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The Effect of Charophyte Communities on Phosphorus Flow in Lake Shinji, Japan

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Abstract

Charophytes are a group of aquatic algae similar to vascular plants; they play an important role in the nutrient cycling of lakes. Specifically, under eutrophication, charophytes have a greater capacity for phosphorus accumulation than vascular plants. During their development, charophytes accumulate calcium within their structures, along with dissolved phosphorus from the water column. The calcified structures are deposited onto the lakebed after the death of the plant, with phosphorus co-precipitating with calcium, limiting its return to the water. Lake Shinji is one of many lakes in Japan where the charophyte population is in decline or extinct. Using aerial photographs acquired 70 years ago, we estimate the extent of the historical charophyte community in Lake Shinji, and quantitatively evaluate and examine the extent to which charophytes accumulated phosphorus in the lake sediment. The amount of phosphorus accumulated by the charophyte community in Lake Shinji is estimated to be 0.56–25.5 t P y^{-1}. Charophytes are not found in Lake Shinji today, and although various species of vascular plants have recolonised the lake, no significant improvement in water transparency has been observed. This study concludes that charophytes are likely to be a more effective measure in combating eutrophication than vascular plants.

Keywords: Eutrophication, Charophyte, Diffusive phosphate, Calcite accumulation

1. Introduction

Eutrophication in lakes is caused by an increase in the concentrations of nitrogen and phosphorus in the water column. Although these nutrients are naturally supplied by soils to lake waters, industrial liquid waste from catchment basins, chemical fertilisers used on farms, and grey water from houses can also flow into the lakes, leading to over-nutrition. In particular, when the N/P ratio is lowered by an increase in phosphorus concentrations, the resulting cyanobacterial bloom typically causes drinking water supplied by the lake to have a foul odour (Poste et al., 2013; Ganguly et al., 2013). Although phosphorus is an essential nutrient in lake ecosystems, high concentrations can lead to over-nutrition, resulting in eutrophication and a reduction in water transparency. In general, phosphorus is not only supplied to lakes from the catchment basins, but also from an increase in sediment input during the summer, mainly as a result of high water temperatures and anoxia. Within the sediments, phosphorus bonds to iron and manganese, however under anoxic conditions, it diffuses back into the water along with these elements (Søndergaard et al., 2000). Thus, even if the external load is reduced, the internal load is not necessarily diminished as the phosphorus diffuses upwards from the sediments (Graneli, 1999; Scharf, 1999). Owing to the fact that such diffusion is more marked for phosphorus than it is for nitrogen (Kamiya et al., 1996), measures for reducing eutrophication should aim to limit the input of this diffusive phosphorus.

Submerged plants are particularly susceptible to the effects of eutrophication in lake ecosystems. These aquatic plants play a key role in the maintenance of a high degree of water transparency by preventing resuspension of sediment; the also serve as nesting areas for juvenile fish, and absorb nutrients (Timms and Moss, 1984; Scheffer et al., 1993). Occurring within highly dense communities that are completely underwater, submerged plants are able to absorb nutrients from lake waters (Blindow, 1992a; Kufel and Ozmek, 1994). However, in such communities, the dissolved oxygen concentration is reduced throughout the night-time, and in mature communities, it can also be lower during the day in the layers beneath the canopy (Wetzel, 2001; Carpenter and Lodge, 1986; Frodge et al., 1990; Miranda et al., 2000). The dissolved oxygen concentration in the water directly overlying the lake bottom has been reported to decrease as the biomass of submerged plants increases (Haga et al., 2006). Thus, although the presence of aquatic plants is generally considered to reduce the concentration of nutrients, some nutrients actually diffuse from the sediment into the water under the plant-induced hypoxia. Furthermore, in many lakes with advanced eutrophication, the submerged plant communities have decayed or completely disappeared, due to the reduction in water transparency brought about by eutrophication, as the amount of light required for overgrowth does not reach the lakebed through the dense blooms of phytoplankton (Scheffer, 1989; Hilt et al., 2006). Accordingly, it is difficult to accurately determine the removal of nutrients by submerged plants in eutrophic lakes.

Submerged plants include both vascular plants such as *Potamogeton*, and rootless algae such as charophytes. Charophytes are classified within the family Characeae, within which six genera have been confirmed: *Chara*, *Nitella*, *Tolypella*, *Nitellopsis*, *Lamprothamnium*, and *Lychnothamnus*. Together, charophytes play an important role in the nutrient cycling of lakes (Kufel and Kufel, 2002; Blindow et al., 2002; Rodrigo et al., 2007). In contrast to vascular plants, the nutrient cycles of charophytes have four prominent water column effects (Asaeda and Fujino, 2006), namely: the absorption and accumulation of nutrients in the water; the facilitation of particle sedimentation and the suppression of their subsequent diffusion; the maintenance of the oxygen supply from gyttja stratification that is high in reducibility; and the accumulation of phosphorus due to the abundance of HCO_3^- ions. Notably, the capacity for phosphorus accumulation is considerably greater in charophytes than in vascular plants such as *Potamogeton* (Van den Berg et al., 1998; Kufel and Kufel, 2002; Nõges et al., 2003; Hargeby et al., 2007; Bakker et al., 2010).

In neutral or slightly alkaline aquatic habitats, the main inorganic carbon species is HCO_3^-. Since the substrate for carbon fixation is CO_2, OH^- is inevitably generated during photosynthesis (Prins et al., 1982). In charophyte cells, OH^- occurs in limited areas called alkaline bands. HCO_3^- or CO_2 combines with Ca^{2+} to form carbonate, and environmental Ca^{2+} reacts with biocarbonate or carbonate ions to produce calcium carbonate outside the charophyte cell walls.

During their development, charophytes deposit dissolved calcium in the water and within their algal structures. The rapid production of biomass and calcite encrustation by charophytes allows a large amount of nutrient to be accumulated from the water (Kufel and Kufel, 2002).

Charophytes can be divided into two distinct types: corticate species that have a cortex outside the cell wall, and ecorticate species that do not have such a structure. Generally, calcification is thought to only occur in ecorticate species of *Chara* (García, 1994). However, Kawahata et al. (2013) found that the corticated species can also calcify the entire cortex and cell wall.

When this calcification occurs, phosphorus from the water column is co-precipitated with the calcium, resulting in a reduction of its concentration in the water, which in turn decreases the phosphorus available for phytoplankton, thus maintaining lake transparency (Hilt et al., 2006; Kufel et al., 2013). Calcium and bonded phosphorus cannot be used by phytoplankton, and the bonded phosphorus deposited in the sediment tends to remain there, thanks to its shape, which makes its subsequent diffusion less likely (Ishii et al., 2008; Yokoyama et al., 2008). Additionally, while the phosphorus accumulated by vascular plants is returned to the water after the plant’s death, for charophytes, the phosphorus remains bonded to calcium even after plant death, and is not returned to either the water or the sediment. In an analysis of columnar sediments from Lake Beloye in West Sibera, Krivonogov et al. (2012) observed that the phosphorus concentration of the sediment increased along with the appearance of charophyte oospores and with a sharp decrease in diatoms.

The present study focuses on Lake Shinji in Japan, in which a calcifying type of charophyte once flourished. Using this lake as a case study, we examine whether charophytes are truly effective water purifiers in lakes, by quantitatively analysing the extent to which lakebed diffusion is affected by the accumulation of phosphorus by charophytes. Charophytes were used as fertiliser for agricultural land in the surrounding area when they were flourishing in the studied lake (Hiratsuka et al., 2006). Therefore, it is assumed that the phosphorus bonded within the charophytes in this lake was either removed through harvesting or remains as apatite.

2. Materials and Methods

2.1 Study Area

Lake Shinji is located in the northeastern part of Shimane Prefecture, Japan and is a brackish lake with an area of 79.8 km^2 and a mean depth of 4.5 m (Figure 1). It is classified as a eutrophic lake with water quality in the centre of the lake as follows: TP: 0.053 mg L-^{1}, TN: 0.52 mg L^{-1} (Kamiya et al., 2015a). Lake Shinji is registered under the Ramsar Convention as a Wetland of International Importance, and is maintained as a place of recreation and relaxation for local residents, as well as a tourist attraction. However, in recent years, water quality conservation has become a pressing issue due to eutrophication within the water column, and the accumulation of blue-green algae.

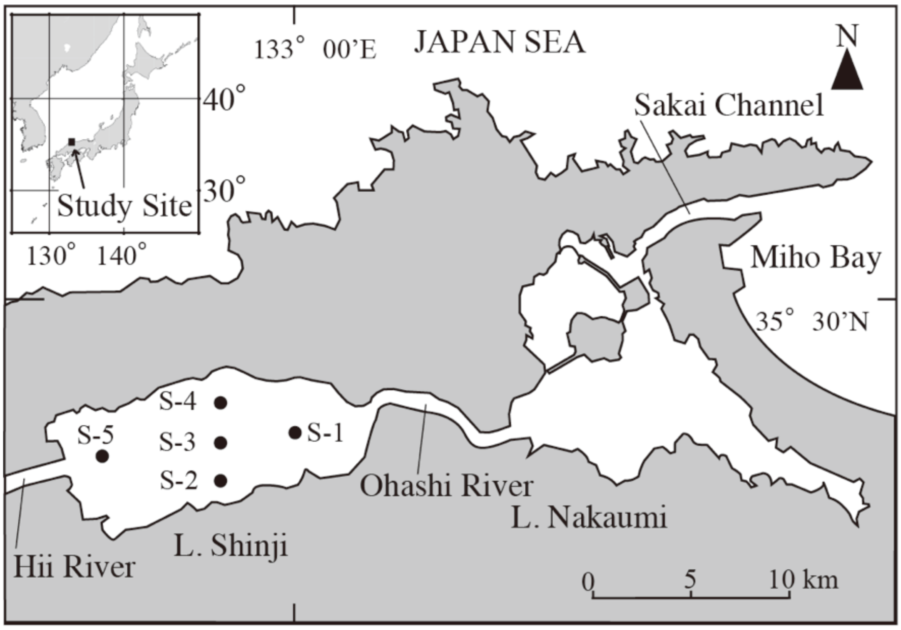

Figure 1. Study site and water sampling points (S-1~S-5)

Notes: Water sampling was performed at S-1, S-2, S-3, S-4 and S-5 by the Shimane Prefectural Institute of Public Health and Environmental Science.

Lake Shinji receives 75% of its freshwater inflow from the Hii River to the west, and outflows to the east into Lake Nakaumi via the Ohashi River. Saline water backflows along the Ohashi River under certain atmospheric pressure conditions, as a result of the nearby Lake Nakaumi being connected to the Sea of Japan. Lake Shinji therefore receives a supply of saline water from Lake Nakaumi via the Ohashi River (Ishitobi et al., 1988, 1993), and thus has an annual average salinity of 3.7 practical salinity units (PSU). Furthermore, at the bottom of the lake, a thin layer of high-salinity water flows from Lake Nakaumi via the Ohashi River. During the warm season, salinity stratification occurs, and dissolved oxygen is consumed by bacteria in the bottom layer, resulting in hypoxia at the lake bed. These conditions favour the diffusion of phosphorus from the sediments (Kamiya et al., 2011). According to Kamiya et al. (2008), the annual phosphorus inflow to the lake is 62.4 t y^{-1}. This is significantly less than the 95.6 t y^{-1} recorded when observations began in 1983, suggesting that the phosphorus inflow to the basin has decreased, mostly likely as a result of improvements to sewage systems and septic tank devices.

Prior to the 1950s, at least five types of charophyte were present among the submerged plants that inhabited Lake Shinji: *Chara braunii* C.C. Gmelin, *Chara corallina* Willdenow, *Chara fibrosa* C. Agardh ex A. Bruzelius, *Chara sp*, and *Nytella hyalina* (De Candolle) C. Agardh. (Kasaki, 1964; Komuro et al., 2016). Among these, *C. corallina* covered the lake bottom as the dominant species (Komuro and Yamamuro, 2013; Komuro et al., 2016). By the 1960s, only two species of charophytes remained, *C. braunii* and *Nitella hyalina* (Kasaki, 1964), and today, none of these species can be confirmed to currently inhabit Lake Shinji. Herbicides were first introduced to Lake Shinji at around this time, leading to a sudden decrease in the number of submerged plants, which in turn facilitated eutrophication, thus resulting in a sudden decrease in the degree of water transparency (Hiratsuka et al., 2006; Yamamuro et al., 2014). Moreover, the dominant species of submerged plant inhabiting the lake in 1961 was not a charophyte, but the submerged macrophyte *Potamogeton malanianus* Miq (Miyadi, 1962). By the 1990s, most of the *P. malaninus* had also disappeared and no aquatic plants remained in Lake Shinji. Since 2009, however, a number of submerged macrophytes, including *Potamogeton crispus* L., have been reported to flourish, and yet there have been no discernible improvements in eutrophication conditions and water transparency (Yamamuro et al., 2014).

Aerial photographs of Lake Shinji acquired 70 years ago reveal a high degree of water transparency, which allows confirmation of lake-bottom conditions (Figure 2). Komuro and Yamamuro (2013) concluded that in 1947, water transparency in Lake Shinji was at its greatest, exceeding a depth of 4 m. In the aerial photograph taken in October of 1947, it was noted that submerged aquatic plants did not grow in the shallowest (0–0.5 m) and deepest areas (>3 m). This photograph was taken during October, therefore we assume that the majority of the submerged macrophytes were mowed and harvested during the summer, for use as fertilizer. Charophytes can thrive in water depths that are approximately the same as the transparency depth (Yoshimura, 1938; Middle and Markager, 1997), and so we consider it likely that the charophytes covered the lake bottom, and that mowing may be performed to a depth of 4 m. In this study, we used aerial photographs taken in 1947 to confirm that a charophyte community existed at water depths of 0.5–3 m, and up to 4 m in one area (Komuro and Yamamuro, 2013). We consider it highly likely that charophytes did not grow in areas with water depths of 0–0.5 m, due to the agitating effects of waves; and their extent could not be confirmed from the aerial photographs in such regions, as they were covered with meadows. Thus, to avoiding overestimation, we restricted the possible habitation depth of charophytes to 0.5–3 m.

2.2 Amount of Phosphorus Accumulated by the Charophyte Community

In order to investigate the phosphorus accumulation capacity by the charophyte community in Lake Shinji, we examined the amount of phosphorous accumulated by communities within two distinct regions: (i) an area of 2.95 km^2 covered by aerial photographs taken by the U.S. Armed Forces in October of 1947 (Komuro and Yamamuro, 2013) (Figure 2); and (ii) the total lake area in which water depths range from 0.5 m to 3 m. The areal extent of (ii) was calculated from a polygon created on the basis of the end member water depths, using ArcGIS 10 (ESRI Inc.) and a bathymetry map surveyed between 1962 and 1963 by the Geographical Survey Institute (now the Geospatial Information Authority of Japan). Thus, the charted lake area between the water depths of 0.5 m and 3 m, forming community (ii), totalled 15.02 km^2.

Both submerged macrophytes (vascular plants) and charophytes use phosphate to aid in their growth. The accumulation of phosphate by 1 m^{-2} of a charophyte community has been previously reported in the literature to be 0.5–1.7 gP m^{-2} (Pereya-Ramos, 1981), 0.4 g P m^{-2} (Blindow, 1992b), and 0.19 gP m^{-2} (Królikowska, 1997). In this study, we calculated the amount of phosphorus that the charophyte communities could absorb using the minimum and maximum published values, for community areas (i) and (ii).

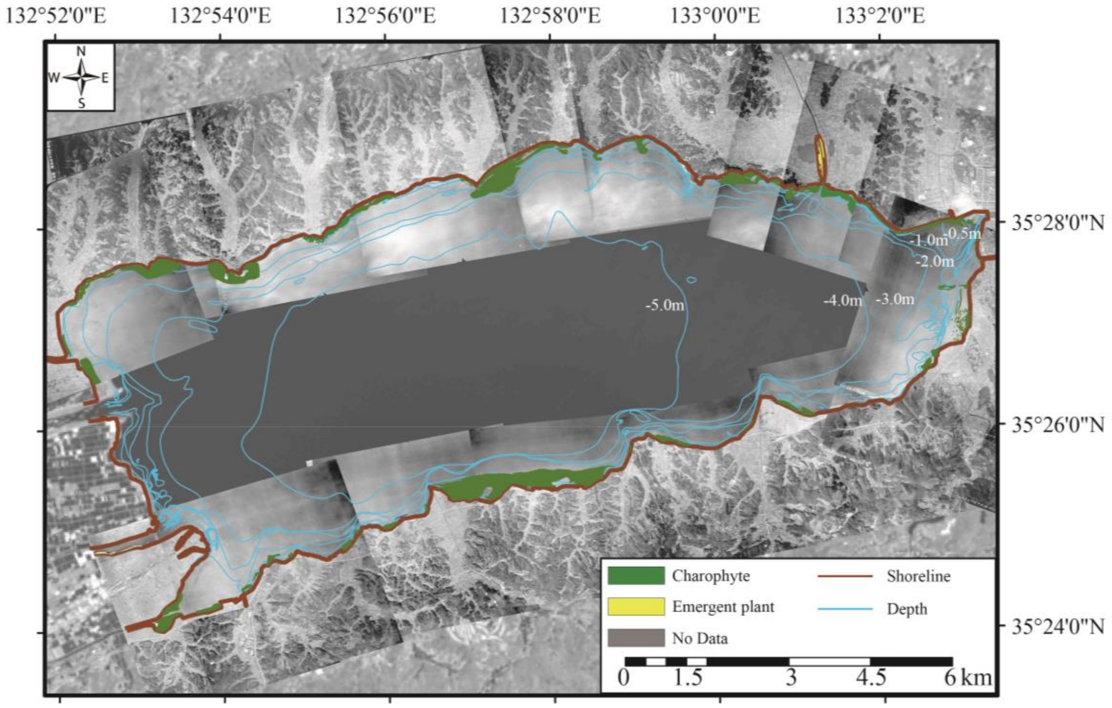

Figure 2. Distribution of the charophyte community at Lake Shinji in 1947 (Komuro and Yamamuro., 2013)

Notes: The textured part is a Landsat-8 picture taken in 2013. The brown border demarcates the lake. The green area is charophyte community and the yellow is an emergent plant community. Community area was detected by aerial photographic interpretation on ArcGIS.

2.3 Calculation of Diffusive TP and Phosphate from Sediment

In order to calculate the amount of total phosphorus (TP) released from the sediments, we used the formulation in Equation 1 (Kamiya et al., 2015b), in addition to the L-Q equation (Equation 2) which provides the inflow load L, needed to calculate the amount of diffusion (Miyazako et al., 2014).

In the case of Lake Shinji, land water (Q_0) flows into the lake between the measurement days of one month and the next, and an inflow load (L_s) is imposed on the lake. The TP concentration in Lake Shinji during a particular month is defined as C_{sn}, and one month later as C_{sn+1}. Likewise, subsequent monthly TP concentrations in Lake Nakaumi are defined as C_{nn} and C_{nn+1}. The mean TP concentration of water flowing out of Lake Shinji and into Lake Nakaumi over one month was calculated as the mean of C_{sn} and C_{sn+1}, whereas the TP concentrations of water flowing back to Lake Shinji from Lake Nakaumi is the mean of C_{nn} and C_{nn+1}. Thus, the amount of TP released from the bottom of Lake Shinji (A_{sn+1}: kg month^{-1}) can be calculated as follows:

$$A_{sn+1}=V_s\,(C_{sn+1} - C_{sn}) + Q_2 * (C_{sn+1} + C_{sn})/2 - Q_1 * (C_{nn+1} + C_{nn})/2 - L_s \qquad (1)$$

where Vs is the volume of Lake Shinji (m^3), Q_1 is the monthly backflow water volume (m^3 month^{-1}) from Lake Nakaumi to Lake Shinji, Q_2 is the monthly water volume (m^3 month^{-1}) flowing from Lake Shinji to Lake Nakaumi, C_{sn} and C_{sn+1} are the TP concentrations (mg L^{-1}) of Lake Shinji during one month and the following month, respectively, C_{nn} and C_{nn+1} are the

TP concentrations (mg L^{-1}) of Lake Nakumi during one month and the following month, respectively, and L_s is the load inflow to Lake Shinji (kg $month^{-1}$). TP values were measured at five stations from 1985-2013, S-1–S-5 in Figure 1, by the Shimane Prefectural Institute of Public Health and Environmental Science (SPIPHES). The TP for each month is the average of the values obtained at S-1–S-5. Water was sampled from the bottom. To determine TP, SPIPHES added 10 ml of a potassium persulfate solution (40 g L^{-1}) to a 50-ml sample; this was then decomposed by autoclaving at 120℃ for 30 min. The mixture was then measured and analysed with an autoanalyzer (Kamiya et al., 2011).

The inflow load to Lake Shinji can be expressed using Miyasako et al.'s (2014) quadratic L-Q equation as follows:

$$\text{Log } L = 0.34(\text{Log}F)^2 + 0.25\text{Log}F - 5.60 \tag{2}$$

where L is the TP load (kg s^{-1}), and F is the flow (m^3 s^{-1}).

In addition, we calculated the ratio of bottom phosphate (PO_4-P) to bottom TP using data from 1985–2013, and used the ratio of diffusive TP (D-TP) to estimate diffusive phosphate (D-P) in 1985–2013. The methods of obtaining these data are described below. We then created a regression for D-TP and D-P in 1985–2010, and confirmed the relationship. We inserted the D-TP calculated using the following methods into the regression (power approximation), and calculated the D-P in 1929 and 1961–1962.

In order to calculate the amount of D-P supplied to the lake when it was inhabited by charophytes, we first created a regression for 28 years (1985–2013) of surface layer TP and bottom layer TP, and then confirmed the relationship. Transparency, as well as surface and bottom TP values were observed at S-1–S-5 from 1985 to 2013. We calculated the average of S-1–S-5 for each month, and then calculated an annual average. Next, we created a regression for 28 years (1985–2013) of water transparency and surface layer TP, and calculated the historical surface layer TP by inserting into this equation with the measured degrees of water transparency in 1929 (Yoshimura 1938), 1947 (Komuro and Yamamuro, 2013), and 1961−1962 (Miyadi, 1962). Only one measurement of transparency was available from 1929 and 1947, and so we assume that this value represented the annual average for those years.

We then inserted the degrees of water transparency from each time period into the previously created regression for surface layer TP and bottom layer TP, and calculated the bottom layer TP. We used this estimated bottom TP as an annual average of conditions each year. In order to estimate the past TP diffusion from the sediment, we created a regression for the amount of phosphorus diffusion as calculated in Equation 1, and the annual average bottom layer TP. Using the past diffusion calculated from this regression, we determined the maximum, minimum, and average amounts of phosphorus diffusion that was accumulated by the charophyte community over the 28 years in community areas (i) and (ii).

3. Results

3.1 Amount of Phosphate Accumulated by the Charophyte Community

We calculated the amount of phosphate accumulation for each of the two community areas: (i) the charophyte community identified from aerial photographs with surface area of 2.95 km^2; and (ii) the surface areas obtained from charting depths of 0.5−3 m on the bathymetry map. The following results were obtained: (i) 0.56−5.02 tP; and (ii) 2.85−25.53 tP (Table 1). Based on these results, we estimate that the charophyte community of Lake Shinji once accumulated between 0.56 and 25.53 tP.

Table 1. Amount of phosphate accumulation by charophytes in the two community areas

Area Type	Area (m^2)	Amount of accumulation Phosphate (tP)	
		0.19 $gP\ m^{-2}$	1.7 $gP\ m^{-2}$
(i) Photo Area	2,950,000	0.56	5.02
(ii) 0.5 m–3 m	15,018,675	2.85	25.53

3.2 Calculation of Diffusive Phosphorus from Sediments.

Based on the results of our calculation, the minimum D-TP from 1985 to 2013 was 4.40 $t\ y^{-1}$, while the maximum was 65.40 $t\ y^{-1}$ (Figure 3). Additionally, the minimum D-P was 0.31 $t\ y^{-1}$, while the average was 7.14 $t\ y^{-1}$ (Figure 4; Table 2). Although some fluctuation in the amount of diffusion was observed, an overall trend of gradual increase prevailed during this period. A significant correlation was observed between the calculated amounts of diffusion and bottom layer TP (Figure 4), and we obtained the following regression (Equation 3):

$$\text{Diffusive TP} = 929.14\ \text{Bottom TP} - 15.383\ (R^2 = 0.50,\ p < 0.001,\ n = 28) \tag{3}$$

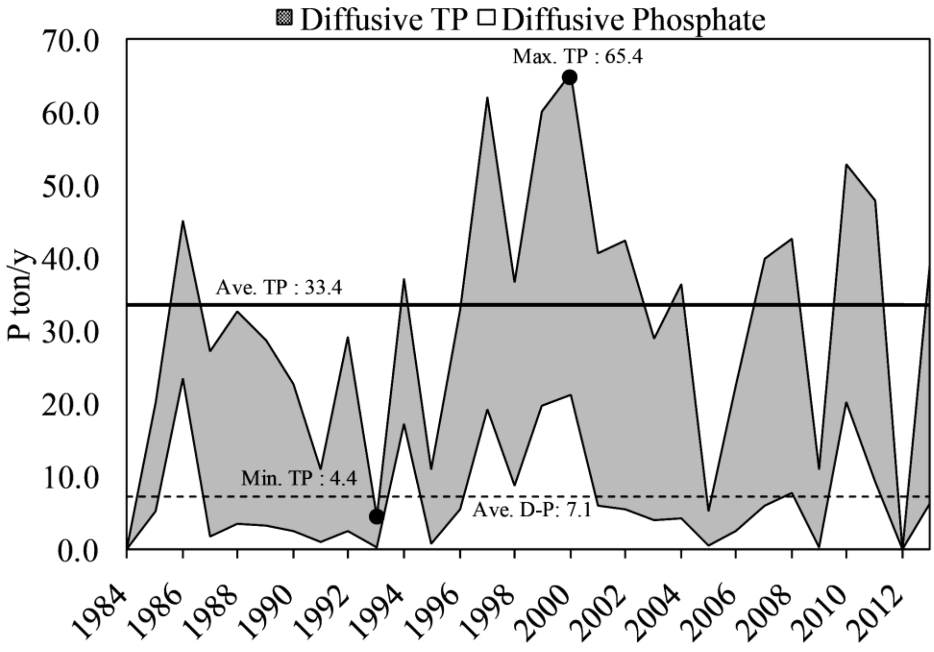

Figure 3. Amount of diffusive phosphorus between 1985 and 2013

Table 2. Minimum, Maximum, and Average amounts of D-TP and D-P on the basis of the degrees of water transparency

1985–2013	D-TP ($t\ y^{-1}$)	D-P ($t\ y^{-1}$)
Minimum	4.40	0.31
Maximum	65.40	23.37
Average	33.38	7.14

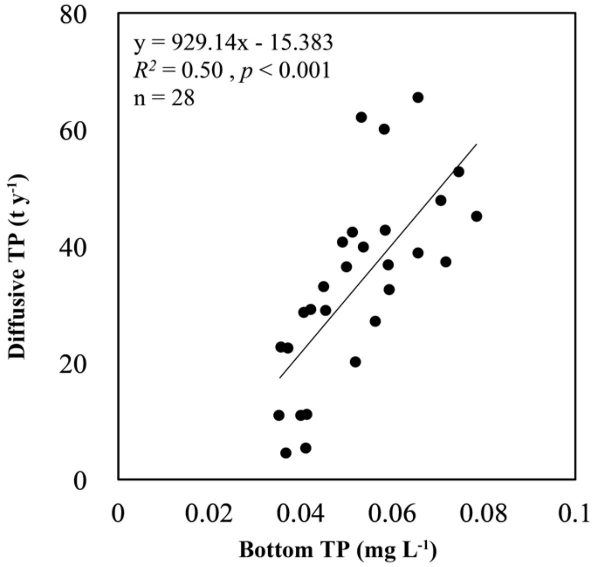

Figure 4. Relationship between diffusive TP and bottom TP

Note: The regression in Equation 3 derived from the correlation is shown here.

Using equation 3, we created a regression for the surface layer TP and bottom layer TP, in order to calculate the amount of diffusion at the time of charophyte inhabitation (Equation 4, Figure 5). We used the annual average surface and bottom TP values for the calculation.

$$\text{Bottom TP} = 1.0365 \text{ Surface TP} + 0.0061 \ (R^2 = 0.64, p < 0.001, \text{n} = 28) \qquad (4)$$

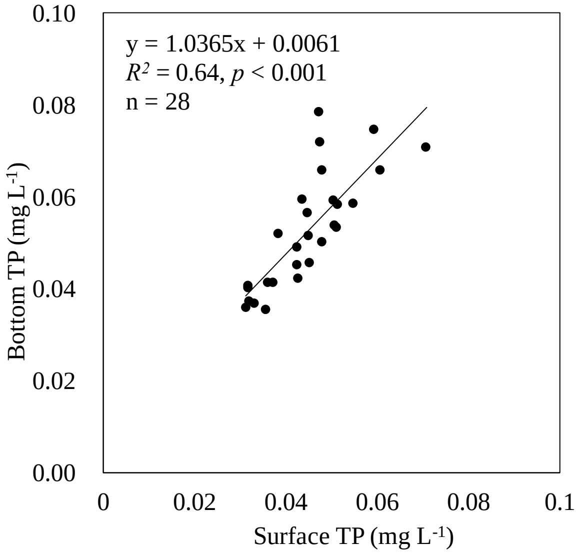

Figure 5. Relationship between surface TP and bottom TP

Note: The regression in Equation 4 derived from the correlation is shown here.

Next, in order to calculate past TP, we created a regression linking TP and the degree of water transparency (Equation 5, Figure 6). We used the annual average TP and transparency values for the calculation.

$$\mathrm{TP} = -0.035\ \mathrm{Log}\ (\mathrm{Tra}) + 0.0554\ (R^2 = 0.31,\ p = 0.002,\ \mathrm{n} = 26) \quad (5)$$

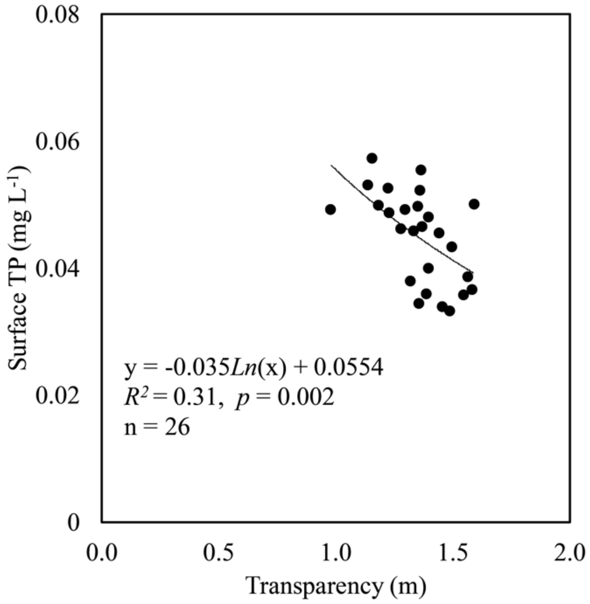

Figure 6. Relationship between average annual surface TP and transparency

Note: The regression in Equation 5 derived from the correlation is shown here

Table 3 shows the results for historical bottom layer TP, calculated using equations 4 and 5. In 1929, the TP in the surface and bottom layers were 0.02 mg L^{-1} and 0.03 mg L^{-1}, respectively; in 1947, 0.007 mg L^{-1} and 0.011 mg L^{-1}, respectively; and from 1961 to 1962, 0.04–0.05 mg L^{-1} and 0.05–0.06 mg L^{-1}, respectively.

Table 3. Estimation of total phosphorus on the basis of the degree of water transparency

Observation Year	Transparency (m)	Amount of TP (mg L^{-1})	
	Obs	Cal Surface	Bottom
1929 (Yoshimura, 1938)	2.7	0.02	0.03
1947 (Komuro and Yamamuro, 2013)	4.0	0.007	0.011
1961–1962 (Miyadi, 1962)	1.1 – 1.6	0.04 – 0.05	0.05 – 0.06
Average of 1984–2011	1.3	0.045	0.052

Notes: Obs: Observed data; Cal: Calculated data.

Table 4 shows the results for D-TP and D-P in 1929, 1947 and 1961–1962. We calculated the D-P using the following equation.

$$\text{D-P} = 0.0146 * \text{D-TP}^{1.6866}\ (R^2 = 0.85,\ \mathrm{n} = 25) \quad (6)$$

Equation 6 was calculated from D-TP and D-P from 1985 to 2010. We inserted the values of D-TP in 1929 and 1961–1962, which were calculated based on transparency, into equation 6 to estimate D-P. However, due to the fact that TP did not diffuse from the sediment, but was accumulated into the sediment in 1947 (Table 4), we did not calculate the diffused phosphate (D-P) for this year. As a result, D-P was found to be 2.4 t y^{-1} in 1929 and 5.9–7.6 t y^{-1} in 1961–1962.

Table 4. Estimation of D-TP and D-P on the basis of the degree of water transparency in 1929, 1947 and 1961–1962

Observation Year	D-TP (t y $^{-1}$)	D-P (t y $^{-1}$)
1929	12.49	2.36
1947	-5.16	-
1961–1962	31.07–40.36	5.88–7.63

4. Discussion

Previous studies into phosphorus accumulation by charophytes (Kufel et al., 2013; Kufel et al., 2016) have not yet investigated the effects of accumulation on the amount of diffusion from the sediment. The present study is the first to quantitatively demonstrate that, in places where the lake bottom is covered by charophytes, the amount of accumulation is equivalent to the amount of phosphorus diffused from the sediment.

We calculate the rate of charophyte phosphate accumulation of diffused phosphate (D-P), using the two charophyte community areas (i) and (ii). Area (i) is 2.95 km^2, representing the charophyte communities present in 1947, while area (ii) is 15.02 km^2, representing a theoretically plausible range based on habitable depths. In addition, we used the minimum and maximum amounts of accumulation (gP m^{-2}) from previous reports: 0.19 gP m^{-2} (Królikowska, 1997) and 1.7 gP m^{-2} (Pereya-Ramos, 1981).

The amounts of D-P in 1929 and 1961–1962, when charophytes still existed in the lake, were 1.03 t y^{-1} and 4.59–6.94 t y^{-1}. In 1929 when charophytes existed in Lake Shinji, area (i) ×0.19 gP m^2 equals 54% of the D-P and (i) ×1.7 gP m^2 equals 487% of the D-P. For area (ii), the charophyte accumulation rate becomes consistently >100%. Figure 7 shows the charophyte accumulation rate for maximum D-P in 1961–1962 when the charophytes had begun to decline. For this period, (i) ×0.19 gP m^2 gives an accumulation rate of 8% and (i) ×1.7 gP m^2 equals 72%. In the case of area (ii) ×1.7 gP m^2, D-P accumulation by charophytes was still >100%. These estimations suggest that the amount of accumulation by charophytes may have exceeded the amount of diffusion before the charophytes disappeared from the lake, as at that time, D-TP was at the same level as today. We did not perform these calculations for 1947 because the value for diffusion was negative, implying that phosphorus sinks into the sediment from the water column (Table 4). Such a situation is possible if the bottom layer did not become anoxic and so no diffusion occurred. Alternatively, charophytes may have accumulated phosphate from the bottom waters, which could have also led to a negative value for diffusion.

Furthermore, we calculated the D-P accumulation rate by charophytes during1985–2013, when the charophytes had disappeared from the lake. The amount of D-TP and D-P released from the sediment before the charophyte loss was 12.49 t y^{-1} and 2.36 t y^{-1}, respectively, but after the 1960s when eutrophication occurred in the lake, it was a maximum of 65.40 t y^{-1} and

23.40 t y^{-1}, respectively. Figure 8 shows the charophyte accumulation rate for maximum D-P (23.40 t y^{-1}) from 1985 to 2013. Here, we estimated four patterns of accumulation using the two community areas, and calculations for the maximum diffusion over a 28-year period. From the results, we determined that the overall phosphorus accumulation capacities in community areas (i) and (ii) were 0.56–5.02 t P and 2.85–25.53 t P, respectively (Table 1). The effects of charophyte accumulation on the maximum D-P in Lake Shinji are (i): 2%–21%, and (ii): >12% from 1985 to 2013. Komuro and Yamamuro (2013) revealed that the lakebed has previously been inhabited by charophytes, and therefore it is conceivable that their presence did have an effect on community surface area (i).

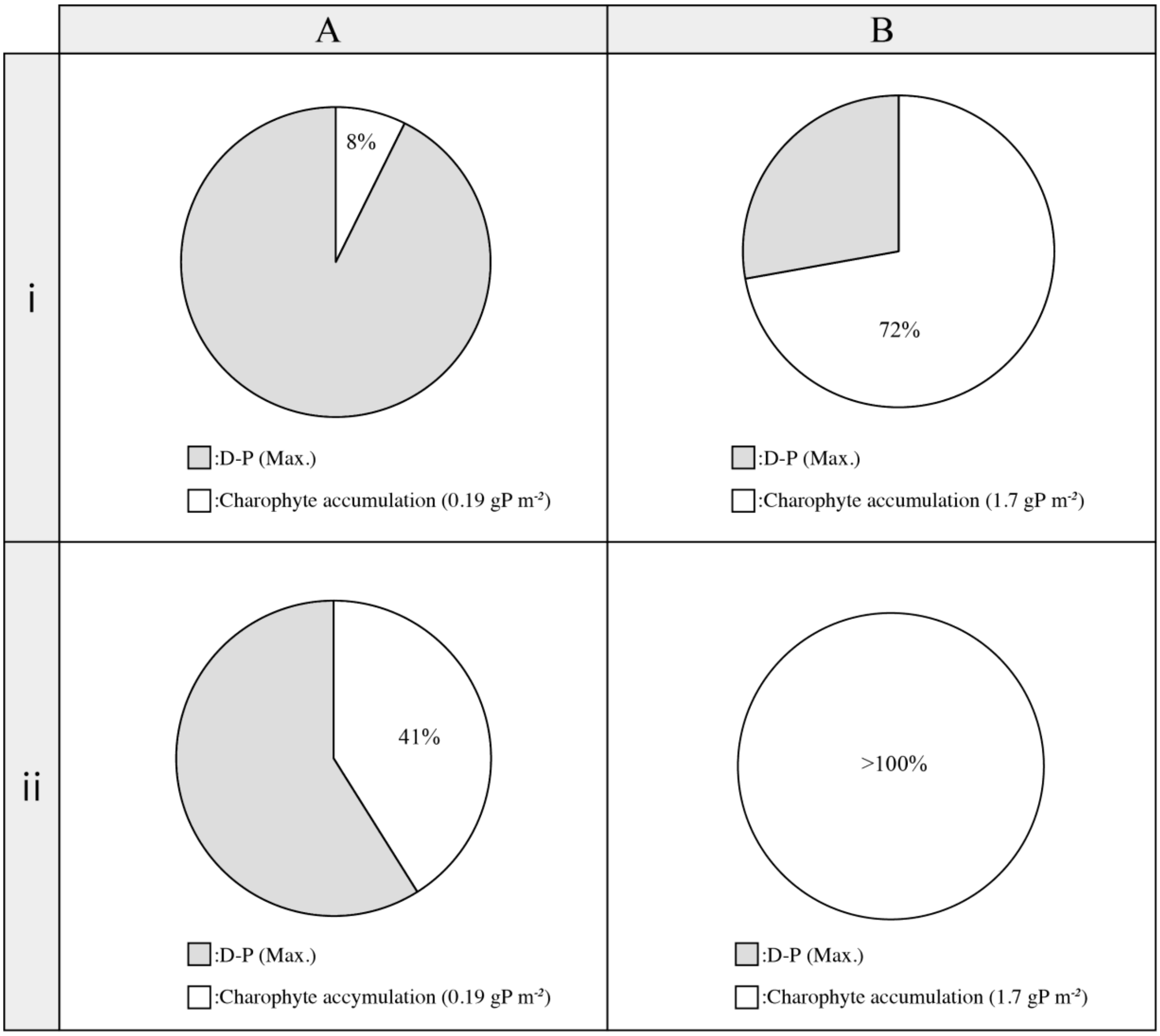

Figure 7. The ratio of charophyte accumulation to D-P during 1961–1962

Notes: The graph shows maximum D-P (grey) and charophyte accumulation (white) in 1961–1962, in two community areas: the photographed area, equivalent to area (i) in the text (2,950,000 m^2), and the area between water depths 0.5–3.0 m, equivalent to area (ii) in the text (15,018,675 m^2). (A) shows the amount of phosphorus accumulation at a rate of 0.19 gP m^{-2}, while (B) shows the amount of phosphorus accumulation at a rate of 1.7 gP m^{-2}.

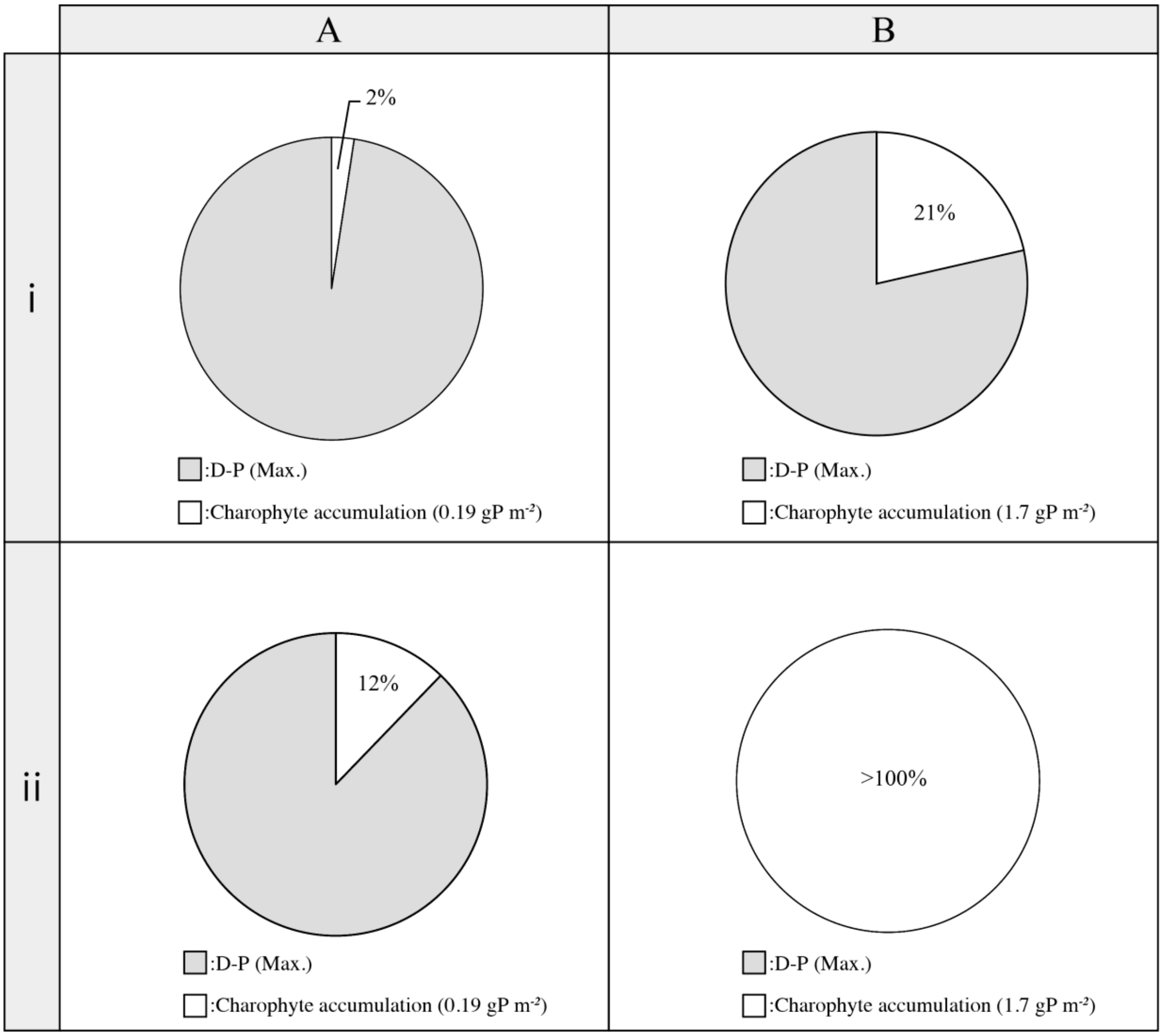

Figure 8. The ratio of charophyte accumulation to maximum D-P from 1985 to 2013

Notes: The graph shows a maximum D-P (grey) and charophyte accumulation (white) from 1985 until 2013, in two community areas: the photographed area, equivalent to area (i) in the text (2,950,000 m^2), and the area between water depths 0.5–3.0 m, equivalent to area (ii) in the text (15,018,675 m^2). (A) shows the amount of phosphorus accumulation at a rate of 0.19 gP m^{-2}, while (B) shows the amount of phosphorus accumulation at a rate of 1.7 gP m^{-2}.

The amount of the D-TP released from the sediment in modern times was similar to when charophytes were extant in Lake Shinji. This suggests that increase of the phytoplankton, and therefore the decrease in transparency, was not induced by an increase in diffusive phosphorus but by the disappearance of the charophytes.

Phosphorus diffused from the lakebed hinders the amelioration of eutrophication. Within the sediments, phosphorus bonds with iron and manganese. When the bottom layer of the lake becomes anoxic, phosphorus is released from the iron and manganese and diffuses upwards (Søndergaard et al., 2000). Therefore, it can be argued that the higher the concentration of iron, the larger the diffusion suppression (Hupfer and Lewandowski, 2008). Accordingly, this suppressed diffusion suggests that the dispersal of iron and nitric acid may be an effective mitigating measure (Søndergaard et al., 2000; Søndergaard et al., 2003; Immers et al., 2015), and the dispersal of iron has not been found to directly affect charophytes (Immers et al., 2013). However, iron diffused from the sediment has recently been linked to the outbreak of cyanobacteria (Molot et al., 2014; Orihel et al., 2015), and therefore the dispersal of iron cannot be considered a valid measure against eutrophication.

In contrast to other submerged plant types, charophytes are able to form dense communities, and flourish when they entirely cover the lake bottom. As the community increases in size, the algae deliver oxygen to the anaerobic layers of lake bottom sediments, forming an oxygen gradient that may inhibit the release of iron-bound phosphorus from the sediment and promote nitrification/denitrification-related nitrogen losses (Lijklema, 1994; Weisner et al., 1994). Moreover, highly dense charophyte communities also suppress the resuspension of bottom sediments, which in turn traps nutrients that would otherwise be consumed by planktonic algae (Kufel and Kufel, 2002). After death, charophytes form a highly viscous substance called gyttja, and through their fixation on the lake bottom, are considered to be an effective measure against the progression of eutrophication.

In general, the decline of submerged plants in eutrophic lakes is caused by a decrease in light penetration due to phytoplankton covering the water surface (Phillips et al., 1978). In contrast to vascular plants, charophytes can grow in deeper waters with lower light levels (Middelboe and Markager, 1997), and therefore the reduction in light penetration is not necessarily the cause of decline or the reason that the submerged plants cannot be revived.

In Japan, the use of herbicides in paddy fields is consistent with the decay of lake stoneworts and aquatic plants in rice paddy areas (Yamamuro et al., 2014). In the past, charophytes have been collected from Lake Shinji to be used as fertiliser, but since paddy field herbicides were introduced in the late 1950s, there has been a sudden decline in the submerged plant population (Hiratuka et al., 2006). Pentachlorophenol (PCP) was registered as a germicide in 1955, and as herbicide in 1956, but was later found to be highly toxic, so its use was restricted in 1963, and from 1972 there was a sharp decline in its production and import. By 1990 all PCP in Japan had expired (Uemura et al., 2002). In an experiment where PCP was applied to *C. braunii*, obstruction to its photosystems was observed at a dose range of 10^{-7}–10^{-6} M, and subsequent plant death was reported (Sato and Shiraiwa, 2012). Furthermore, when compared to other herbicides, these fatal effects occurred at a very low concentration. Accordingly, as the decline of charophytes in lakes coincided with PCP use in Japan, PCP has been implicated as a causal candidate for the population decline. In addition, owing to the fact that even today charophyte growth is limited in areas where there are no effects from herbicides, there is a high probability that herbicides act to continually suppress the growth of charophytes.

According to hysteresis, if the nutrient concentrations present at the beginning of the decline of the submerged vascular plants were to decrease further, then the aquatic plants would be able to restore (Scheffer et al., 2001). However, charophytes flourish in paddy fields abundant in nutrients (Pinke et al., 2014), and therefore even if the nutrient concentration remained unchanged, if herbicide use were reduced to the extent that it no longer affected charophytes, or if substances that prevented withering were developed, it would be possible to suppress phosphorus diffusion in lakes through the restoration of charophyte communities. Therefore, we consider charophytes to be superior to vascular plants as a measure against eutrophication and its effects on submerged plants.

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References

Asaeda, T., & Fujino, T. 2006. Effect of *Charophytes* on Nutrient Cycling in Lake. *The Science and Engineering Reports of Saitama University*, *39*, 152-165

Bakker, E. S., Van Donk, E., Declerck, S. A. J., Helmsing, N. R., Hidding, B., & Nolet, B. A. 2010. Effect of macrophyte community composition and nutrient enrichment on plant biomass and algal blooms. *Basic and Applied Ecol.*, *11*, 432-439. https://doi.org/10.1016/j.baae.2010.06.005

Blindow, I. 1992a. Long-and short-term dynamics of submerged macrophytes in two shallow eutrophic lakes. *Freshwater Biol.*, *28*, 15-27. https://doi.org/10.1111/j.1365-2427.1992.tb00558.x

Blindow, I. 1992b. Decline of charophytes during eutrophication: comparison with angiosperms. *Freshwater Biol.*, 28, 9-14. https://doi.org/10.1111/j.1365-2427.1992.tb00557.x

Blindow, I., Hargeby, A., & Andersson, G. 2002. Seasonal changes of mechanisms maintaining clear water in a shallow lake with abundant Chara vegetation. *Aquat. Bot.*, *72*, 315-334. https://doi.org/10.1016/S0304-3770(01)00208-X

Carpenter, S. R., & Lodge, D. M. 1986. Effects of submerged macrophytes on ecosystem processes., *Aquat. Bot.*, *16*, 341-370. https://doi.org/10.1016/0304-3770(86)90031-8

Frodge, D. J., Thomas, G. L., & Pauley, G. B. 1990. Effects of canopy formation by floating and submerged aquatic macrophytes on the water quality of two shallow Pacific northwest lakes. *Aquat. Bot.*, *38*, 231-248. https://doi.org/10.1016/0304-3770(90)90008-9

Ganguly, D., Robin, R. D., Vardhan, K. V., Muduli, P. R., Abhilash, K. R., Patra, S., & Subramanian, B. R. 2013. Variable response of two tropical phytoplankton species at different salinity and nutrient condition. *J. Exp. Mar. Biol. Ecol.*, *440*, 244-249. https://doi.org/10.1016/j.jembe.2013.01.008

García, A. 1994. Charophyta: their use in paleolimnology. *J Paleolimnol.*, *10*, 43-52.

Haga, H., Ashiya, M., Ohtsuka, T., Matsuda, M., Tuji A., Baba, K., Numahata, S., & Yamane, T. 2006. Relationship between dissolved oxygen concentration of bottom water and macrophyte biomass in the southern basin of Lake Biwa, Japan. *Jpn. J. Limnol.*, *67*, 23-27. https://doi.org/10.3739/rikusui.67.23

Hargeby, A., Blindow, I., & Andersson, G. (2007). Long-term patterns of shifts between clear and turbid states in Lake Krankesjön and lake Tåkern. *Ecosystems.*, *10*, 28-35. https://doi.org/10.1007/s10021-006-9008-5

Hilt, S., Gross, E. M., Hupfer, M., Morscheid, H., Mahlmann, J., Melzer, A., Poltz, J., Sandrock, S., Scharf, E-M., Schneider, S., & van de Weyer, K. 2006. Restoration of submerged vegetation in shallow eutrophic lakes- A guideline and state of the art in Germany. *Limnologica-Ecology and Management of Inland Waters*, *36*(3), 155-171.

https://doi.org/10.1016/j.limno.2006.06.001

Hiratsuka, J., Yamamuro, M., & Ishitobi, Y. 2006. Satoumi Moku tori monogatari. (The story of harvesting submerged aquatic plant), Seibutsu Kennkyu-Sha, Tokyo, 141 p.

Hupfer, M., & Lewandowski, J. 2008. Oxygen controls the phosphorus release from lake sediments: A long - lasting paradigm in limnology. *Int. Rev. Hydrobio*., *93*(4-5), 415-432. https://doi.org/10.1002/iroh.200711054

Immers, A. K., Bakker, E. S., Van, Donk E., Ter Heerdt, G. N. J., Geurts, J. J. M., & Declerck, S. A. J. 2015. Fighting internal phosphorus loading: An evaluation of the large-scale application of gradual Fe-addition to a shallow peat lake. *Ecol. Eng*., *83*, 78-89. https://doi.org/10.1016/j.ecoleng.2015.05.034

Immers, A. K., van der Sande, M. T., Van der Zande, R. M., Geurts, J. J. M., Van Donk, E., & Bakker, E. S. 2013. Iron addition as a shallow lake restoration measure: impacts oncharophyte growth. *Hydrobiologia*., *710*(1), 241-251. https://doi.org/10.1007/s10750-011-0995-7

Ishii, Y., Komatsu, N., Harigae, S., Yabe, T., Watanabe, K., Negisi, M., & Iwasaki, J. 2008. Characteristics of fractional phosphorus distribution in sediment of fish farming area at Lake Kasumigaura, Japan. *Jpn. J. Nippon Suisan Gakkaishi*., *74*(4), 607-614. https://doi.org/10.2331/suisan.74.607

Ishitobi, Y., Kamiya, H., & Itogawa, H. 1993. Tidal, meteorological and hydrological effects on the water level variation in a lagoon, Lake Shinji. *Jpn. J. Limnol*., *54*, 69-79. https://doi.org/10.3739/rikusui.54.69

Ishitobi, Y., Kawatsu, M., Kamiya, H., Hayashi, K., & Esumi, H. 1988. Estimation of Water Quality and Nutrient Loads in the Hii River by Semi-Daily Sampling. *Jpn. J. Limnol*., *49*(1), 11-17. https://doi.org/10.3739/rikusui.49.11

Kamiya, H., Ishitobi, Y., Inoue, T., Nakamura, Y., & Yamamuro, M. 1996. Origin of nutrients accumulated in the bottom water of Lake Shinji in summer. *Jpn. J. Limnol*., *57*(4), 313-326. https://doi.org/10.3739/rikusui.57.313

Kamiya, H., Kano, Y., Mishima, K., Yoshioka, K., Mitamura, O., & Ishitobi, Y. 2008. Estimation of long-term variation in nutrient loads from the Hii River by comparing the change in observed and calculated loads in the catchments. *Landscape Ecol. Eng*., *4*, 39-46. https://doi.org/10.1007/s11355-008-0040-9

Kamiya, H., Ohshiro, H., Saga, Y., Sato, S., Nojiri, Y., Kishi, M., Fujihara, A., Godo, T., Sugahara, S., Inoue, T., & Yamamuro, M. 2015a. Effects of residence time and nutrient concentration to the internal COD production in shallow lakes. *Jpn. J. Ecol. Civil Eng*., *17*, 79-88. https://doi.org/10.3825/ece.17.79

Kamiya, H., Ohshiro, H., Tabayashi, Y., Kano, Y., Mishima, K., Godo, T., Yamamuro, M., Mitamura, O., & Ishitobi, Y. 2011. Phosphorus release and sediment in three contiguous shallow brackish lakes, as estimated from changes in phosphorus stock and loading from catchment. *Landscape Ecol. Eng*., *7*, 53-64. https://doi.org/10.1007/s11355-010-0110-7

Kamiya, H., Sugahara, S., Saga, Y., Sato, S., Nojiri, Y., Kishi, M., Fujihara, A., & Godo, T. 2015b. The mechanism and rate of re-sedimentation of phosphorus released from the sediment during the summer months in shallow brackish lakes. *Jpn. J. Limnol*., *76*, 139-148.

https://doi.org/10.3739/rikusui.76.139

Kasaki, H. 1964. The Charophyta from the lakes of Japan. *J. Hattori Botanical Lab*., *27*, 217-314.

Kawahata, C., Yamamuro, M., & Shirakawa, Y. 2013. Changes in alkaline band formation and calcification of corticated charophyte *Chara globularis*. *Springer plus*., *2*, 85. https://doi.org/10.1186/2193-1801-2-85

Komuro, T., Sakayama, H., Kamiya, H., & Yamamuro, M. 2016. Reconstruction of the charophyte community of Lake Shinji by oospore collection. *Knowl. Manag. Aquat. Ecosyst*., *417*, 12. https://doi.org/10.1051/kmae/2015045

Komuro, T., & Yamamuro, M. 2013. Estimation of the aquatic plant community area in Lake Shinji reconstructed with the aerial photographs taken by the US forces in 1940's. *Jpn. J. Ecol. Civil Eng*., *16*(1), 51-59. https://doi.org/10.3825/ece.16.51

Krivonogov, S. K., Yamamuro, M., Takahara, H., Kazansky, A. Y., Klimin, M. A., Bobrov, V. A., Safonova, I. Y., Phedorin, M. A., & Bortnikova, S. B. 2012. An abrupt ecosystem changes in Lake Beloye, southern Western Siberia: Paleoclimate versus local environment. *Paleogeog., Paleoclimat., Paleoecol*., *331-332*, 194-206. https://doi.org/10.1016/j.palaeo.2012.02.030

Królikowska, J. 1997. Eutrophication process in a shallow, macrophyte-dominted lake-species differentiation, biomass and the distribution of submerged macrophytes in Lake Łuknajno (Poland). *Hydrobiologia*., *342/343*, 411-416. https://doi.org/10.1023/A:1017055827120

Kufel, L., & Ozimek, T. 1994. Can Chara control phosphorus cycling in lake Łuknajno (Poland)?. *Hydrobiologia*., *275/276*, 277-283. https://doi.org/10.1007/BF00026718

Kufel, L., Biardzka, E., & Strzałek, M. 2013. Calcium carbonate incrustation and phosphorus ractions in five species. *Aquat. Bot*., *109*, 54-57. https://doi.org/10.1016/j.aquabot.2013.04.002

Kufel, L., & Kufel, I. 2002. Chara beds acting as nutrient sinks in shallow lakes – a review. *Aquat. Bot*., *72*, 249-260. https://doi.org/10.1016/S0304-3770(01)00204-2

Kufel, L., Strzałek, M., & Biardzka, E. 2016. Site- and species-specific contribution of charophytes to calcium and phosphorus cycling in lake. *Hydrobiologia*., *767*, 185-195. https://doi.org/10.1007/s10750-015-2498-4

Miranda, L. E., Driscoll, M. P., & Allen, M. S. 2000. Transient physiochemical microhabitats facilitate fish survival in inhospitable aquatic plant stands. *Freshwater Biol*., *44*, 617-628. https://doi.org/10.1046/j.1365-2427.2000.00606.x

Miyadi, D. 1962. Nakaumi Kanntaku Tannsuika Jigyou ni tomonau Gyozoku Seitai Chousa Houkoku.

Miyazako, T., Sugahara, S., Tabayashi, Y., Ohshiro, H., Koyama, Y., Nakashima, Y., Kamiya H., & Seike, Y. 2014. Comparison of measured and calculated loads using quadratic LQ equation for Hii River flows, eastern Shimane prefecture, Japan. *Jpn. J. Limnol*., *75*, 151-159. https://doi.org/10.3739/rikusui.75.151

Molot, L. A., Watson, S. B., Creed, I. F., Trick, C. G., McCabe, S. K., Verschoor, M. J., Sorichetti, R. J., Powe, C., Venkiteswaran, J. J., & Schiff, S. L. 2014. A novel model for cyanobacteria bloom formation: the critical role of anoxia and ferrous iron. *Freshwater Biol.*, *59*, 1323-1340. https://doi.org/10.1111/fwb.12334

Nõges, P., Tuvikene, L., Feldmann, T., Tõnno, I., Künnap, H., Luup, H., Salujõe, J., & Nõges, T. 2003. The role of charophytes in increasing water transparency; a case study of two shallow lakes in Estonia. *Hydrobiologia.*, *506-509,* 567-573. https://doi.org/10.1023/B:HYDR.0000008625.36438.75

Orihel, D. M., Schindler, D. W., Ballard, N. A., Graham, M. D., O'Connell, D. W., Wilson, L. R., & Vinebrooke, R. D. 2015. The "nutrient pump:" Iron-poor sediments fuel low nitrogen-to-phosphorus ratios and cyanobacterial blooms in polymictic lakes. *Limnol. Oceanog.*, *60*, 856-871. https://doi.org/10.1002/lno.10076

Pereya-Ramos, E. (1981). The ecological role of Characeae in the lake littoral. *Ekol. Pol.*, *29*, 167-209.

Phillips, G. L., Eminson, D., & Moss, B. 1978. A mechanism to account for macrophytes decline in progressively eutrophicated freshwater. *Aquat. Bot.*, *4,* 103-126. https://doi.org/10.1016/0304-3770(78)90012-8

Pinke, G., Csiky, J., Mesterházy, A., Tari, L., Pál, R. W., Botta-Dukát, Z., & Czúcz, B. 2014. The impact of management on weeds and aquatic plant communities in Hungarian rice crops. *Weed Ress.*, *54*, 388-397. https://doi.org/10.1111/wre.12084

Poste, A. E., Hecky, R. E., & Guildford, S. J. 2013. Phosphorus enrichment and carbon depletion contribute to high *Microcystis* biomass and microcystin concentrations in Ugandan lakes. *Limnol. Oceanog.*, *58*(3), 1075-1088. https://doi.org/10.4319/lo.2013.58.3.1075

Prins, H. B. A., Snel, J. F. H., Zanstra, P. E., & Helder, R. J. (1982). The mechanism of biocarbonate assimilation by the polar leaves of *Potamogeton* and *Elodea*. CO_2 concentrations at the leaf surface. *Plant Cell Environ.*, *5*, 207-214. https://doi.org/10.1111/1365-3040.ep11571916

Sato, M., & Shiraiwa, Y. 2012. In Handbook of Algae-Their diversity and utilization. Watanabe N:376-379. NTS, Tokyo, p824.

Scharf, W. 1999. Restoration of the highly eutrophic Lingese Reservoir. *Hydrobiologia.*, *416*, 85-96. https://doi.org/10.1023/A:1003851102095

Scheffer, M. 1989. Alternative stable states in eutrophic freshwater systems: A minimal model. *Hydrobiol. Bull.*, *23*(1), 73-83. https://doi.org/10.1007/BF02286429

Scheffer, M., Carpenter, S., Foley, J. A., Folke, C., & Walker, B. 2001. Catastrophic shifts in ecosystems. *Nature.*, *413,* 591-596. https://doi.org/10.1038/35098000

Scheffer, M., Hosper, S. H., Meijer, M. L., Moss, B., & Jeppesen, E. 1993. Alternative equilibria in shallow lakes. *Trends. Ecol. Evol.*, *8*, 275-279. https://doi.org/10.1016/0169-5347(93)90254-M

Søndergaard, M., Jensen, J. P., Jeppesen, E. 2003. Role of sediment and internal loading of phosphorus in shallow lakes. *Hydrobiologia.*, *506-509,* 135-145. https://doi.org/10.1023/B:HYDR.0000008611.12704.dd

Søndergaard, M., Jeppesen, E., & Jensen, J. P. 2000. Hypolimnetic nitrate treatment to reduce internal phosphorus loading in a stratified lake. *Lake and Reservoir Management*., *16*, 195-204. https://doi.org/10.1080/07438140009353963

Timms, R. M., & Moss, B. 1984. Prevention of growth of potentially dense phytoplankton populations by zooplankton grazing, in the presence of zooplanktivorous fish, in a shallow wetland ecosystem. *Limnol. Oceanogr*., *29*(3), 472-486. https://doi.org/10.4319/lo.1984.29.3.0472

Uemura, S., Kawamura, H., & Tsuji, M. 2002. *Dictionary of herbicide pesticide*. (3rd ed.). 311-313, Sanseido, Tokyo, p640.

Van den Berg, M. S., Coops, M. L., Meijer, M., Scheffer, M., & Simons, J. 1998. *Clear water associated with the dense Chara vegetation in the shallow and turbid Lake Veluwemeer, the Netherland*. *In*: Jeppesen, E., Sondergaard, M., Sondergaard, M., Christoffersen, K (eds). The Structuring Role of Submerged Macrophytes in Lakes. Springer, New York, 339-352. https://doi.org/10.1007/978-1-4612-0695-8_25

Wetzel, R. G. 2001. *Oxygen. In Limnology: Lake and River Ecosystems* (3rd ed.). Wetzel RG: 151-168. Saunders College Publication, Philadelphia, 1006 p.

Yamamuro, M., Kamiya, H., & Ishitobi, Y. 2014. Water quality before and after the break out of submerged plants at Lake Shinji. *Jpn. J. Limnol*., *75*, 99-105. https://doi.org/10.3739/rikusui.75.99

Yokoyama, K., Yamamoto, K., & Kawano, S. 2008. Morphodynamics and transport of sediment phosphorus in the northeast part of the Ariake Sea and the estuarine channel of The Chikugogawariver. *Japanese Journal*, *Doboku Gakkai Ronbunshuu B*., *64*(2), 83-98. https://doi.org/10.2208/jscejb.64.83

Yoshimura, S. 1938. Dissolved oxygen of the lake wates of japan. *Science Reports of the Tokyo Bunrika Daigaku*., *8*, 63-277.

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