in addition to this, species such as *A. circinalis* Rabenh., *A. perturbata* Hill, *A. curva* Hill, and *A. fusca* Hill have been identified more recently, and may even dominate. In some cases, *A. crassa* (Lemm.) Komark.-Legn. & Cronberg and *A. lemmermannii* P. Richter were also found to be among the dominants in the northern lake basin, L. Peipsi *s.s.*, while *A. compacta* (Nyg.) Hickel was frequently found (but was not dominant) in the southern parts. Patchiness of the distributions seems to be obvious, especially in the case of *Anabaena* and also *Gloeotrichia*, which occurred in samples only sporadically and was not found in open water at all in some years. At the same time, these cyanobacteria were abundant at some sites in the coastal region.

Principal component analysis placed all four genera (*Gloeotrichia*, *Aphanizomenon*, *Microcystis*, *Anabaena*) separately from each other (Fig. 3). Two main groupings of components explained 63% of the variance found. *Microcystis*, TP, and Fe formed a quite close group; *Aphanizomenon*, residual algae (others), and TN the other somewhat diffusive group, where *Anabaena* was linked with temperature and *Gloeotrichia* with water level and with temperatures over 22 °C. According to stepwise multiple analysis, the most distinguishable cyanobacterium in L. Peipsi, *Gloeotrichia echinulata*, is visible by the naked eye during two months each year, commonly from the end of June to mid-August, and it seems to be fairly independent of nutrients. The principal factor affecting all cyanobacterial genera taken together was temperature (33% of the variance), followed by nutrients (27%) and water

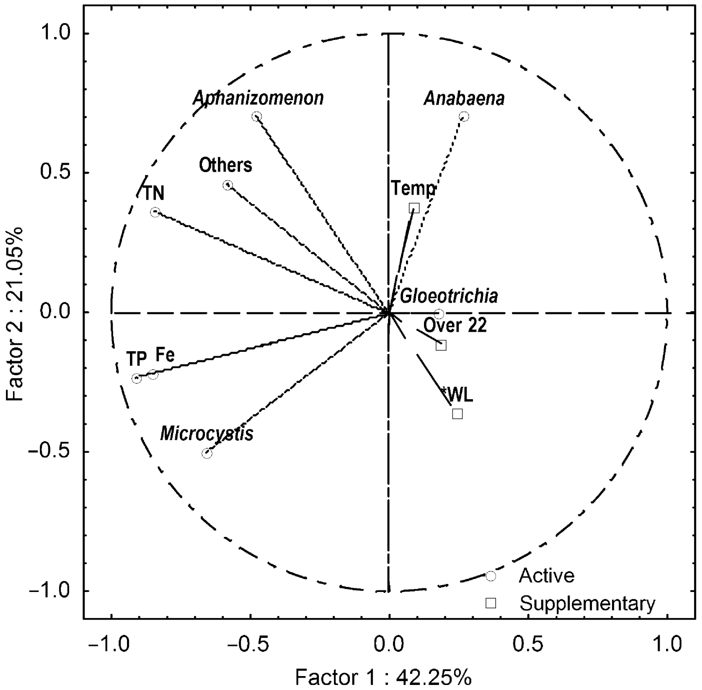

Fig. 3. Grouping of selected variables in PCA analysis. Abbreviations: TN – total nitrogen; TP – total phosphorus; WL – average water level for June, July, August; Temp – average of water temperatures in July–August; Over 22 – number of days over 22 °C.

level (15%). Factor 1 (temperature) was positively correlated with *Microcystis* ($p < 0.001$); factor 2 (nutrients) was positively correlated with *Microcystis*, *Aphanizomenon*, and the residual group ($p < 0.001$). *Anabaena* was negatively correlated with water level (factor 3, $p = 0.023$); *Aphanizomenon* was negatively correlated with temperature and water level ($p < 0.001$); *Microcystis* and the residual group were negatively correlated with water level ($p = 0.02$ and $p < 0.001$, respectively). Negative connections between *Aphanizomenon* and water temperature reflect the domination of *Aphanizomenon* at the end of summer and in autumn at lower temperatures (Fig. 4). Spearman correlation analysis displayed that most relationships concerning the heterocystous forms were influenced by *Aphanizomenon* ($r = 0.95$, $p < 0.001$), and those of all potentially toxic cyanobacteria by *Microcystis* ($r = 0.88$, $p < 0.001$). *Anabaena* had positive correlations with TN : TP ratios ($r = 0.39$, $p < 0.001$) and behaved in an opposite manner to the other bloom-forming genus *Microcystis*. At the same time, *Anabaena* and *Microcystis* were found to be present in the plankton mass together with all bloom-forming genera, and in some cases they were subdominants. Correlations between cyanobacterial genera and mineral forms of nutrients (DIN, PO_4, DIN : PO_4) were lacking, except for the correlation of *Aphanizomenon* and residual cyanobacteria with DIN. *Microcystis* and potentially toxic cyanobacteria were associated

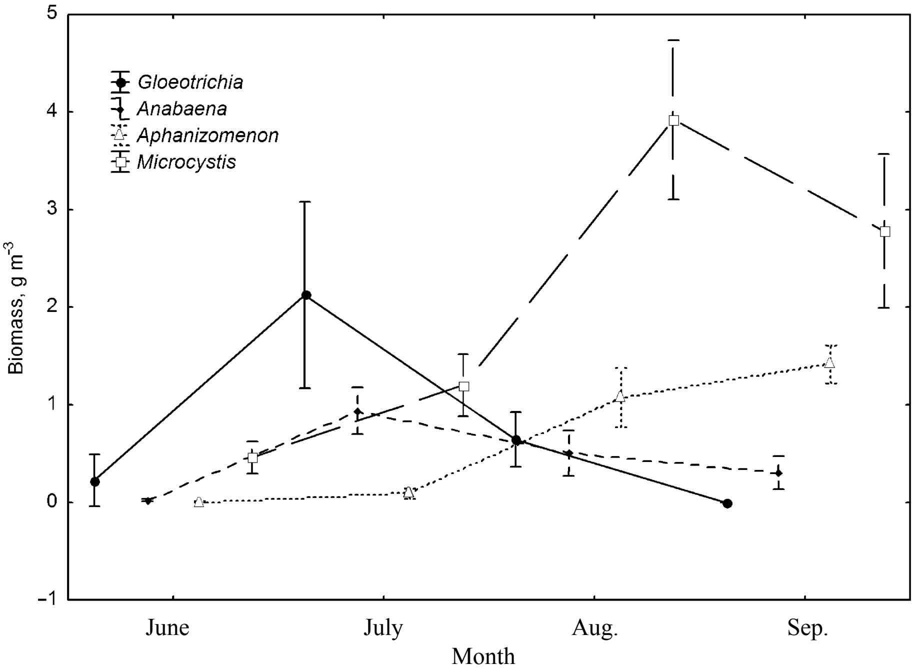

Fig. 4. Biomass of cyanobacterial genera (mean ± 0.95 conf. interval) in the whole lake in summer and early autumn.

with iron ($r = 0.67$ and 0.71, respectively, $p < 0.001$). The residual group showed a reasonably positive correlation with DIN and TN and with the ratio of mineral forms of N and P and a negative correlation with water level. This group links with other ions (Cl^-, SO_4^{2-}, K^+, Na^+, Si^{2-}; $r = -0.34$ to -0.42, $p < 0.001$). At the same time, quite remarkable negative correlations occurred in both lake parts between water level and the above-mentioned constituents ($r = -0.44$ to -0.58, $p < 0.001$), and also between the residual group and water levels ($r = -0.53$, $p < 0.001$). Evidently, any correlation of the residual cyanobacterial group with the measured constituents is mediated principally by water level. As to nutrients, TN and TP were weakly correlated with water level in all studied months ($r = -0.26$ to -0.45, p 0.02–0.006).

According to the data for 15 years, the main cyanobacterial genera appeared in the open water on the following days of the year: *Anabaena* 167th day (mid-June), *Microcystis* 176th day, *Aphanizomenon* 177th day, and *Gloeotrichia* 180th day (end of June). *Anabaena* and *Gloeotrichia* appeared earlier in the northern lake part, while *Microcystis* and *Aphanizomenon* were observed earlier in the southern part. However, *Anabaena*, *Microcystis*, and *Aphanizomenon* occurred among the dominants in both lake parts. The biomass of *Gloeotrichia* on the sampling day was positively affected most of all by water temperatures ten days prior to sampling, and the biomass of *Anabaena* and *Microcystis* was positively affected by water temperatures 30 days prior to sampling ($p < 0.0001$). At the same time, the biomass of *Aphanizomenon* was negatively affected by water temperatures during the whole August ($p = 0.004$–0.0009) but not by those in earlier months.

DISCUSSION

According to the long-term data, the effects of water level, as well as the mechanical influence of wind and waves, are stronger in the shallower part, L. Pihkva, due to its 8-fold smaller volume than that of L. Peipsi *s.s.* (Milius et al., 2005). *Gloeotrichia echinulata* is common in well-mixed mesotrophic and eutrophic lakes found at temperate latitudes (Karlsson-Elfgren et al., 2003), and was recently found blooming in oligo- to mesotrophic lakes throughout the northeastern USA (Carey & Rengefors, 2010). It was found to inhabit mainly the moderately eutrophic northern basin of L. Peipsi, appearing into the littoral water along with *Anabaena*, however, later than other genera into the open water. This can be explained by the slower warming of the larger and deeper northern part of the lake, L. Peipsi *s.s.*, where *Gloeotrichia* dominates in midsummer. Among four studied genera it had also the strongest connections with temperature and was not related with nutrients. This may possibly be due to two reasons: firstly, the colonies were found only by chance in the open water due to their very irregular (patchy) distribution; secondly, its ability to assimilate large amounts of P from nutrient-rich sediments, above its immediate needs, and thus the colony can store P for subsequent growth. Thus, germinating colonies can sustain substantial growth even when nutrient supplies in water are low (Karlsson-Elfgren et al., 2004). Besides, the germination of this alga is light-dependent, and recruitment from shallow sediments forms the important seed for the pelagic population in Lake Erken (Karlsson-Elfgren et al., 2003). In L. Peipsi, where transparency is low (not more than 2–3 m in the spring clear-water period in the northern part), germination can take place in some shallow areas, and colonies would be carried into open water by currents. The structure of the currents is complicated, depending on the shape of the lake basin and the effects of prevalent winds (Jaani et al., 2008). Thus, the colonies can distribute in random and incalculable ways, being sometimes completely lacking in fixed monitoring sites. As a result, the observed relative importance of *G. echinulata* in the lake may be far less than the reality.

It is interesting that the genus *Anabaena*, as well as the N_2-fixing group, was found to have quite strong positive correlations with the TN : TP ratio and weak correlations with the nutrient forms. According to Reynolds (2006), for N_2 fixers nitrogen fixation response is a preferential reaction to the concentration of NH_4-N <0.5 µM N (<7 mg N m^{-3}). In August these values were on average 32 mg N m^{-3} in L. Pihkva and up to 49 mg N m^{-3} in L. Peipsi *s.s.* Reynolds stresses also that potential N_2 fixers can increase to significant levels without producing any heterocysts, which are produced abundantly by nitrogen limitation. Thus, the occurrence of significant numbers of *Anabaena* filaments without heterocysts in L. Peipsi may indicate that there was no nitrogen limitation in most cases. Nitrogen fixation was measured in the lake in July to September 2005 and July to August 2006 (Laugaste et al., 2008). From 18 determinations, significant N_2 fixation was found in four sites in July 2005 (NH_4-N 22–87 mg m^{-3}, TN : TP 12–27; dominated by *G. echinulata*, *Anabaena lemmermannii*, and in southern parts, *Microcystis* spp.), and at one site in July 2006 (NH_4-N 33 mg m^{-3}, TN : TP 11; dominated by *Microcystis* spp. and in small quantities, *Aphanizomenon flos-aquae*). In August of 2003–2010, N_2 fixers achieved maximum biomass at a TN : TP mass ratio between 8 and 19, while *Microcystis* (not fixing N_2) occurred at a ratio below 10 in the lake. Ferber et al. (2004) stressed that *Anabaena* and *Aphanizomenon* can compensate for their N demand by vertical migrations into the hypolimnion or the sediments, thus avoiding the need to fix nitrogen (low DIN hypothesis). Thus, connections of heterocystous cyanobacteria with different forms of N in the water as well as with the TN : TP ratio are quite weak or lacking altogether. Ferber et al. (2004) suggested that the low DIN hypothesis is valid for both heterocystous and nonheterocystous vacuolated cyanobacteria. Consequently, we conclude that the domination of nitrogen-fixing cyanobacteria is far from being associated with real N fixing in the lake nor is it associated with the content of nitrogen in the water.

The response of N_2 fixers as well as *Microcystis* to the TN : TP ratio in L. Peipsi contradicted numerous data found in the literature: in our lake the biomass of heterocystous forms and their percentage in the cyanobacterial biomass increased with the TN : TP ratio, while the biomass of *Microcystis* fell (Fig. 5). Downing et al.

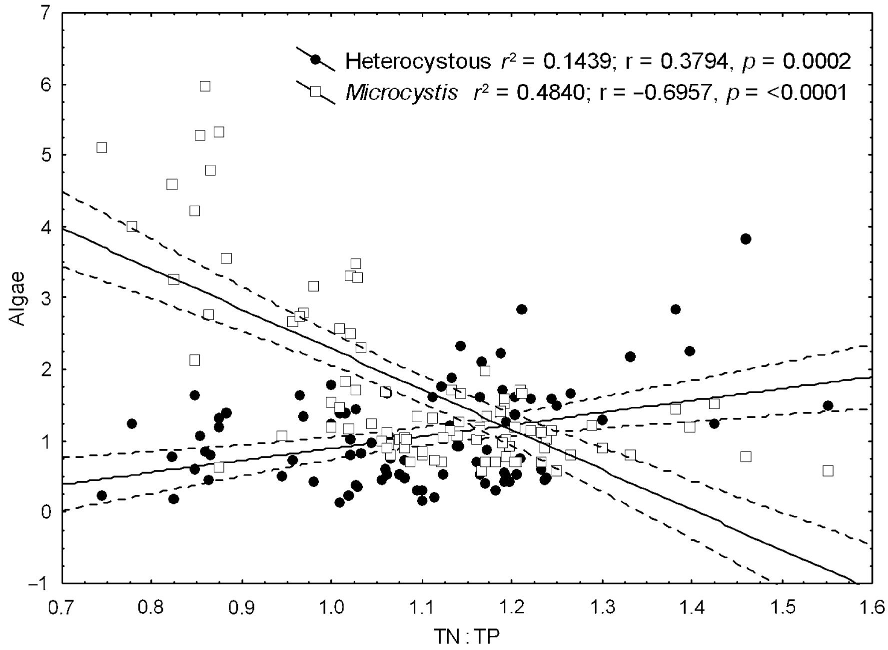

Fig. 5. Relationship between square root of heterocystous cyanobacteria and *Microcystis* biomass and the logarithmic TN : TP ratio in Lake Peipsi proper.

(2001) analysed 269 observations collected from 99 lakes around the world. These authors found the TN : TP ratio to be the poorest predictor of cyanobacterial dominance, while the probability for total P concentration to predict dominance is 30% higher. They noted that average summer P concentrations above 70 µg L^{-1} provide an 80% guarantee for cyanobacterial dominance. This threshold is appropriate for L. Pihkva but is too high for L. Peipsi *s.s.* Although such values are rare for the latter lake, cyanobacteria prevail in the summer months in all years. According to Nõges et al. (2008), the biomasses of all cyanobacteria and N_2-fixing species, as well as the proportion of cyanobacteria and N_2-fixing species found, achieved maximum values at a TN : TP mass ratio at or below 30 (for both total and mineral forms) in L. Peipsi. In our opinion, this threshold is not very strict, particularly for L. Peipsi *s.s.* The percentage of cyanophytes in the phytoplankton biomass had some correlations with nutrient concentrations in the northern part of the lake but not in the southern part L. Pihkva, evidently because that basin has permanent sufficiency of nutrients. Wang et al. (2008) could not identify a limiting nutrient with respect to TN : TP ratios in Chinese shallow lakes and fish ponds, and these authors suggested that total P is the primary factor regulating phytoplankton. Also some other authors have noted the importance of P and not the TN : TP ratio in cyanobacterial dominance (Trimbee & Prepas, 1987; Scheffer et al., 1997; Vrede et al., 2009; Arvola et al., 2011).

Reynolds (2006) pointed out that in experiments mineral N forms, particularly NH_4-N, are important for N_2 fixers and also for *Microcystis* growth. According to our data, *Anabaena*, *Microcystis*, and the residual group all had negative correlations with ammonium; N_2 fixers were positively correlated with TN and *Microcystis* with TP. Reynolds (2006) also noted that *Microcystis* is among the species that have a faster uptake capacity at a low P content, thus it has a greater affinity for P and a greater ability to compete for scarce resources. However, in L. Peipsi, long-term data indicate a phosphorus peak for August (Milius & Haldna, 2008) when *Microcystis* was seen to prevail. Marinho & Huszar (2002) noted a *M. aeruginosa* bloom in a tropical reservoir when DIN concentrations were <5 µM and TN : TP ratios below 10. In warm summers in Müggelsee in Germany, a bloom of *Aphanizomenon flos-aquae* is accompanied by the mass development of several *Microcystis* species (Teubner et al., 1999). Such a pattern of dominants is quite common also in L. Peipsi, particularly in warm weather.

The mineral iron showed moderate negative correlations with heterocystous forms and positive ones with *Microcystis* biomass in Lake Peipsi. Some authors (Ou et al., 2006; Li et al., 2009) have stressed the importance of iron for the growth of *M. aeruginosa*. Reynolds (2006) found the requirement of active N_2 fixers for iron to be relatively greater. Ou et al. (2006) found in laboratory conditions that the *Microcystis* growth kinetics with respect to changes in P and Fe reflect its competitive advantage in the natural environment. According to Tan et al. (2008), *Anabaena* and *Aphanizomenon* appear in plankton in late May–early June, *Gloeotrichia* from middle June to late July, *Microcystis* in late July, peaking in August in different lakes of middle latitudes. Unfortunately, it is complicated to attempt to compare these data for different lakes with our study on one lake. In the shallow and medium-sized Shelburne Pond (USA), *Aphanizomenon flos-aquae* dominates in June–July, *Microcystis* species and *Planktothrix* dominate in July–August, and *Anabaena* occurs throughout the vegetation period with a maximum in autumn (Ferber et al., 2004). In Lake Balaton, *Aphanizomenon flos-aquae* appears and becomes dominant in the period characterized by low temperatures and high light intensities (late spring and early summer), and is followed by other heterocystous species such as *Anabaena* spp. (Kovács et al., 2012). In two Japanese lakes, the growth of *Aphanizomenon flos-aquae*, which usually blooms in summer but can also tolerate low temperatures in winter, is promoted at high temperatures (Yamamoto, 2009). The author also noted that nutrient concentrations and pH has no evident impact on *A. flos-aquae*, and further expansion of this cyanobacterium will be determined by the eutrophication process and by the absence of other cyanoprocaryotes. Such a pattern could be applied to L. Peipsi as well.

In summer months, the maximum biomass values of genera in L. Peipsi occurred in the following order: *Anabaena* + *Gloeotrichia*, *Microcystis*, *Aphanizomenon*. According to Laugaste et al. (2008), the dominance of the last genus continues up to November in some warm autumns. Evidently, unlike *Anabaena* filaments, the bundles of *Aphanizomenon* are rather resistant to waves. Jensen et al. (1994) found that in shallow lakes, *Anabaena* and *Aphanizomenon* prevail in early summer along with abundant N content, while *Microcystis* dominates at the end of summer. According to literature data, the period of the domination of *Aphanizomenon* appears to be extremely variable in different lakes.

It would be quite confusing to consider all these statements in the literature when analysing the succession of different algal genera in L. Peipsi. Jensen et al. (1994) stressed that in reality, the growth of organisms requires a complex of conditions, while we like simple explanations. As Oliver & Ganf (2002) noted, the occurrence and abundance of various types of gas-vacuolated cyanobacteria is not reliant on any particular environmental stimulus, but depends on a complex interplay of factors.

In conclusion, the succession of cyanobacterial genera in L. Peipsi started with *Anabaena*, then *Gloeotrichia* appeared (in the larger and deeper eutrophic northern lake part), followed by *Microcystis* and *Aphanizomenon*. The domination by *G. echinulata* was most affected by high water temperatures. Frequent occurrences of *Anabaena* species without heterocysts indicates the abundance of nitrogen in the lake. It seems also that *Anabaena* filaments are most susceptible to wave action. The connections of most heterocystous forms with nitrogen were weak. The biomass of the genus *Microcystis* was found to have evidently the strongest positive connections with phosphorus and also with iron, as well as with all potentially toxic (vacuolated) forms of cyanobacteria. *Aphanizomenon flos-aquae* was most frequent at the end of summer and in early autumn, while any connections between its presence and nutrients remained unclear. Further research is needed to establish whether altered P loading has brought about any change in the biomass or dominant species of cyanobacteria over a longer time period.

ACKNOWLEDGEMENTS

The research was supported through targeted financing by the Estonian Ministry of Education and Research (project SF 0170006s08) and the Estonian Science Foundation (grant 7643). Hydrobiological collections from the Centre for Limnology and data from the Estonian State Monitoring Programme were used. We are indebted to Dr Renee Miller (UK) for revising the English text of this paper. The contribution of the referees is highly appreciated.

REFERENCES

Arvola, L., Järvinen, M., & Tulonen, T. 2011. Long-term trends and regional differences of phytoplankton in large Finnish lakes. *Hydrobiologia*, **660**, 125–134.

Carey, C. C. & Rengefors, K. 2010. The cyanobacterium *Gloeotrichia echinulata* stimulates the growth of other phytoplankton. *Journal of Plankton Research*, **32**, 1349–1354.

Dokulil, M. T. & Teubner, K. 2000. Cyanobacterial dominance in lakes. *Hydrobiologia*, **468**, 1–12.

Downing, J. A., Watson, S. B., & McCauley, E. 2001. Predicting cyanobacteria dominance in lakes. *Canadian Journal of Fisheries and Aquatic Sciences*, **58**, 1905–1908.

Ferber, L. R., Levine, S. N., Lini, A., & Livingston, G. P. 2004. Do cyanobacteria dominate in eutrophic lakes because they fix atmospheric nitrogen? *Freshwater Biology*, **49**, 690–708.

Haberman, J., Haldna, M., Laugaste, R., & Blank, K. 2010. Recent changes in large and shallow Lake Peipsi (Estonia/Russia): causes and consequences. *Polish Journal of Ecology*, **58**, 645–662.

Havens, K. E., James, R. T., East, T. L., & Smith, V. H. 2003. N : P ratios, light limitation, and cyanobacterial dominance in a subtropical lake impacted by non-point source nutrient pollution. *Environmental Pollution*, **122**, 379–390.

Jaani, A. 2001. The location, size and general characterization of Lake Peipsi and its catchment area. In *Lake Peipsi: Meteorology, Hydrology, Hydrochemistry* (Nõges, T., ed.), pp. 10–17. Sulemees Publishers, Tartu.

Jaani, A., Klaus, L., Pärn, O., Raudsepp, U., Zadonskaja, O., Gronskaja, T., & Solntsev, V. 2008. Hüdroloogia. In *Peipsi* (Haberman, J., Timm, T., & Raukas, A., eds), pp. 113–155. Eesti Loodusfoto, Tartu.

Jensen, J. E., Jeppesen, E., Olrik, K., & Kristensen, P. 1994. Impacts of nutrients and physical factors on the shift from cyanobacterial to chlorophyte dominance in shallow lakes. *Canadian Journal of Fisheries and Aquatic Sciences*, **51**, 1692–1699.

Kangur, K. & Möls, T. 2008. Changes in spatial distribution of phosphorus and nitrogen in the large north-temperate lowland Lake Peipsi (Estonia/Russia). *Hydrobiologia*, **599**, 31–39.

Karlsson-Elfgren, I., Ryding, E., Hyenstrand, P., & Pettersson, K. 2003. Recruitment and pelagic growth of *Gloeotrichia echinulata* (Cyanophyceae) in Lake Erken. *Journal of Phycology*, **39**, 1050–1056.

Karlsson-Elfgren, I., Rengefors, K., & Gustafsson, S. 2004. Factors regulating recruitment from the sediment to the water column in the bloom-forming cyanobacterium *Gloeotrichia echinulata*. *Freshwater Biology*, **49**, 265–273.

Kovács, A. W., Tóth, V. R., & Vörös, L. 2012. Light-dependent germination and subsequent proliferation of N_2-fixing cyanobacteria in a large shallow lake. *International Journal of Limnology*, **48**, 177–185.

Laugaste, R., Jastremskij, V., & Ott, I. 1996. Phytoplankton of Lake Peipsi-Pihkva: species composition, biomass and seasonal dynamics. *Hydrobiologia*, **338**, 49–62.

Laugaste, R., Nõges, P., Nõges, T., Yastremskij, V. V., Milius, A., & Ott, I. 2001. Algae. In *Lake Peipsi. Flora and Fauna* (Pihu, E. & Haberman, J., eds), pp. 31–49. Sulemees Publishers, Tartu.

Laugaste, R., Nõges, T., & Tõnno, I. 2008. Vetikad. In *Peipsi* (Haberman, J., Timm, T., & Raukas, A., eds), pp. 251–270. Eesti Loodusfoto, Tartu.

Li, H., Murphy, T., Guo, J., Parr, T., & Nalewajko, C. 2009. Iron-stimulated growth and microcystin production of *Microcystis novacekii* UAM 250. *Limnologica*, **39**, 255–259.

Marinho, M. M. & Huszar, V. L. M. 2002. Nutrient availability and physical conditions as controlling factors of phytoplankton composition and biomass in a tropical reservoir (Southeastern Brazil). *Archiv für Hydrobiologie*, **153**, 443–468.

Milius, A. & Haldna, M. 2008. Hüdrokeemia. In *Peipsi* (Haberman, J., Timm, T., & Raukas, A., eds), pp. 157–178. Eesti Loodusfoto, Tartu.

Milius, A., Laugaste, R., Möls, T., Haldna, M., & Kangur, K. 2005. Water level and water temperature as factors determining phytoplankton biomass and nutrient content in Lake Peipsi. *Proceedings of the Estonian Academy of Sciences*, **54**, 5–17.

Ni, W., Zhang, J., Ding, T., Stevenson, R. J., & Zhu, Y. 2012. Environmental factors regulating cyanobacteria dominance and microcystin production in a subtropical lake within the Taihu watershed, China. *Journal of Zheijang University SCIENCE*, A **13**, 4, 311–322.

Nõges, T., Laugaste, R., Nõges, P., & Tõnno, I. 2008. Critical N : P ratio for cyanobacteria and N_2-fixing species in large shallow temperate lakes Peipsi and Võrtsjärv, North-East Europe. *Hydrobiologia*, **599**, 77–86.

OECD. 1982. *Eutrophication of Waters, Monitoring, Assessment and Control*. OECD, Paris.

Oliver, R. L. & Ganf, G. G. 2002. Freshwater blooms. In *The Ecology of Cyanobacteria* (Whitton, B. A. & Potts, M., eds), pp. 149–194. Kluwer Academic Publishers, Dordrecht, The Netherlands.

Ou, M.-M., Wang, Y., Zhou, B.-X., & Cai, W.-M. 2006. Effects of iron and phosphorus on *Microcystis* physiological reactions. *Biomedical and Environmental Sciences*, **19**, 399–404.

R Development Core Team. 2011. *R: A Language and Environment for Statistical Computing*. R Foundation for Statistical Computing, Vienna, Austria. http://www.R-project.org (accessed 21.09.2013).

Reynolds, C. S. 2006. *The Ecology of Phytoplankton*. Cambridge University Press, New York.

Salmaso, N. 2000. Factors affecting the seasonality and distribution of cyanobacteria and chlorophytes: a case study from the large lakes south of the Alps, with special reference to Lake Garda. *Hydrobiologia*, **438**, 43–63.

Scheffer, M. S. 1998. *Eclogy of Shallow Lakes*. Chapman & Hall, London.

Scheffer, M., Rinaldi, S., Gragnani, A., Mur, L. R., & Vannes, E. H. 1997. On the dominance of filamentous cyanobacteria in shallow, turbid lakes. *Ecology*, **78**, 272–282.

Tan, X., Kong, F.-x., Cao, H.-s., Yang, Y., & Zhang, M. 2008. Recruitment of bloom-forming cyanobacteria and its driving factors. *African Journal of Biotechnology*, **7**, 4726–4731.

Tanner, R., Kangur, K., Spoof, L., & Meriluoto, J. 2005. Hepatotoxic cyanobacterial peptides in Estonian freshwater bodies and inshore marine water. *Proceedings of the Estonian Academy of Sciences. Biology. Ecology*, **54**, 40–52.

Teubner, K., Feyerabend, R., Henning, M., Nicklisch, A., Woitke, P., & Kohl, J.-G. 1999. Alternative blooming of *Aphanizomenon flos-aquae* or *Planktothrix agardhii* induced by the timing of the critical nitrogen : phosphorus ratio in hypertrophic riverine lakes. *Archiv für Hydrobiologie, Special Issue Advances in Limnology*, **54**, 325–344.

Trimbee, A. M. & Prepas, E. E. 1987. Evaluation of total phosphorus as a predictor of the relative biomass of blue-green algae with emphasis on Alberta lakes. *Canadian Journal of Fisheries and Aquatic Sciences*, **14**, 1337–1342.

Utermöhl, H. 1958. Zur Vervollkommnung der quantitativen Phytoplankton-Methodik. *Mitteilungen - Internationale Vereinigung für theoretische und angewandte Limnologie*, **9**, 1–38.

Vrede, T., Ballantyne, A., Mille-Lindblom, C., Algesten, G., Gudasz, C., Lindahl, S., & Brunberg, A. K. 2009. Effects of N : P loading ratios on phytoplankton community composition, primary production, and N fixation in a eutrophic lake. *Freshwater Biology*, **54**, 331–344.

Wang, H.-J., Liang, X.-M., Jiang, P.-H., Wang, J., Wu, S.-K., & Wang, H.-Z. 2008. TN : TP ratio and planktivorous fish do not affect nutrient–chlorophyll relationships in shallow lakes. *Freshwater Biology*, **53**, 935–944.

Yamamoto, Y. 2009. Environmental factors that determine the occurrence and seasonal dynamics of *Aphanizomenon flos-aquae*. *Journal of Limnology*, **68**, 122–132.