Changes in submerged macrophyte colonization in shallow areas of an oligo-mesotrophic lake and the potential role of groundwater

Cécile Périllon https://orcid.org/0000-0003-2387-4387, Klaus van de Weyer, Jens Päzolt, Peter Kasprzak, Sabine Hilt https://orcid.org/0000-0002-0585-2822

DOI
10.1016/j.limno.2017.03.002

Original publication date
7 April 2017 (Available online)

Document version
Accepted manuscript

Published in
Limnologica

Citation
Périllon C, Van de Weyer K, Päzolt J, Kasprzak P, Hilt S. Changes in submerged macrophyte colonization in shallow areas of an oligo-mesotrophic lake and the potential role of groundwater. Limnologica. 2018;68:168-76.

Accepted Manuscript

Title: Changes in submerged macrophyte colonization in shallow areas of an oligo-mesotrophic lake and the potential role of groundwater

Authors: Cécile Périllon, Klaus van de Weyer, Jens Päzolt, Peter Kasprzak, Sabine Hilt

PII:	S0075-9511(16)30042-1
DOI:	http://dx.doi.org/doi:10.1016/j.limno.2017.03.002
Reference:	LIMNO 25576

To appear in:

Received date:	15-6-2016
Revised date:	7-3-2017
Accepted date:	21-3-2017

Please cite this article as: {http://dx.doi.org/

Changes in submerged macrophyte colonization in shallow areas of an oligo-mesotrophic lake and the potential role of groundwater

Cécile Périllon[1,2*], Klaus van de Weyer[3], Jens Päzolt[4], Peter Kasprzak[5], Sabine Hilt[1]

[1] Leibniz-Institute of Freshwater Ecology and Inland Fisheries, Department of Ecosystem Research, Berlin, Germany

[2] Institute of Biochemistry and Biology, University of Potsdam, Germany

[3] Lanaplan, Nettetal, Germany

[4] Landesumweltamt Brandenburg, Groß Glienicke, Germany

[5] Leibniz-Institute of Freshwater Ecology and Inland Fisheries, Department of Experimental Limnology, Stechlin, Germany

*Corresponding author: perillon@igb-berlin.de

Highlights

- Shallow littoral macrophyte communities changed strongly in a mesotrophic lake
- Charophytes were replaced by angiosperms and vegetation-free areas
- Changes in water column nutrients and transparency do not explain macrophyte changes
- Groundwater-mobilized nutrients promote periphyton
- Periphyton can impair macrophytes by shading, higher sensitivity to waves and benthivorous fish

Abstract

Groundwater influx can significantly contribute to nutrient budgets of lakes and its influence is strongest in shallow littoral areas. In oligo- or mesotrophic systems, additional nutrient supply by groundwater influx may affect benthic primary producers and their interactions. Potential changes can be expected in community composition, biomass, stoichiometry and interactions between submerged macrophytes and epiphyton.

This study aimed at investigating whether enhanced epiphyton growth at sites with groundwater discharge may have contributed to a significant change in shallow littoral macrophyte abundance reported from oligo-mesotrophic Lake Stechlin during the last 50 years. In the 1960s, shallow littoral areas were dominated by small charophyte species such as *Chara aspera, C. filiformis* and *C. rudis*. Recent mappings indicated a strong decline of this shallow water charophyte community from 42 ha to 3 ha and a shift to the occurrence of macrophyte species typical of eutrophic lakes such as *Potamogeton perfoliatus, P. pectinatus* and *Myriophyllum spicatum.* We analyzed the nutrient content of macrophytes, and measured epiphyton growth at sites with different groundwater influence. Water column nutrient enrichment may have increased the abundance of eutrophic species, but this did not explain the decrease of charophytes. Our data suggest that enhanced epiphyton growth in shallow littoral areas with groundwater influx could impair the development of small charophytes by shading, increasing drag forces and the charophytes’ sensitivity to herbivory.

1. Introduction

Submerged macrophytes have important functions in littoral zones of many lakes by influencing suspended solid retention, sediment oxygenation, and providing shelter or support for other primary producers and grazers (Carpenter and Lodge, 1986). They have been suggested to stabilize clear-water conditions in both shallow (Scheffer et al., 1993) and deeper lakes (Hilt et al., 2010; Sachse et al., 2014). During the last century, higher nutrient loading to temperate lakes resulted in a decrease of charophytes (Baastrup-Spohr et al., 2013; Blindow, 1992) and an increase of faster growing macrophyte species such as *Potamogeton pectinatus* (recently named *Stuckenia pectinata)*, *Myriophyllum spicatum*, or *Ceratophyllum demersum* (Sand-Jensen et al., 2000). Eutrophication has also reduced the maximum colonization depth (Middelboe and Markager, 1997), caused a shift to species with a shorter vegetation period (Hilt et al., 2013; Sayer et al., 2010) and ultimately led to a complete decline of submerged macrophytes (Körner, 2002; Sand-Jensen et al., 2000).

A major nutrient-promoted process impeding macrophytes is the development of phytoplankton and epiphyton competing for light. As macrophytes in deeper water are first affected by shading, maximum colonization depth of macrophytes are widely used as an indicator for lake water quality (Kolada et al., 2014; Lyche-Solheim et al., 2013; Penning et al., 2008, Søndergaard et al., 2013). In the shallow littoral, however, macrophytes are supposed to be less affected by turbid water. Macrophytes therefore often find a refuge in shallow water of highly eutrophic lakes (Hilt et al., 2013). However, additional stress factors can affect macrophyte growth even in shallow waters. Macrophytes in the upper littoral may be influenced by water level fluctuations (Deegan et al., 2012), shading by shore vegetation (Köhler et al., 2010) and by epiphyton (periphyton growing on macrophytes, Phillips et al., 1978; Sand-Jensen and Søndergaard, 1981; Tóth and Palmer, 2016) and wave action

(Chambers and Kalff, 1987; Schutten et al., 2004). Shallow macrophytes may also be influenced by groundwater inflow (in the following termed lacustrine groundwater discharge, LGD), which predominantly takes place close to the shoreline (McBride and Pfannkuch, 1975; Rosenberry et al., 2015).

LGD may constitute a significant component of the nutrient budget in nutrients-limited lakes (Lewandowski et al., 2015). Groundwater-borne nutrients may influence macrophyte biomass (Frandsen et al., 2012; Lillie and Barko, 1990; Lodge et al., 1989; Loeb and Hackley, 1988), and the stoichiometry (Sebestyen and Schneider, 2004) and total phosphorus (TP) content of their tissue (Ommen et al., 2012). LGD, however, can also promote epiphyton growth (Hagerthey and Kerfoot 1998, 2005) which may increase shading and drag forces on macrophytes in shallow habitats (Périllon and Hilt, 2016).

Here, we evaluate the changes in the abundance and species composition of shallow littoral macrophytes in a groundwater-fed oligo-mesotrophic hardwater lake and the potential role of LGD in this process. In a previous study, a potential impact of groundwater-mobilized nutrients on periphyton growth has been shown for this lake (Périllon et al., in revision). We hypothesize that this process may contribute to a change in macrophyte species composition towards a community with more species typical for eutrophic lakes and a decline of charophytes in shallow littoral areas. To test these hypotheses, we compared the macrophyte species composition and abundance in shallow areas in 1962, 2002, 2007, 2008 and 2014. In addition, macrophyte tissue nitrogen (N) and phosphorus (P) concentrations were measured in five macrophyte species at locations with and without LGD in 2014. Epiphyton development was monitored in summer 2014 on artificial substrates at four locations with or without LGD.

2. *Materials and methods*

2.1 Lake Stechlin

Lake Stechlin is a temperate, monomictic hard-water lake in northeastern Germany (Table 1), fed by groundwater and rainfall, with a stable water level since 1962. Short-term water level changes are controlled by climatic conditions such as wind and precipitations (Kirillin et al., 2013a).

In summer 2012, a piezometer campaign aimed at localizing areas with LGD using stable isotopes as indicators (Périllon et al., in revision). We generalized these data for the present study area (0 to 2 m deep) using the Voronoi polygons tool (QGIS 2.12.0) and selected the area situated between the shore and the 2 m depth line, using a bathymetric map (Fig. 1A). The areas located next to a piezometer with low δ^{18}O signature (between -10‰ and -6‰) were characterized as "LGD" and areas with higher δ^{18}O values (between -6‰ and -2‰) as "C" (control). The most eastern bay was excluded from the analysis due to its anthropogenic use as beach area (Fig. 1A). The main locations for LGD are in the southern, south-eastern and western littoral, while the outflow is concentrated in the northern littoral of the lake. All our sampling points were located in areas with stable groundwater flow direction, apart from the eastern control which could show inter-annual variation in flow direction, e.g. after wet years (Holzbecher, 2001).

2.2 Macrophyte mapping

Macrophyte surveys have been performed during the summers 1962 (Krausch, 1964), 2002, 2007 (unpublished data of Landesumweltamt Brandenburg), 2008 (Van de Weyer et al., 2009), and 2014 (Van de Weyer et al., 2015, Fig. 1B). From 1962 we could only access the maps (Fig. 1B) and the list of species present in the whole lake (Table 2). In 2002, 2007, 2008 and 2014, macrophytes have been surveyed on 7 identical transects (straight lines that begin perpendicular to the shore). 13 further transects were surveyed in 2008 and 2014.

The mappings performed in 2008 and 2014 (20 transects) were most detailed. First, vegetation zones were mapped in June/July from a boat using an underwater camera and macrophyte were identified after sampling with a rake. Additionally, a diver followed the borders of specific populations of vegetation with a GPS buoy. Finally, divers mapped 20 transects to define more precisely macrophyte habitats and identify maximum colonization depths. Macrophyte species were determined following Van de Weyer and Schmitt (2011) and the macrophyte zones were identified after Berg et al. (2004). For each vegetation zone, the coverage was estimated in the field using the decimal Londo scale (Londo, 1976) and then translated into percentage of coverage, with values ranging between 0.1% (single macrophyte) to 97.5% (single species continuous cover).

Macrophyte species were classified following the indicator values defined in Schaumburg et al. (2015) for the lake type TKg13 (carbonate-rich stratified water body of northern German lowlands with small watershed). “A” species are typical for pristine undisturbed conditions characteristic of this lake type, “B” species are more indifferent and “C” species indicate a deviation from reference conditions for this lake type (Schaumburg et al., 2004). The classification of charophytes (Kabus and Mauersberger, 2011) and angiosperms (Ristow et al., 2006) in red list categories for Brandenburg, are presented in Table 2.

For data evaluation, we selected macrophyte data from the two first meters depth using QGIS. The indicator values were attributed following the species and the depth limits of vegetation zones: when the zone upper limit were shallower than 1m, the indicator values corresponding to 0-1 m (Schaumburg et al., 2015) were attributed to the macrophytes.

Indicator values corresponding to 1-2 m were attributed to deeper zones.

First data analysis consisted of the comparison of the number of macrophytes species present at 0 – 2 m depth, in the 7 common transects studied in 2002, 2007, 2008 and 2014 (Fig. 2A). The number of macrophyte species typically growing in shallow littoral, are also represented for the year 1962 (Fig. 2A).

Further analysis required the calculation of coverage data within transects, using the data from 20 transects, in 2008 and 2014. The coverage of each macrophyte species were added for each indicator value and transects. The percentage of the littoral area covered by the vegetation zones were used as an adjustment value. Often species were observed as single plants, or only in few transects, therefore the obtained values averaged among transects and species, are low.

2.3 Epilimnion water quality

Data on surface water quality (Secchi depth, total phosphorus (TP) concentration) were gathered from regular monthly monitoring in the middle of Lake Stechlin (Kasprzak et al., 2017) from 1970 onwards. TP concentration were analysed according to DIN 38405. We selected data from the water surface (< 1m depth), which were supposed to best represent conditions in the littoral zone, and the four years preceding the macrophyte surveys. The data were split in two periods: summer (from May to September, when the lake is stratified) and winter. The 20 to 40 single data per period and parameter were presented as boxplot (Fig. 3). Previous analyses of water sampled at different littoral sites showed no difference to data of the lake centre (data not shown).

2.4 Macrophyte tissue nutrient concentration

We sampled submerged macrophytes in 40 to 70 cm water depth at four sites of Lake

Stechlin in July and August 2014 (Fig. 1A, Fig. 4). At this depth, the macrophyte community only consisted of single plants. Five macrophyte species were present in sufficient quantity to be included: *Myriophyllum spicatum*, *Potamogeton lucens*, *P. perfoliatus*, *P. pectinatus* and *Najas marina*. We sampled selectively young tissue to reduce the possible effect of age on nitrogen (N) and phosphorus (P) contents. For *M. spicatum*, *P. lucens* and *P. perfoliatus* we selected new leaves, for *N. marina* and *P. pectinatus* we sampled the whole above-ground plant. For each macrophyte species, several leaves or plants were sampled per replicate, and we took 4 replicates per site and sampling date. The samples were gently cleaned from epiphyton and dried at 60°C until the constant weight. The ground probes were analyzed with the ammonium molybdate spectrometric method for P. TC (total carbon) and TN (total nitrogen) tissue content were determined with a CHN elemental analyser (Vario EL, Germany).

2.5 Epiphyton biomass and shading

Epiphyton was sampled from transparent polypropylene sheets (IBICO, GBC, Chicago. Il, USA), that mimic macrophytes by being flexible and having a slightly rough surface. Strips (2 x 12cm) were installed vertically in 50 cm water depth, fixed to the sediment with a metallic structure. The sampled surface extended from 5 cm above the sediment surface to around 30cm from the water surface. The strips were installed parallel to the shore line, and in areas with limited shading from trees or macrophytes (Fig. 1A).

After four weeks exposure, we sampled eight stripes at each location and transported them to the laboratory in a dark, humid and cooled box. Stripes were exposed to carbonated water to remove grazers, and epiphyton was scrubbed with a toothbrush into filtered (0.45 µm cellulose acetate filters, Sartorius, Göttingen) lake water. The suspension obtained from two stripes was filtered on to a pre-weighed GF/F glass-fibre filter and dried for 12 hours at 105°C for obtaining epiphyton dry weight (dw). We obtained 4 replicates (each from two stripes), per site and per sampling campaign. Light attenuation by epiphyton was calculated following Köhler et al. (2010) using the formula: light attenuation=(108 × epiphyton dw)/(9.2 + epiphyton dw).

Carbon and N concentrations were obtained from the filters, and P concentrations from the suspension, following the same methods as described for the macrophytes (Fig. 5).

2.6 Statistics

To evaluate differences of epiphyton and macrophyte parameters between samples in LGD or in control (C) conditions, or between years, we applied Student's t-test for normally distributed data with equal variances, or for small samples sizes. When normality could not be attained, we applied the non-parametrical Mann-Whitney-U-test. All statistical analyses were performed with the software R (R Development Core Team, 2014) and the data represented with ggplot2 (Wickham, 2009).

3. Results

3.1 Macrophyte community composition and abundance

In 1962, low-growing charophyte meadows were abundant along two third of Lake Stechlin's littoral shore line and covered 42 ha (Fig. 1C). In 2008 and 2014, this area had shrunk to 3 ha, present in the northern bay and few stands further from the shore, next to the eastern bay (Fig. 1C). Submerged angiosperms were present along most of the lake shore, with a prevalence on the western shore and bay ends. They covered 17 ha in 1962, 13 ha in 2008 and 33 ha in 2014. Areas without submerged vegetation were only mapped in 2014, and covered 31 ha of the whole littoral.

Since 1962, species of all three indicator values (A, B and C) have been present in the lake, but the number of A species tended to decrease (Fig. 2A). Specifically, the A species *Chara aspera and C. rudis* have not been observed after 1962, and *C. intermedia* after 2002 (Table 2). *Najas marina ssp. intermedia* and *P. crispus* appeared as additional C species in 2002 and 2008, respectively. While healthy stands of the A specie *C. filiformis* were observed in 1962, in the last campaigns only isolated plants were reported.

The percentage of transect coverage per species have been calculated for 20 transects in 2008 and 2014, and show different patterns for A, B, C species (Fig. 2B). “A” species were less abundant in 2014 than in 2008 (Mann-Whitney-U-test, $p<0.01$). There were no significant differences in the abundance between A and C species in 2008 (Mann-Whitney-U-tests, $p>0.05$), while C species became more abundant than A species in 2014 (Mann-Whitney-U-tests, $p<0.001$). The number of species per transect was extremely variable, and no significant differences between years were found. Also, we could not find any significant differences in macrophyte coverage between transects located in LGD sites and other transects.

3.2 Surface water quality

Average TP concentrations of the surface water of Lake Stechlin were 12.0 ± 4.6 (sd) µg L^{-1} during the 16 observed years, with slightly higher values during the winter period (10.6 ± 3.3 µg L^{-1} in summer and 13.2 ± 5.3 µg L^{-1} in winter). Summer TP concentrations during the periods 2005 - 2008 and 2011 - 2014 were significantly higher than in the two previous time period (1970 - 1973 and 1999 - 2002, $p<0.001$, Mann-Whitney-U-tests, Fig. 3). Winter TP concentrations in surface water of Lake Stechlin have increased from 1970-73 to 1999-2002 and to 2005-08 and did not significantly differ between the two last periods (Mann-Whitney-U-tests, $p<0.001$, $p<0.001$ and $p>0.05$ respectively). Summer Secchi depths significantly decreased during the three last periods ($p<0.001$, Mann-Whitney-U-tests), with means of 8.9 ± 1.3 m, 7.4 ± 1.3 m and 5.9 ± 1.4 m, respectively (Fig. 3).

3.3 Macrophyte tissue nutrient concentrations

Macrophyte tissue P content ranged between 0.07 % dw and 0.41 % dw and the N content varied between 1.04% dw and 3.58% dw (Fig. 4). 89 % of the P values were above 0.13 % dw, and 98 % of the N data were above 1.3 % dw, the thresholds for P and N limitation suggested by Gerloff and Krombholz (1966).

Macrophyte tissue nutrient content mostly did not differ significantly between LGD and C sites (Student's t-tests, $p>0.05$), apart from *N. marina spp. intermedia* and *P. lucens* ($p<0.05$).

3.4 Epiphyton shading

Epiphyton dry weight varied between 0.97 g m^{-2} and 13.5 g m^{-2}, with averages of 7.4 g m^{-2} in LGD sites and 5.4 g m^{-2} in control sites. Light attenuation by epiphyton varied between 10.3 % and 64.3 % of incoming photosynthetically active radiation (PAR), and was significantly higher in LGD sites than in control sites (Student's t-test, $p<0.05$, Fig. 5A). More P accumulated per area in epiphyton at LGD sites (12.4 ± 5.4 mg m^{-2}) compared to C sites (8.3 ± 3.9 mg m^{-2}) (Student's t-test, $p<0.01$, Fig. 5D). Most of C:P ratio and all C:N ratio in epiphyton were above the thresholds for P or N limitation identified by Kahlert (1998).

4. Discussion

Shallow littoral macrophyte communities of Lake Stechlin showed strong changes in the last

50 years, both in species composition and abundance. Low-growing charophyte communities in shallow water have almost completely disappeared while macrophyte species typical of eutrophic conditions have become abundant, and more areas completely lack vegetation. Increasing summer TP concentrations and decreasing Secchi depth in surface water of Lake Stechlin indicate a process of slight eutrophication, especially during the last decade. This process may have promoted the occurrence and dispersal of angiosperm species typical for eutrophic conditions in the shallow littoral areas. A competition for space in the shallow littoral between low-growing charophytes and angiosperms seems unlikely as the vegetation is generally rather sparse. Our data on higher periphyton biomass at shallow areas without charophyte stands and with groundwater discharge and our previous detailed study on groundwater-mobilized nutrients potentially affecting periphyton growth (Périllon et al., in revision) indicate that groundwater may indirectly affect both macrophyte species composition and abundance in shallow littoral areas due to a stimulation of epiphyton growth shading the macrophytes, increasing the drag forces of waves, especially on small rootless charophytes and enhancing the sensitivity of macrophytes to disturbances by herbivorous and benthivorous cyprinids.

4.1 Macrophyte development in shallow littoral areas

In the shallow littoral areas of Lake Stechlin, we observed a strong decline of both presence and abundance of macrophyte species characteristic for the reference oligotrophic conditions in this lake type. The observed change in macrophyte abundance is especially visible between 2008 and 2014, however, this could be partly explained by inter-annual fluctuation, and a confirmation of this trend would need further data acquisition. The once abundant charophyte species *C. aspera* and *C. intermedia* had completely disappeared in 2008 and 2014, while *C. rudis* had declined in abundance. A decline of charophytes with increasing eutrophication has been described for the period since 1940 in several lakes in Southern Sweden (Blindow, 1992) and in Denmark (Baastrup-Spohr et al., 2013). Both studies point to a disappearance of macrophytes in deep waters in alkaline lakes and consequently a prevalence of reduced light availability as a main reason for this historical decline. The situation in Lake Stechlin, and dozens of other lakes in the region (R. Mauersberger, pers. comm.) seems different.

Secchi disk transparencies and TP concentrations indicate a slight eutrophication trend, especially during the last decade. The reasons for this development are subject of current research (Kirillin et al., 2013b). The effect of decreasing light availability in the water column for shallow littoral habitats, however, was supposed to be of low relevance. Minimum light requirements for charophytes and submerged angiosperms are comparable (Sand-Jensen and Madsen, 1991). In addition, Kovtun-Kante et al. (2014) have shown that shallow water *C. aspera* communities could adapt to reduced light availability. In contrast to light, the increasing P availability in the water column may have contributed to the decline of shallow water charophytes by promoting macrophyte species typical for eutrophic conditions. Richter and Gross (2013) reported that charophyte stands can negatively affect tall-growing angiosperms under low P availability and high water clarity. This implies that taller angiosperms may outcompete low-growing charophytes at increasing TP concentrations. The tissue P- and N-contents of the five angiosperms abundant in Lake Stechlin were all well above the threshold levels indicating limiting conditions (Gerloff and Krombholz, 1966), which suggests that nutrient availability in the sandy littoral was sufficient. P- and N- contents of *M. spicatum* were similar to the values obtained by Gross (2009) in the littoral of Lake Constance, at a time when charophytes increased and started replacing *Myriophyllum* during re-oligotrophicaton. However, angiosperm densities are still rather low and vegetation-free areas are abundant indicating that competition with angiosperms is at least not the only reason for the observed charophyte decline.

4.2 Potential influence of groundwater

A direct influence of groundwater discharge on angiosperm biomass or nutrient content could not be found in Lake Stechlin, contrary to other studies which showed a promotion of macrophytes by LGD (Frandsen et al., 2012; Lodge et al., 1989; Ommen et al., 2012). However, our data show an increased epiphyton growth at the LGD sites. A previous study indicated that sediment nutrients, and especially phosphorus, may have been mobilized by groundwater discharge (Périllon et al., in revision) and stimulated epiphyton growth.

The present epiphyton biomass production in Lake Stechlin was higher than 40 years ago. Scheffler (1981) applied the ^{14}C method and measured an epiphyton production of approximately 28 ± 7 mg C m^{-2} d^{-1} on artificial substrates exposed for 4 weeks at a sunny littoral site during May-September of 1971-74 while we measured 64.5 mg C m^{-2} d^{-1} at LGD sites and 51.6 mg C m^{-2} d^{-1} at control sites during the same season in 2014. The question, whether the observed development of epiphyton was significantly affected by changes in the groundwater discharge, P availability in the groundwater or in the littoral sediment, remains open. Since 2011, several wet summers caused increasing water levels in many lakes of the region after 30 years of droughts and falling lake levels (Kaiser et al., 2014). Changes in groundwater flow may have influenced nutrient fluxes and mobilization processes, but groundwater-induced nutrient fluxes were not studied in the past. However, one could speculate that P availability in the littoral may have increased due to a feedback with epi- and periphyton growth in the groundwater-influenced locations resulting in effects similar to those described as “nearshore P shunt” for areas with abundant dreissenids (Hecky et al., 2004).

This groundwater-promoted epiphyton growth in shallow littoral areas may have affected the low-growing charophyte species, which can currently only be found in littoral areas without LGD, in three ways. It may firstly have increased shading. In our study, epiphyton shading reached up to 64 % reduction of incoming PAR and growth limitations have been observed for values between 26 % PAR reduction for *P. pectinatus* (Vierssen and Hootsmans, 1994) to 95 % for charophytes (Middelboe and Markager, 1997). Most of the littoral of Lake Stechlin is shaded by trees which together with epiphyton can significantly contribute to macrophyte biomass reductions (Ali et al., 2011; Köhler et al., 2010). Secondly, epiphyton may enhance sensitivity of macrophytes to wave drag forces (Schutten et al., 2004). Especially charophytes may be more sensitive to high fetch than other macrophytes, as observed by Schmieder and Lehmann (2004) in Lake Constance. This could explain the loss of charophytes at the wind-exposed northern sites. Finally, it has been shown in a modeling study and by a meta-analysis that epiphyton increases the susceptibility of macrophytes to herbivory (Hidding et al., 2016). An influence of herbivorous and benthivorous cyprinids on charophytes in Lake Stechlin has been suggested based on the increase of vegetation-free areas and typical sediment patterns indicating physical disturbance by benthivorous fish (Van de Weyer et al., 2015). Negative effects on charophytes have been shown in other lakes for a number of fish species such as common carp (Laguna et al., 2016), grass carp (Krupska et al., 2012), bream (Ten Winkel and Meulemans, 1984) and rudd (Lake et al., 2002). Long-term exclosure experiments should be performed in the shallow littoral of Lake Stechlin to clarify the impact of herbivorous and benthivorous fish.

5. Conclusion

We conclude that there is a strong need for further research on the reasons of charophyte decline observed in the shallow littoral area of Lake Stechlin and other oligo-mesotrophic lakes in North-Eastern Germany. Their decline does not follow the typical pattern described for eutrophication of freshwater habitats with an initial loss of species in deeper water habitats. Groundwater-mobilized nutrients may play a role by promoting epiphyton growth leading to increased shading and sensitivity of wave action and herbivory, but further detailed studies need to clarify the origin of the nutrients and the impact of other factors such as disturbances by herbivorous and benthivorous cyprinids.

Acknowledgements

The authors thank Sebastian Meis, Volker Krautkrämer and Berta Ortiz-Crespo for their participation in field and lab work. The Department of Experimental Limnology of the IGB kindly provided data from the long-term Stechlin lake monitoring, and the possibility of using material and lab space. We also acknowledge the help of the Department of Chemical Analytics and Biogeochemistry of the IGB in nutrient analyses of epiphyton and macrophytes, and of Jörg Lewandowski for the information on groundwater. The wind data were kindly provided by Markus Wallasch (Umweltbundesamt). Finally, we thank the reviewers and the guest editor Henning Jensen for their valuable comments on an earlier version of the manuscript. The first author was funded by the Leibniz Association (SAW- 2012-IGB 4167).

References

Ali, M.M., Hassan, S.A., Shaheen, A.-S.M., 2011. Impact of riparian trees shade on aquatic plant abundance in conservation islands. Acta Bot. Croat. 70, 245–258. doi:10.2478/v10184-010-0012-7

Baastrup-Spohr, L., Iversen, L.L., Dahl-Nielsen, J., Sand-Jensen, K., 2013. Seventy years of changes in the abundance of Danish charophytes. Freshw. Biol. 58, 1682–1693. doi:10.1111/fwb.12159

Berg, C., Dengler, J., Abdank, A., Isermann, M., 2004. Die Pflanzengesellschaften Mecklenburg-Vorpommern und ihre Gefährdung. 2 Bände. Weissdorn-Verlag, Jena.

Blindow, I., 1992. Decline of charophytes during eutrophication: comparison with angiosperms. Freshw. Biol. 28, 9–14.

Carpenter, S.R., Lodge, D.M., 1986. Effects of submersed macrophytes on ecosystem processes. Aquat. Bot. 341–370.

Chambers, P.A., Kalff, J., 1987. Light and nutrients in the control of aquatic plant community structure. I. In situ experiments. J. Ecol. 75, 611–619.

Deegan, B.M., White, S.D., Ganf, G.G., 2012. Nutrients and water level fluctuations: a study of three aquatic plants. River Res. Appl. 28, 359–368. doi:10.1002/rra.1461

DIN 38405, n.d. Deutsche Einheitsverfahren (DEV) zur Wasser-, Abwasser- und Schlammuntersuchung. Anal. Stand. DIN 38405, part 21.

Frandsen, M., Nilsson, B., Engesgaard, P., Pedersen, O., 2012. Groundwater seepage stimulates the growth of aquatic macrophytes. Freshw. Biol. 57, 907–921. doi:10.1111/j.1365-2427.2012.02747.x

Gerloff, G.C., Krombholz, P.H., 1966. Tissue analysis as a measure of nutrient availability for the growth of angiosperm aquatic plants. Limnol. Oceanogr. 11, 529–537.

Gross, E.M., 2009. Impact of reoligotrophication in Lake Constance on nutrient content and defensive phenolic compounds in Eurasian watermilfoil. Int. Assoc. Theor. Appl. Limnol. Vol 30, Pt 6, Proc. 30, 897–899.

Hagerthey, S.E., Kerfoot, W.C., 2005. Spatial variation in groundwater-related resource supply influences freshwater benthic algal assemblage composition. J. North Am. Benthol. Soc. 24, 807–819.

Hagerthey, S.E., Kerfoot, W.C., 1998. Groundwater flow influences the biomass and nutrient ratios of epibenthic algae in a north temperate seepage lake. Limnol. Oceanogr. 43, 1127–1242.

Hecky, R.E., Smith, R.E.H., Barton, D.R., Guildford, S.J., Taylor, W.D., Charlton, M.N., Howell, T., 2004. The nearshore phosphorus shunt: a consequence of ecosystem engineering by dreissenids in the Laurentian Great Lakes. Can. J. Fish. Aquat. Sci. 61, 1285–1293. doi:10.1139/f04-065

Hidding, B., Bakker, E.S., Hootsmans, M.J.M., Hilt, S., 2016. Synergy between shading and herbivory triggers macrophyte loss and regime shifts in aquatic systems. Oikos 125, 1489–1495. doi:10.1111/oik.02629

Hilt, S., Köhler, J., Adrian, R., Monaghan, M.T., Sayer, C.D., 2013. Clear, crashing, turbid and back - long-term changes in macrophyte assemblages in a shallow lake. Freshw. Biol. 58, 2027–2036. doi:10.1111/fwb.12188

Hilt, S., Van de Weyer, K., Köhler, A., Chorus, I., 2010. Submerged macrophyte responses to reduced phosphorus concentrations in two peri-urban lakes. Restor. Ecol. 18, 452–461. doi:10.1111/j.1526-100X.2009.00577.x

Holzbecher, E., 2001. The dynamics of subsurface water divides - watersheds of Lake Stechlin and neighbouring lakes. Hydrol. Process. 15, 2297–2304. doi:10.1002/hyp.261

Kabus, T., Mauersberger, R., 2011. Liste und Rote Liste der Armleuchteralgen (Characeae) des Landes Brandenburg. Natur und Landschaftpfl. Brand.

Kahlert, M., 1998. C:N:P ratios of freshwater benthic algae. Arch. Hydrobiol. (Suppl) (Advanc. Limnol.) 51, 105–114.

Kaiser, K., Koch, P.J., Mauersberger, R., Stüve, P., Dreibrodt, J., Bens, O., 2014. Detection and attribution of lake-level dynamics in north-eastern central Europe in recent decades. Reg. Environ. Chang. 14, 1587–1600. doi:10.1007/s10113-014-0600-5

Kasprzak, P., Shatwell, T., O.Gessner, M., Gonsiorczyk, T., Kirillin, G., Selmeczy, G., Padisák, J., Engelhardt, C., 2017. Extreme weather event triggers cascade towards extreme turbidity in a clear-water lake. Ecosystems in press.

Kirillin, G., Phillip, W., Engelhardt, C., Nützmann, G., 2013a. Net groundwater inflow in an enclosed lake: From synoptic variations to climatic projections. Hydrol. Process. 27, 347–359. doi:10.1002/hyp.9227

Kirillin, G., Shatwell, T., Kasprzak, P., 2013b. Consequences of thermal pollution from a nuclear plant on lake temperature and mixing regime. J. Hydrol. 496, 47–56. doi:10.1016/j.jhydrol.2013.05.023

Köhler, J., Hachoł, J., Hilt, S., 2010. Regulation of submersed macrophyte biomass in a temperate lowland river: interactions between shading by bank vegetation, epiphyton and water turbidity. Aquat. Bot. 92, 129–136. doi:10.1016/j.aquabot.2009.10.018

Kolada, A., Willby, N., Dudley, B., Nõges, P., Søndergaard, M., Hellsten, S., Mjelde, M., Penning, E., Van Geest, G., Bertrin, V., Ecke, F., Mäemets, H., Karus, K., 2014. The applicability of macrophyte compositional metrics for assessing eutrophication in European lakes. Ecol. Indic. 45, 407–415. doi:10.1016/j.ecolind.2014.04.049

Körner, S., 2002. Loss of submerged macrophytes in shallow lakes in north-eastern Germany. Int. Rev. Hydrobiol. 87, 375–384. doi:10.1002/1522-2632(200207)87:4<375::AID-IROH375>3.0.CO;2-7

Kovtun-Kante, A., Torn, K., Kotta, J., 2014. In situ production of charophyte communities under reduced light conditions in a brackish-water ecosystem. Est. J. Ecol. 63, 28–38. doi:10.3176/eco.2014.1.03

Krausch, H.D., 1964. Die Pflanzengesellschaften des Stechlinsee-Gebietes I. Die Gesellschaften des offenen Wassers. Limnologica 2, 145–203.

Krey, L., 1985. The lakes of the Lake Stechlin area: aspects of their morphometry, in: Lake Stechlin. Dr. W. Junk Publishers, Dordrecht, pp. 29–40.

Krupska, J., Pełechaty, M., Pukacz, A., Ossowski, P., 2012. Effects of grass carp introduction on macrophyte communities in a shallow lake. Oceanol. Hydrobiol. Stud. 41, 35–40. doi:10.2478/s13545-012-0004-4

Laguna, C., López-Perea, J.J., Viñuela, J., Florín, M., Feliu, J., Chicote, Á., Cirujano, S., Mateo, R., 2016. Effects of invasive fish and quality of water and sediment on macrophytes biomass, and their consequences for the waterbird community of a Mediterranean floodplain. Sci. Total Environ. 551–552, 513–521. doi:10.1016/j.scitotenv.2016.02.059

Lake, M.D., Hicks, B.J., Wells, R.D.S., Dugdale, T.M., 2002. Consumption of submerged aquatic macrophytes by rudd (*Scardinius erythrophthalmus* L.) in New Zealand. Hydrobiologia 470, 13–22. doi:10.1023/A:1015689432289

Lewandowski, J., Meinikmann, K., Nützmann, G., Rosenberry, D.O., 2015. Groundwater - the disregarded component in lake water and nutrient budgets. 2. Effects of groundwater on nutrients. Hydrol. Process. 29, 2922–2955. doi:10.1002/hyp.10384

Lillie, R.A., Barko, J.W., 1990. Influence of sediment and groundwater on the distribution and biomass of *Myriophyllum spicatum* L. in Deveil's Lake, Wisconsin. J. Freshw. Ecol. 5, 417–426.

Lodge, D.M., Krabbenhoft, D.P., Striegl, R.G., 1989. A positive relationship between groundwater velocity and submersed macrophyte biomass in Sparkling lake, Wisconsin. Limnol. Oceanogr. 34, 235–239.

Loeb, S.L., Hackley, S.H., 1988. The distribution of submerged macrophytes in Lake Tahoe, California and Nevada, and the possible influence of groundwater seepage. Verhandlungen der Int. Vereinigung für Theor. und Angewante Limnol. 23, 1927–1933.

Londo, G., 1976. The decimal scale for releves of permanent quadrats. Vegetatio 33, 61–64.

Lyche-Solheim, A., Feld, C.K., Birk, S., Phillips, G., Carvalho, L., Morabito, G., Mischke, U., Willby, N., Søndergaard, M., Hellsten, S., Kolada, A., Mjelde, M., Böhmer, J., Miler, O., Pusch, M.T., Argillier, C., Jeppesen, E., Lauridsen, T.L., Poikane, S., 2013. Ecological status assessment of European lakes: a comparison of metrics for phytoplankton, macrophytes, benthic invertebrates and fish. Hydrobiologia 704, 57–74. doi:10.1007/s10750-012-1436-y

McBride, M.S., Pfannkuch, H.O., 1975. The distribution of seepage within lakebeds. J. Res. USGS 3, 505–512.

Middelboe, A.L., Markager, S., 1997. Depth limits and minimum light requirements of freshwater macrophytes. Freshw. Biol. 37, 553–568.

Ommen, D.A.O., Kidmose, J., Karan, S., Flindt, M.R., Engesgaard, P., Nilsson, B., Andersen, F.Ø., 2012. Importance of groundwater and macrophytes for the nutrient balance at oligotrophic Lake Hampen, Denmark. Ecohydrology 5, 286–296. doi:10.1002/eco.2

Penning, W.E., Mjelde, M., Dudley, B., Hellsten, S., Hanganu, J., Kolada, A., van den Berg, M., Poikane, S., Phillips, G., Willby, N., Ecke, F., 2008. Classifying aquatic macrophytes as indicators of eutrophication in European lakes. Aquat. Ecol. 42, 237–251. doi:10.1007/s10452-008-9182-y

Périllon, C., Hilt, S., 2016. Groundwater influence differentially affects periphyton and macrophyte production in lakes. Hydrobiologia 778, 91–103. doi:10.1007/s10750-015-2485-9

Périllon, C., Pöschke, F., Lewandowski, J., Hupfer, M., Hilt, S., in revision. Stimulation of epiphyton growth by lacustrine groundwater discharge to an oligo - -mesotrophic hardwater lake. Freshw. Sci.

Phillips, G.L., Eminson, D., Moss, B., 1978. A mechanism to account for macrophytes decline in progressively eutrophicated freshwaters. Aquat. Bot. 4, 103–126.

R Development Core Team, R., 2014. R: a language and environment for statistical computing. R Found. Stat. Comput. doi:10.1007/978-3-540-74686-7

Richter, D., Gross, E.M., 2013. *Chara* can outcompete *Myriophyllum* under low phosphorus supply. Aquat. Sci. 75, 457–467. doi:10.1007/s00027-013-0292-9

Ristow, M., Herrmann, A., Illig, H., Klage, H.-C., Klemm, G., Kummer, V., Machatzi, B., Rätzel, S., Schwarz, R., Zimmermann, F., 2006. Liste und Rote Liste der etablierten

Gefäßpflanzen Brandenburgs. Naturschutz und Landschaftspfl. Brand. Bg.

Rosenberry, D.O., Lewandowski, J., Meinikmann, K., Nützmann, G., 2015. Groundwater - the disregarded component in lake water and nutrient budgets. Part 1: effects of groundwater on hydrology. Hydrol. Process. 29, 2895–2921. doi:10.1002/hyp.10403

Sachse, R., Petzoldt, T., Blumstock, M., Moreira, S., Rücker, J., Janse, J.H., Mooij, W.M., Hilt, S., Marlene, P., 2014. Extending one-dimensional models for deep lakes to simulate the impact of submerged macrophytes on water quality. Environ. Model. Softw. 61, 410–423. doi:10.1016/j.envsoft.2014.05.023

Sand-Jensen, K., Madsen, T.V., 1991. Minimum light requirements of submerged freshwater macrophytes in laboratory growth experiments. J. Ecol. 79, 749–764.

Sand-Jensen, K., Riis, T., Vestergaard, O., Larsen, S.E., 2000. Macrophyte decline in Danish lakes and streams over the past 100 years. J. Ecol. 88, 1030–1040. doi:10.1046/j.1365-2745.2000.00519.x

Sand-Jensen, K., Søndergaard, M., 1981. Phytoplankton and epiphyte development and their shading effect on submerged macrophytes in lakes of different nutrient status. Int. Rev. der gesamten Hydrobiol. 66, 529–552.

Sayer, C.D., Davidson, T.A., Jones, J.I., 2010. Seasonal dynamics of macrophytes and phytoplankton in shallow lakes: A eutrophication-driven pathway from plants to plankton? Freshw. Biol. 55, 500–513. doi:10.1111/j.1365-2427.2009.02365.x

Schaumburg, J., Schranz, C., Hofmann, G., Stelzer, D., Schneider, S., Schmedtje, U., 2004. Macrophytes and phytobenthos as indicators of ecological status in German lakes—a contribution to the implementation of the Water Framework Directive. Limnologica 34, 302–314.

Schaumburg, J., Schranz, C., Stelzer, D., Vogel, A., 2015. Verfahrensanleitung für die ökologische Bewertung von Seen zur Umsetzung der EU- Wasserrahmenrichtlinie :

Makrophyten und Phytobenthos Phylib.

Scheffer, M., Hosper, S.H., Meijer, M., Moss, B., Jeppesen, E., 1993. Alternative equilibria in shallow lakes. Tree 8, 275–279.

Scheffler, W., 1981. Quantitative Erfassung des Periphytons im Stechlinsee. Limnologica 13, 101–122.

Schmieder, K., Lehmann, A., 2004. A spatio-temporal framework for efficient inventories of natural resources: a case study with submersed macrophytes. J. Veg. Sci. 15, 807–816.

Schutten, J., Dainty, J., Davy, A.J., 2004. Wave-induced hydraulic forces on submerged aquatic plants in shallow lakes. Ann. Bot. 93, 333–341. doi:10.1093/aob/mch043

Sebestyen, S.D., Schneider, R.L., 2004. Seepage patterns, pore water, and aquatic plants: hydrological and biogeochemical relationships in lakes. Biogeochemistry 68, 383–409.

Søndergaard, M., Phillips, G., Hellsten, S., Kolada, A., Ecke, F., Mäemets, H., Mjelde, M., Azzella, M.M., Oggioni, A., 2013. Maximum growing depth of submerged macrophytes in European lakes. Hydrobiologia 704, 165–177. doi:10.1007/s10750-012-1389-1

Team, Q.D., 2015. QGIS Geographic Information System.

Ten Winkel, E.H., Meulemans, J.T., 1984. Effects of fish upon submerged vegetation. Hydrobiol. Bull. 18, 157–158.

Tóth, V.R., Palmer, S.C.J., 2016. Acclimation of *Potamogeton perfoliatus* L. to periphyton accumulation-induced spectral changes in irradiance. Hydrobiologia 766, 293–304. doi:10.1007/s10750-015-2462-3

Van de Weyer, K., Meis, S., Krautkrämer, V., 2015. Die Makrophyten des Großes Stechlinsees, des Wummsees und des Wittwensees. Fachbeiträge des LUGV, H. 145.

Van de Weyer, K., Päzolt, J., Tigges, P., Raape, C., Oldorff, S., 2009. Flächenbilanzierungen submerser Pflanzenbestände – dargestellt am Beispiel des Großen Stechlinsees (Brandenburg) im Zeitraum von 1962-2008. Naturschutz und Landschaftspfl. Brand. 18, 1–6.

Van de Weyer, K., Schmitt, C., 2011. Bestimmungsschlüssel für die aquatischen Makrophyten (Gefäßpflanzen, Armleuchteralgen und Moose) in Deutschland: Band 1: Bestimmungsschlüssel. Fachbeiträge des LUGV Brand.

Vierssen, W. Van, Hootsmans, M.J.M., 1994. Photoperiodic effects on photosynthesis and tuber production in *Potamogeton pectinatus* L., in: Vierssen, W. Van, Hootsmans, M.J.M., J.E.Vermaat (Eds.), Lake Veluwe, A Macrophyte-Dominated System Under Eutrophication Stress. Kluwer Academic Publishers, Dordrecht, p. 118–134.

Wickham, H., 2009. ggplot2: Elegant graphics for data analysis. Springer-Verlag, New York.

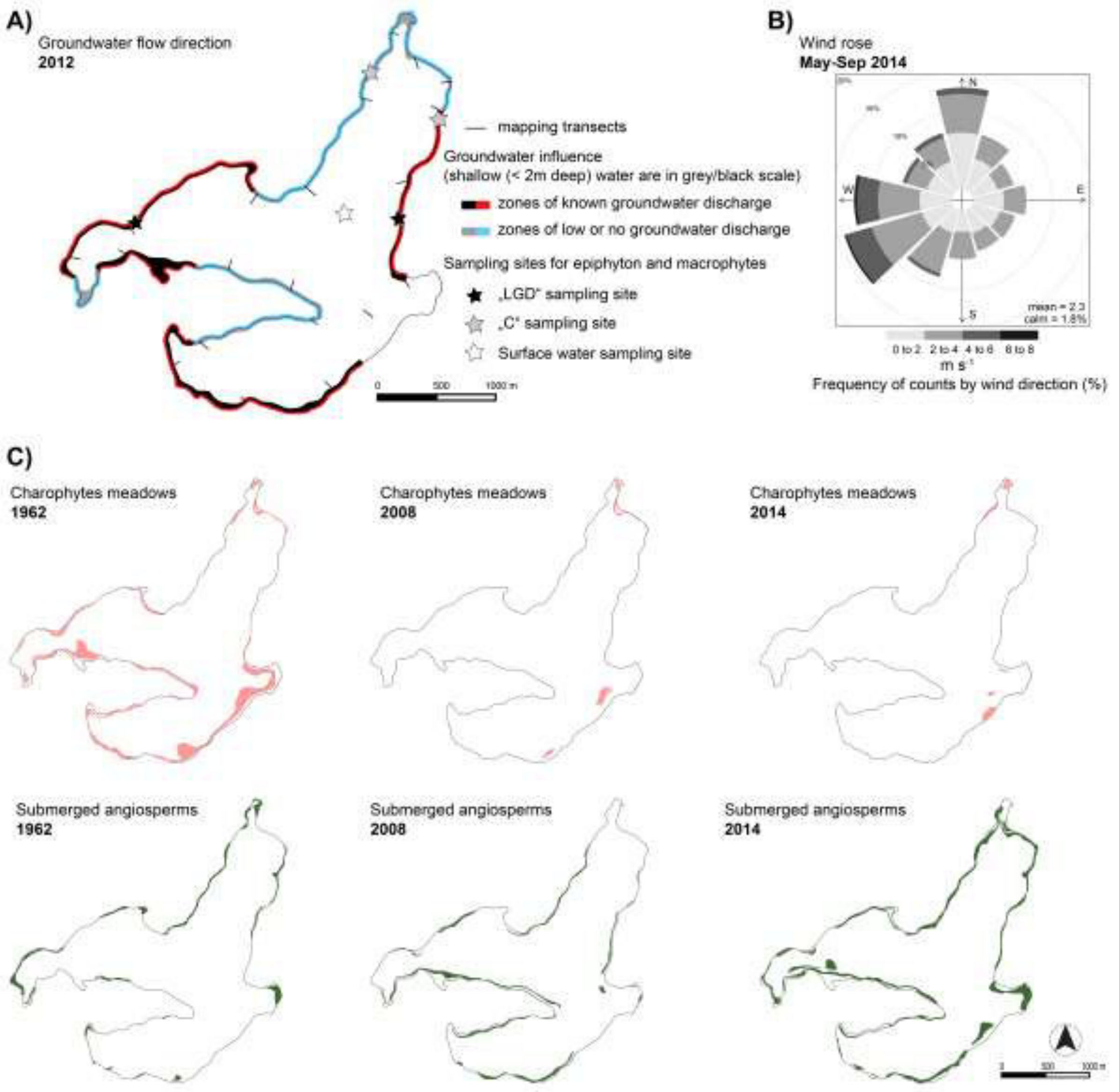

Fig. 1: A) Areas of lacustrine groundwater discharge (LGD), mapped transects and macrophyte and epiphyton sampling points in Lake Stechlin (black stars for LGD sites, grey stars for control sites). B) Wind strength and directions, measured at the shore in summer 2014. C) Distribution of charophyte meadows and angiosperms in shallow littoral areas of Lake Stechlin in 1962, 2008 and 2014.

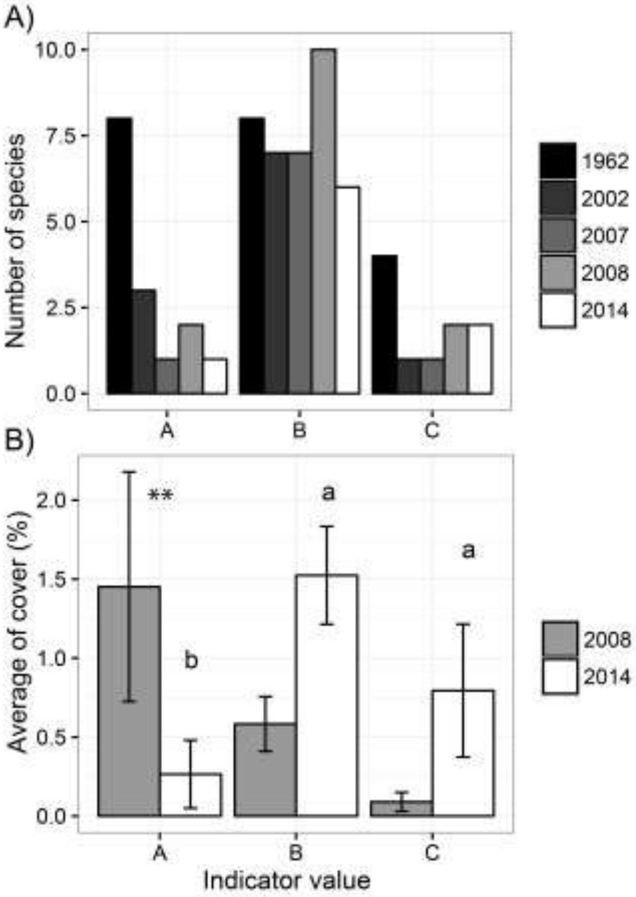

Fig. 2: A) Number of macrophyte species with A, B and C indicator values present in shallow littoral areas (0-2 m depth) of Lake Stechlin during the survey in 1962 (whole lake), and in 2002, 2007, 2008 and 2014 (data from 7 identical transects). B) Average coverage (± standard error) of macrophyte species with A, B and C indicator values observed in 0-2 m depth of 20 transects surveyed in 2008 and 2014. The significant differences between years are indicated (Mann-Whitney-U-test, **: $p<0.01$) and for each year, different letters are attributed to significantly different (Mann-Whitney-U-test, $p<0.05$) coverages. The indicator values are attributed after Schaumburg et al. (2015) for the "TKg13" lake type.

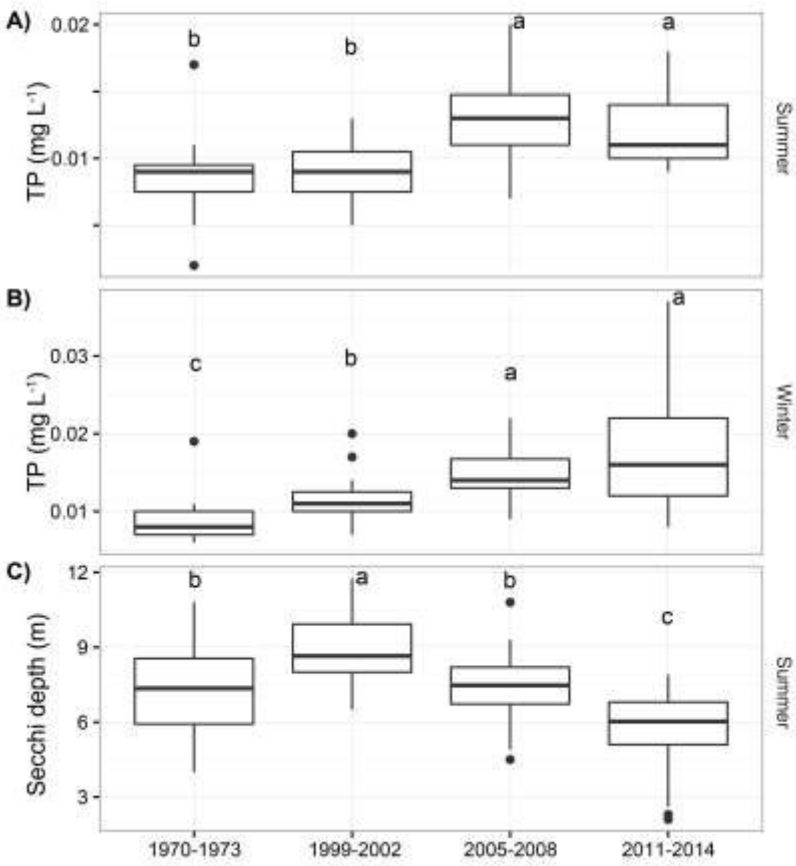

Fig. 3: Characteristics of surface water during the 4 years preceding macrophyte mappings in Lake Stechlin. (A) Total phosphorus (TP) concentrations during the summer months: May to September, (B) TP concentrations during the winter months: October to April, (C) Secchi depth during the summer months. Box-and whisker plots boxes represent first, second and third quartiles, with upper and lower whiskers extending until respectively the highest and the lowest values that are within 1.5 x inter-quartile range. The small letters represent the rank of the datasets, after repeated Mann-Whitney-U-tests.

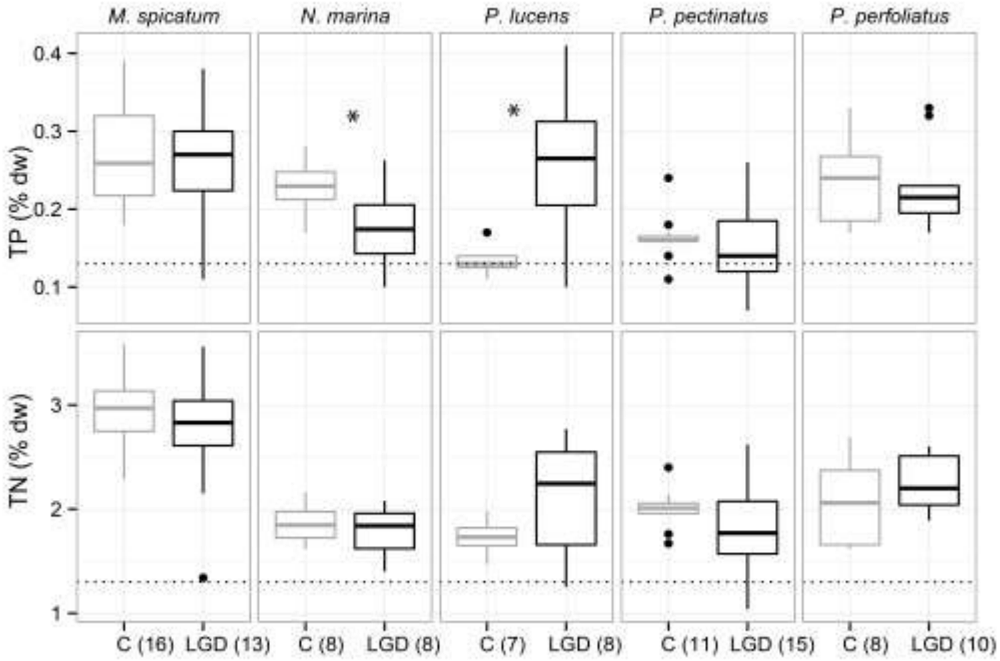

Fig. 4: Nitrogen (N) and phosphorus (P) tissue concentrations in submerged macrophytes at locations with and without lacustrine groundwater discharge (LGD) in Lake Stechlin in 2014. The horizontal lines indicate the threshold of nutrient limitation (Gerloff and Krombholz, 1966). Box-and whisker plots boxes represent first, second and third quartiles, with upper and lower whiskers extending until respectively the highest and the lowest values that are within 1.5 x inter-quartile range. An asterisk (*) is included if the result of the Student's t-test is significant (p<0.05). The number of samples is indicated on the x-axis.

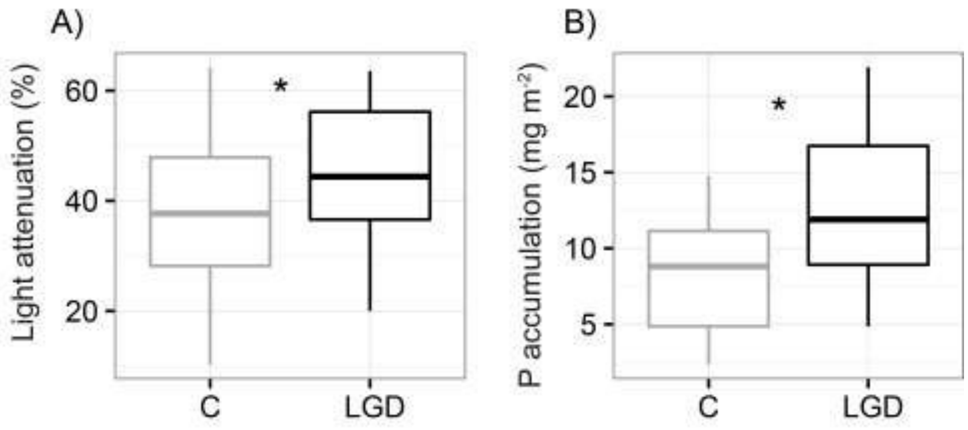

Fig. 5: Light attenuation (A) and phosphorus accumulation (B) in the epiphyton at locations with (LGD) and without (C) lacustrine groundwater discharge in the littoral of Lake Stechlin in summer 2014. Box-and whisker plots boxes represent first, second and third quartiles, with upper and lower whiskers extending until respectively the highest and the lowest values that are within 1.5 x inter-quartile range. The results of Student's t-tests are included (* for $p < 0.05$).

Table 1: Topographical, morphological, hydrological, and chemical parameters of Lake Stechlin (Krey, 1985; IGB, unpublished data).

Parameter	**Mean ± sd**
Drainage basin	12.6 km^2
Forested area in drainage basin	95 %
Maximum depth	69.5 m
Surface area	4.3 km^2
Volume	96.9 x 10^6 m^3
Mean depth	23.3 m
Effective fetch	2 000 m
Water retention time	>40 yrs
Water temperature[3]	19.1 ± 3.1 °C
Secchi transparency[1]	6.4 ± 1.7 m
Calcium[2]	49.6 ± 6.9 mg L^{-1}
Dissolved inorganic carbon[2]	20.6 ± 1.9 mg L^{-1}
NO_3^-nitrogen[2]	16 ± 24 µg L^{-1}
NH_4^+-nitrogen[2]	32 ± 30 µg L^{-1}
Total phosphorus[2]	11 ± 3 µg L^{-1}
Soluble reactive phosphorus[2]	2 ± 1 µg L^{-1}

[1] seasonal average, May-September, 2001-2010

[2] seasonal averages, May-September, 2000-2008, pooled samples, surface, 5m, 10m

[3] seasonal averages, May-September, 2014, pooled samples, surface, 5m, 10m

Table 2: Presence of macrophyte species in transects of the shallow littoral (0-2 m water depth) of Lake Stechlin in 2002, 2007, 2008 and 2014 (whole lake in 1962). Species group according to Schaumburg et al. (2015) at 0 to 1 m depth in lakes from the "TKg13" category. Taxa from group "A" are only abundant at undisturbed sites, "B" taxa are present in both disturbed and undisturbed sites, and "C" taxa only occur at disturbed sites. Red list categories for Brandenburg according to Kabus and Mauersberger (2011) for charophytes and Ristow et al. (2006) for angiosperms (1: critically endangered, 2: endangered, 3: vulnerable, "V": near threatened, *: least concern)

	Species group	Red list category	1962	2002	2007	2008	2014
Charophytes							
Chara aspera	A	2	x				
Chara contraria	B	V	x	x	x	x	
Chara filiformis	A	1	x	x		x	x
Chara globularis	B	*	x		x	x	x
Chara intermedia	A	3	x	x			
Chara rudis	A	2	x				
Chara tomentosa	A	3	x	x	x	x	x
Chara virgata	B	*	x	x		x	
Angiosperms							
Ceratophyllum demersum	C	*	x		x	x	x
Elodea canadensis	C	*	x				
Myriophyllum spicatum	B	V	x	x	x	x	x
Myriophyllum alterniflorum	B	2	x			x	
Najas marina ssp. intermedia	C	3		x	x	x	x
Nuphar lutea	B	*	x				x
Potamogeton crispus	C	*				x	x
Potamogeton filiformis	A	1	x			x	
Potamogeton gramineus	A	2	x				
Potamogeton lucens	B	3	x	x	x	x	x
Potamogeton natans	A	*	x				
Potamogeton pectinatus	B	*	x	x	x	x	x
Potamogeton perfoliatus	B	V		x	x	x	x

Potamogeton pusillus	C	3	x	x		x	x
Ranunculus circinatus	C	3	x			x	x