

Do benthic algae provide important information over and above that provided by macrophytes and phytoplankton in lake status assessment? – Results from a case study in Norway



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ABSTRACT

To test if phytobenthic algae provide additional important information to macrophytes and phytoplankton for lake monitoring, we sampled two large lakes in Norway. In each lake, we analyzed water chemistry and phytoplankton above the deepest site, recorded macrophytes and non-diatom phytobenthic algae at 20 sites around the shoreline and estimated site-specific nutrient input from land cover. Since no ready-to-use phytobenthos index exists for lakes in Norway, we tested the PIT index developed for rivers, commonly perceived signs of disturbance such as high algal cover, and taxon richness as well as similarity patterns. Both lakes were nutrient poor, but had potential local nutrient inputs (villages, agriculture). In neither of the lakes did phytobenthos indicate a worse overall ecological status than macrophytes and phytoplankton. Our data therefore, did not suggest that it would be useful to add phytobenthos into surveillance monitoring of lakes in Norway. There was a loose correlation between macrophyte and phytobenthic site-specific taxon richness and similarities. This means that macrophytes and phytobenthos do indeed give partly redundant information. High algal cover was found at sites with both high and low phosphorus input. Using algal cover as indicator of site-specific nutrient input is therefore overly simplistic. Urban and cultivated areas were associated with a more eutrophic PIT. This indicates that the PIT, despite being developed for lotic waters, may be used to detect site specific nutrient input in lakes.

1. Introduction

The Water Framework Directive (WFD; [European Commission, 2000](#)) aims to ensure at least good ecological status for all water bodies in Europe. The underlying idea of the WFD is that “good ecological status” reflects “healthy ecosystems”. Consequently, the boundary between good and moderate status in the WFD broadly separates healthy ecosystems (those that are in good or high status) from the rest (those that are in moderate, poor, or bad status; [Kelly et al., 2016](#)). Healthy ecosystems are commonly understood to be able to maintain viable and diverse populations, to maintain ecological processes such as nutrient cycles, as well as being able to provide sustainable ecosystem services for human use ([Grumbine, 1994](#)). Most member states, however, only use abundance/composition or sensitivity/tolerance metrics for WFD-compliant ecological status classification, while functional metrics which could indicate ecological processes are rarely used ([Poikane et al., 2015](#)). For lakes, the WFD defines good ecological status as ecosystems that show only “slight” differences in the composition and abundance of phytoplankton, aquatic flora, benthic invertebrates, and

fish compared to reference status. The good-moderate boundary is at the point where “slight” differences turn into “moderate” differences compared to reference status. Since - possibly costly - measures need to be taken if good ecological status is not achieved, the good-moderate boundary receives much attention in ecological status classification.

The overall classification of a lake into an ecological status class does not yet provide information with respect to which stressors may affect the lake ecosystem. If a lake achieves at least good status, knowledge about stressors is less important, simply because the overall situation generally is accepted as being “good enough”. Consequently, if the required biological quality elements for surveillance monitoring (i.e. for lakes phytoplankton, aquatic flora, benthic invertebrates, and fish) all indicate at least good status for a lake as a whole, there is no legal need to know more about detailed impacts of stressors which may operate only at specific sites or areas within the lake. However, if a lake fails to achieve good status, measures must be taken, and water managers need to know which pressures need to be addressed to improve conditions ([Schneider et al., 2016](#)). Most metrics (often also called “indices”) used for status assessment according to the WFD therefore

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have a “double role”: they are used for overall classification of a water body into one of five status classes (high, good, moderate, poor, and bad), but they also are related to specific stressors, such as nutrient enrichment, organic pollution, or water level fluctuations. This means, if good ecological status is not achieved, these indices can also be used as a source of information with respect to which stressors affect the lake (Birk et al., 2012).

The primary producers among the required biological quality elements, i.e. phytoplankton and aquatic flora – the latter consisting of the sub-elements macrophytes and phytobenthos – are mostly used to assess eutrophication, i.e. the over-enrichment of a water body with nutrients (Birk et al., 2012). However, metrics related to other stressors have also been developed. In Norway, for example, macrophytes in lakes also are used to indicate acidification, and water level fluctuations (Mjelde et al., 2013; Lyche Solheim et al., 2017). Most countries in Europe have assessment systems for phytoplankton and macrophytes in place (Poikane et al., 2015), but fewer countries use phytobenthic algae in lakes (Poikane et al., 2016). Since phytoplankton, macrophytes and phytobenthos all are sensitive to eutrophication, which still is the most widespread stressor in lakes in Europe (Noges et al., 2016), there is a question of redundancy: why should we analyze phytobenthos, if phytoplankton and macrophytes provide the same information? Monitoring is expensive, and the money may e.g. be used for restoration measures instead. There is no reason why phytobenthos has to be the third choice; however, this is the practice that has evolved throughout much of Europe (Kelly et al., 2016).

In the EU intercalibration exercise, i.e. the process that ensured that the good-moderate boundary was comparable among all countries in Europe (Poikane et al., 2015), it was agreed upon that where a biological quality element consists of two components, such as macrophytes and phytobenthos, it may be sufficient to use only one of the two components (European Commission, 2010). However, if only one component is used, then it must be demonstrated that the impacts of the existing pressures are being sufficiently detected by that component (European Commission, 2010). Many countries therefore argued that macrophytes would be sufficient to indicate shoreline eutrophication, and phytobenthos would be redundant (Kelly et al., 2016). The concern that macrophytes react slower to eutrophication than benthic algae and therefore may overlook recent eutrophication processes (Schneider et al., 2012), was responded to with the argument that phytoplankton also was assessed, and this would respond relatively rapidly (Kelly et al., 2016).

It is true that phytoplankton (Carvalho et al., 2013) and macrophytes (Sondergaard et al., 2010) are powerful ecological indicators for lake assessment, and Kelly et al. (2016) have shown that – indeed – phytobenthos is detecting relatively few impacted lakes that have not already been identified by phytoplankton and macrophytes. However, when a lake’s littoral zone is affected by hydrological pressures (such as lake level alterations), the frequency of occurrence of many macrophyte species may decline dramatically (Boschilia et al., 2012). In such cases, and in lakes where the macrophyte community is naturally species-poor, phytobenthos may be a better indicator of nutrient effects in the littoral than macrophytes (DeNicola and Kelly, 2014). Both situations, i.e. water level alterations caused by regulation, and naturally species-poor macrophyte communities are common in Norway, suggesting that phytobenthos in Norwegian lakes could indeed be relevant for lake status assessment. In addition, phytobenthos is known to react more quickly to changes in nutrient input than macrophytes (Schneider et al., 2012) and may therefore be better suited to detect local hotspots of nutrient input (DeNicola and Kelly, 2014). In other words: phytobenthos may be important in operational and investigative monitoring, even if its role in overall lake status classification and surveillance monitoring may be limited (Kelly et al., 2016).

Phytobenthic algae never were part of routine lake monitoring in Norway. Data are therefore scant, inconsistent and lack related information on other organism groups or water chemistry. To test if

phytobenthic algae provide important additional information over and above macrophytes and phytoplankton, we took samples at 40 sites in two large regulated lakes in Norway. We selected regulated lakes because we expected that water level fluctuations may cause depauperate macrophyte assemblages, which in turn may lead to unreliable macrophyte index results. If this was the case, then the phytobenthos assessment should be more informative than macrophytes. We selected large lakes because the detection of local hotspots of nutrient input, for which phytobenthos is assumed to be particularly well suited (DeNicola and Kelly, 2014), may generally be more relevant than in small lakes, which are more likely to have comparatively homogeneous surroundings. Since no ready-to-use phytobenthos index exists for lakes in Norway, a purely statistical approach to address redundancy among index values was not possible (such as in Kelly et al., 2016). In addition, the few WFD indices which have been developed for phytobenthos in lakes all are based on diatoms (Poikane et al., 2016). However, Norway is the only country in Europe which exclusively uses non-diatom benthic algae for freshwater monitoring (Poikane et al., 2016). Consequently, it was not possible to test existing lake indices from other countries on our data. Instead, we tested the PIT index developed for rivers in Norway, looked at commonly perceived signs of disturbance (such as high cover of green algae and cyanobacteria; Hart et al., 2013), and analyzed taxon richness as well as similarities among macrophyte and phytobenthos assemblages. To identify potential local hotspots of nutrient input, we estimated phosphorus export from the adjacent 2.5 ha of land at each of the 40 sampling sites from CORINE land cover (McGuckin et al., 1999) and from the national land resource map of Norway (AR50 site index classification; Ahlstrøm et al., 2014).

We addressed the following questions: (1) Are there signs that phytobenthic algae may indicate a worse overall lake status than phytoplankton and macrophytes? If phytobenthos indicates worse ecological status, then this may indicate that phytobenthos is more sensitive to the existing stressors than macrophytes and phytoplankton. This would be a strong argument for including phytobenthos into surveillance monitoring. (2) Do taxon richness and community similarity patterns of phytobenthos and macrophytes co-vary? Covariation would indicate that similar natural conditions and stressors affect macrophytes and phytobenthos, which in turn would mean that they give redundant information for ecological status evaluation. (3) Are phytobenthic algal assemblages more closely related to local hotspots of nutrient input than macrophyte assemblages? If phytobenthic algae indicate local hotspots of nutrient input better than macrophytes, this would be an argument for including phytobenthos into operational and investigative monitoring.

2. Material and methods

2.1. Study area

Snåsavatnet (122 km², 21 m a.s.l., 121 m maximum depth) and Selbusjøen (57 km², 159 m a.s.l., 206 m maximum depth) are large lakes located in Mid-Norway (Fig. 1). Both lakes have a Calcium concentration around 3.5 mg/l and an alkalinity around 0.18 meq/l (Lyche Solheim et al., 2017). Although both lakes are regarded as clear, Snåsavatnet has a higher TOC concentration (4 mg/l TOC in Snåsavatnet compared to 2.5 mg/l TOC in Selbusjøen). Both lakes are regulated, but in Snåsavatnet the difference between upper and lower regulation level is only 1.4 m, while it is 6.3 m in Selbusjøen. Consequently, Selbusjøen is regarded as heavily modified. For lake status classification, however, the same class boundaries are used as for corresponding lake types (Lyche Solheim et al., 2017).

2.2. Sampling

In both lakes, the complete program of surveillance monitoring according to the WFD was performed in 2016, including analysis of

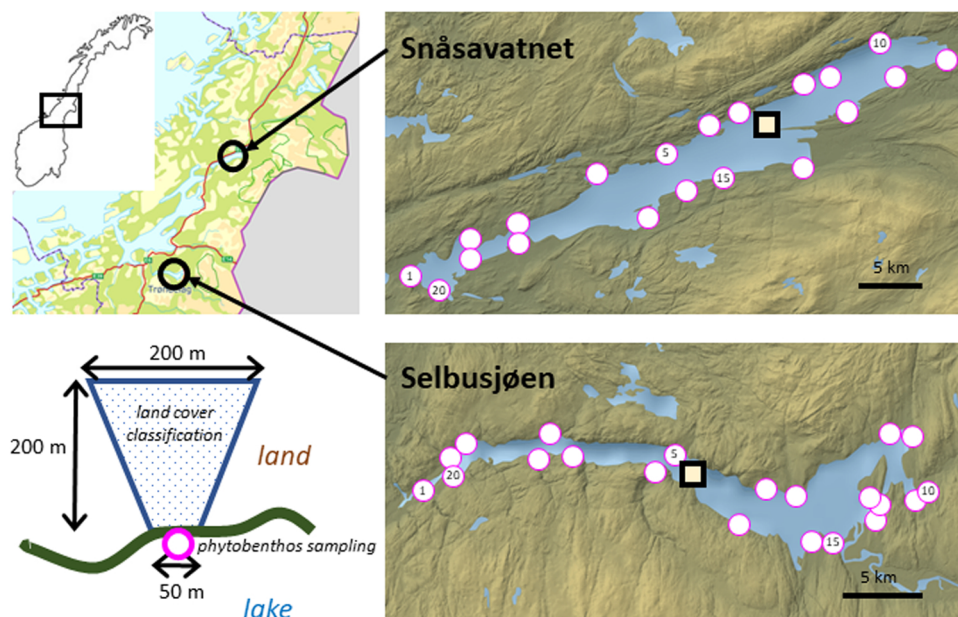


Fig. 1. Location of sampling sites in Snåsavatnet and Selbusjøen. Circles indicate the sampling sites for macrophytes and phytobenthos, quadrats the sites for water chemistry and phytoplankton sampling. The lower left panel exemplifies how the area was delineated, in which CORINE land cover and AR50 site index classifications were quantified at each site.

water chemistry, phytoplankton and macrophytes (Lyche Solheim et al., 2017). Phytoplankton and water chemistry samples were taken monthly during the growth season (from May to October) in form of composite samples taken from 0 to 10 m water depth at the deepest point of the lake. Water chemistry was analyzed according to Norwegian standard procedures (Lyche Solheim et al., 2017). In each lake, macrophytes were mapped once at 20 sites in August 2016, using an bathyscope (a bucket with a transparent bottom) and a rake. Abundance for each species at each site was recorded according to a five-degree scale (1 = very rare, 2 = infrequent, 3 = common, 4 = frequent, 5 = abundant, predominant).

Phytobenthos samples were taken at the same sites and at the same time as the macrophytes. At each site, samples of non-diatom benthic algae were collected according to European standard procedures (EN 15708:2009) at approximately 0.5 to 1 m water depth, along an approximately 10-m length of the shoreline using a bathyscope. The water level at the time of sampling was average to slightly above average for both lakes. However, periphyton growth was clearly visible at the sampling depth, indicating that this area had been inundated for at least several weeks prior to sampling. Percent cover of each form of macroscopically visible benthic algae was recorded, and samples were collected and stored separately in vials for species determination. In addition, microscopic algae were collected from ten cobbles/stones with diameters ranging between approximately 10 and 20 cm, taken from each site. An area of about 8 x 8 cm from the upper side of each cobble/stone was brushed with a toothbrush to transfer the algae into a beaker containing approximately 1 l of lake water from which a subsample was taken. All samples were preserved with a few drops of formaldehyde. The preserved benthic algae samples were later examined in a microscope (200 to 600 × magnification) and all non-diatom algae identified to species, when possible. Diatoms were not included due to the great differences in methodology for sample preparation and enumeration between diatom and non-diatom benthic algae, and to ensure consistency with the Norwegian assessment system for rivers and streams, which exclusively relies on non-diatom benthic algae (Schneider and Lindstrøm, 2011). For some genera of filamentous green algae that could not be determined to species level (e.g. *Spirogyra* Link or *Mougeotia* C. Agardh), categories based mainly on filament width were used (see Schneider and Lindstrøm (2009, 2011) for further details). Abundance of each microscopic taxon was estimated in the laboratory as “rare”, “common” and “abundant”. For data analysis, these estimates were later translated into % cover as 0.001, 0.01 and

0.1%, respectively. Macroscopic algae whose cover was recorded as “< 1%” in the field, were noted as “0.1%” for data analysis. For all other taxa, the cover that was estimated in the field was used.

2.3. Estimation of site specific nutrient input from the near shore surroundings

The CORINE Land Cover map (CLC) covers all Europe and was first developed for the reference year 1990 by EUs CORINE (Coordination of Information on the Environment) program (EEA, 2007). It has since been updated three times, in 2000, 2006, and 2012 (EEA, 2014). The CLC maps classify areas into 44 landcover classes, grouped in a three-level hierarchy, with a minimum mapping unit of 25 ha and 100 m as the minimum width of linear elements (EEA, 2007). The national land resource map of Norway (AR50) is based on the AR5 resource map (Ahlstrøm et al., 2014). Each area is classified using four different characteristics: main land use category, forest productivity, main forest species, and type of agriculture. These four classifications result in 106 area types (legal combinations of the four characteristics). In AR50, the minimum mapping unit is in general 1.5 ha, although smaller units can occur.

For each of the 40 sites sampled for macrophytes and phytobenthos, we used GIS to delineate a 2.5-hectare trapeze that stretched 200 m inland from the shore of the lake (Fig. 1). 200 m was chosen based on the topography of the adjacent land: we aimed at standardizing the area while avoiding including valleys with an orientation parallel to the shoreline (since nutrients from such areas would not be transported towards this lake site). For each trapeze, we calculated the areas of different land cover classes, using both CORINE and the AR50 site index classifications, respectively. Phosphorus export coefficients for each land use category were taken from Smith et al. (2005). The AR50 site index differentiates among high, medium and low productivity within several of the index classes. Within each index class, high values were assigned an export coefficient of “average plus one standard deviation”, while low values were assigned an export coefficient of “average minus one standard deviation” (as tabled in Smith et al., 2005). For each trapeze, the phosphorus export (in kg/year) was calculated by summarizing the products of land cover area (in each land use class) with their export coefficients.

2.4. Data treatment

Ecological status evaluation for macrophytes, phytoplankton and the supporting water chemical parameters followed the Norwegian standard methods (Lyche Solheim et al., 2017). Briefly, phytoplankton assessment in Norway is based on average Chl a concentration, average PTI (an index calculated from taxon specific nutrient preferences), average phytoplankton biovolume, and maximum biovolume of cyanobacteria, calculated from six monthly samples taken between May and October. The overall status classification for phytoplankton is generally derived by averaging these four metrics (however, some extra rules apply; see Lyche Solheim et al., 2017; DG, 2013). The TIC index is used for assessment of eutrophication based on macrophytes, an index reflecting the relative occurrences of species which are sensitive and tolerant to nutrient enrichment (DG, 2013). The TIC is calculated from summarized species lists for entire lakes, resulting in one status assessment for an entire lake. The TIC is not designed for site specific use, i.e. for detecting small-scale differences among sites within a lake. It is, however, nevertheless possible to calculate site-specific TIC indices from the site-specific recordings of macrophyte presence.

Since no phytobenthos metrics exist for the assessment of lakes in Norway, we calculated the Periphyton Index of Trophic status PIT (Schneider and Lindstrøm, 2011), an index reflecting eutrophication which was developed for use in Norwegian rivers and streams. In many cases, the similarities between rivers and the shallow part of lake littoral zones mean that concepts should be transferable (DeNicola and Kelly, 2014). Also for macrophytes, there is a correlation between species specific eutrophication scores for lakes and rivers (Schneider, 2007). The PIT index should therefore reflect eutrophication in lakes as well. For status class boundaries, we used the same boundaries as for rivers with a corresponding water chemistry, i.e. the boundaries given for rivers with a Ca concentration > 1 mg/l (DG, 2013). For this river type, the boundary between high and good status is at PIT = 9.5, while the good-moderate boundary is at 16. In addition to the PIT index, we calculated the following response parameters from the phytobenthos taxon lists: (1) taxon richness of phytobenthic algae, as well as of green algae and cyanobacteria, as a simple means to describe biodiversity; (2) total cover of benthic algae (calculated as sum of cover of all taxa), as well as of green algae and cyanobacteria, because they often are viewed as a sign of degradation (Hart et al., 2013), and because the WFD mentions “accelerated growth of phytobenthos” in the definition of lake ecological status classes (European Commission, 2000).

All phytobenthos response variables were calculated twice: once for the complete taxon list, and a second time for a reduced taxon list, which excluded all taxa which lack hold-fast structures (e.g. the genera *Cosmarium* Corda ex Ralfs and *Closterium* Nitzsch ex Ralfs). This was done because taxa lacking hold-fasts may not be strictly benthic, i.e. they may be transported among sites by wave action, such that their occurrence at a particular site may not necessarily be related to site-specific pressures. In addition, the overall PIT index for each lake was calculated in two different ways: once by averaging the PIT index at the 20 sites analyzed in each lake, and a second time by creating an overall taxon list for each lake, and calculating the PIT from the overall taxon list. This was done because there is no a priori best way how to calculate an overall lake index from several sites, such that we tested both options.

To explore species composition and abundance of the macrophyte and phytobenthos assemblages, an NMDS (non-metric multidimensional scaling) was computed on the square-root transformed data. NMDS was used because, in contrast to other ordination methods, it can also handle non-linear responses. The NMDS was computed using the metaMDS function in R version 2.14.2 (R Development Core Team, 2012), extended with the “vegan” package 2.0–4 (Oksanen et al., 2012). Bray-Curtis was used as dissimilarity measure because it is less dominated by single large differences than many other dissimilarity measures (Quinn and Keough, 2002). Two sites in Selbusjøen had no

macrophyte vegetation, such that it was not possible to calculate dissimilarities. For visualization in the NMDS plots, site scores similar to a site with only one very rare species were manually assigned.

To test if similarities between phytobenthos assemblages were correlated with similarities between macrophyte abundances (i.e. to test if the phytobenthos assemblages differ in a similar way among sites as the macrophyte assemblages), a Mantel test was computed, using Bray-Curtis as dissimilarity measure, and 999 permutations. Two sites without macrophyte vegetation were excluded from this exercise. To make macrophyte and phytobenthos similarities comparable, phytobenthos abundances were translated into 5-scale abundance classes using the German system (1, microscopically rare; 2, microscopically abundant; 3, maximum 5% cover; 4, 5% to 33% cover; 5, more than 33% cover; Schneider et al., 2018). In addition, presence-absence data also were tested.

To explore if land use adjacent to the sampling sites can explain site-specific macrophyte and phytobenthos assemblages, linear models were computed using the MASS-package in R (Venables and Ripley, 2002), separately for each response variable and each lake, with explanatory variables selected through forward selection based on AIC (Akaike information criterion). Variables were log or sqrt transformed where necessary in order to improve normality and homoscedasticity. To avoid overinterpretation, criteria in addition to model significance were applied for identifying informative models: a model was deemed informative if the adjusted R^2 was > 0.2, and the number of model parameters was ≤ 3 , i.e. there occurred no obvious model overfitting.

3. Results

3.1. Overall assessment of lake ecological status

Complete lists of macrophyte and phytobenthos taxa found in Snåsavatnet and Selbusjøen are given in appendix (Tables S1 and S2). Briefly, we found a total of 21 macrophyte species in Snåsavatnet, and the dominant species were *Isoetes echinospora* Durieu, *I. lacustris* L., *Ranunculus reptans* L., *Subularia aquatica* L. and *Myriophyllum alterniflorum* DeCandolle in Lamarck et DeCandolle. Among the phytobenthic algae, the genus *Spirogyra* reached a cover of $\geq 50\%$ at 6 out of 20 sites (sites 5, 8, 9, 10, 15 and 16). Among the taxa which were less abundant, but present at almost all sites were the cyanobacterium *Merismopedia* Meyen sp., and the green algal genera *Bulbochaete* C. Agardh, *Cosmarium*, and *Oedogonium* Link ex Hirn.

We found 20 macrophyte species in Selbusjøen, and the dominant species were *Ranunculus reptans*, *Subularia aquatica*, *Potamogeton perfoliatus* L., *Sparganium angustifolium* Michaux fil. and *Nitella opaca* (C.Agardh ex Bruzelius) C.Agardh. Two sites (sites 5 and 6) at the northern shore were located underneath steep high cliffs, and were devoid of aquatic vegetation. Benthic algae generally occurred in low abundances at all sites, and the most prevalent taxa were the cyanobacterium *Dichothrix orsiniana* Bornet & Flahault, as well as the green algal genera *Bulbochaete*, *Cosmarium*, *Mougeotia*, *Oedogonium* and *Zygnema* C.Agardh.

Of the 86 phytobenthos taxa we registered in Snåsavatnet and Selbusjøen, 58 were assigned a PIT indicator value, i.e. are recognized as having a comparatively narrow ecological amplitude in Norwegian lotic waters. Of these, 7 (i.e. 12% of the indicator taxa) were assigned a PIT indicator value > 16, i.e. worse than the good-moderate boundary for rivers with a Ca-concentration > 1 mg/l. In contrast, 48 (i.e. 83% of the indicator taxa) were assigned a PIT indicator value < 9.5, i.e. indicate “high” ecological status in rivers with a Ca-concentration > 1 mg/l.

Detailed results for phytoplankton are given in Lyche Solheim et al. (2017). Briefly, the phytoplankton assemblages in both lakes were dominated by chrysophytes, cryptophytes, and diatoms. Average Chl a concentration was 0.8 $\mu\text{g/l}$ in both lakes, and the average phytoplankton biovolume was 0.15 mm^3/l in Snåsavatnet, and 0.13 mm^3/l in

Table 1

Ecological status for Snåsavatnet and Selbusjøen, as indicated by phytoplankton, macrophytes and phytobenthos, as well as supporting water chemical parameters. Phytobenthos assessment was based on PIT status classes for rivers with a Ca-concentration > 1 mg/l, i.e. reflecting the Ca-concentrations measured in Snåsavatnet and Selbusjøen. Phytoplankton and water chemistry results were calculated from monthly samples taken between May and October.

	value	ecological status class
Snåsavatnet		
phytoplankton		
Chl a (µg/l)	0.8	high
phytoplankton biovolume (mm ³ /l)	0.15	high
PTI index	2.27	moderate
max biovolume cyanobacteria (mm ³ /l)	0.01	high
macrophytes		
Tlc	81	high
benthic algae		
average PIT of 20 sites	8.1	high
PIT from "overall taxon list"	8.6	high
average PIT of 20 sites only strictly benthic taxa	9.3	high
PIT from "overall strictly benthic taxon list"	9.2	high
water chemistry		
total phosphorus (µg/l)	4.7	good
total nitrogen (µg/l)	281	good
Secchi depth (m)	4.8	high
Selbusjøen		
phytoplankton		
Chl a (µg/l)	0.81	high
phytoplankton biovolume (mm ³ /l)	0.13	high
PTI index	2.09	good
max biovolume cyanobacteria (mm ³ /l)	0.003	high
macrophytes		
Tlc	70	good
benthic algae		
average PIT of 20 sites	6.4	high
PIT from "overall taxon list"	8.9	high
average PIT of 20 sites only strictly benthic taxa	6.7	high
PIT from "overall strictly benthic taxon list"	9.5	high
water chemistry		
total phosphorus (µg/l)	3.3	high
total nitrogen (µg/l)	187	high
Secchi depth (m)	6.6	high

Selbusjøen, respectively. The maximum cyanobacteria biovolume was 0.011 mm³/l in Snåsavatnet, and 0.003 mm³/l in Selbusjøen.

The macrophyte index Tlc indicated “high” ecological status for Snåsavatnet (Table 1). In contrast, the phytoplankton index PTI indicated “moderate” status. However, since Chl a, phytoplankton biovolume and the maximum biovolume of cyanobacteria all indicated “high” status (Table 1), the overall status class for phytoplankton was assessed as “good”. The supporting water chemical parameters indicated “good” status for TP and TN, and “high” status for Secchi depth. For phytobenthos, the Periphyton Index of Trophic status PIT indicated “high” ecological status, irrespective of which method was used for calculating the overall lake status class (Table 1). However, the indices calculated from the “strictly benthic” taxon list were slightly higher, i.e. indicated slightly more nutrient enriched conditions, than when calculated from the complete taxon list. In summary, among the primary producers, phytoplankton indicated “good” ecological status, while macrophytes and phytobenthos indicated “high”.

For Selbusjøen, the macrophyte index Tlc indicated “good” ecological status (Table 1). The phytoplankton index PTI also indicated “good” status, but Chl a, phytoplankton biovolume and the maximum biovolume of cyanobacteria all indicated “high” status, such that the overall phytoplankton assessment resulted in “high” status. All

supporting water chemical parameters also were in “high” status. The Periphyton Index of Trophic status PIT indicated “high” status for all four different ways of calculating overall PIT. However, the PIT calculated from the overall strictly benthic taxon list indicated slightly more nutrient-enriched conditions and was at the boundary between “high” and “good” status (which is at PIT = 9.5; Table 1). In summary, among the primary producers, the macrophytes indicated “good” ecological status, while phytoplankton and benthic algae indicated “high”.

3.2. Richness and diversity patterns

In Snåsavatnet, there were no obvious groups of sites which shared similar macrophyte or phytobenthos assemblages among each other while differing from the remaining sites (Fig. 2a, b). Nevertheless, assemblages at some individual sites differed from the others (i.e. they are located at the edges of the NMDS plots; Fig. 2). With respect to macrophyte assemblages, sites 3 and 10 were slightly different from the remaining sites. Site 10 is situated at a campground, and had a more depauperate macrophyte vegetation than the other sites (Fig. 2a, Table S2a). The macrophyte assemblage at site 3 differed from the other sites by its absence of *Isoetes*. There are some cottages close to site 3, and the surrounding area is agriculturally used, but the reason for the absence

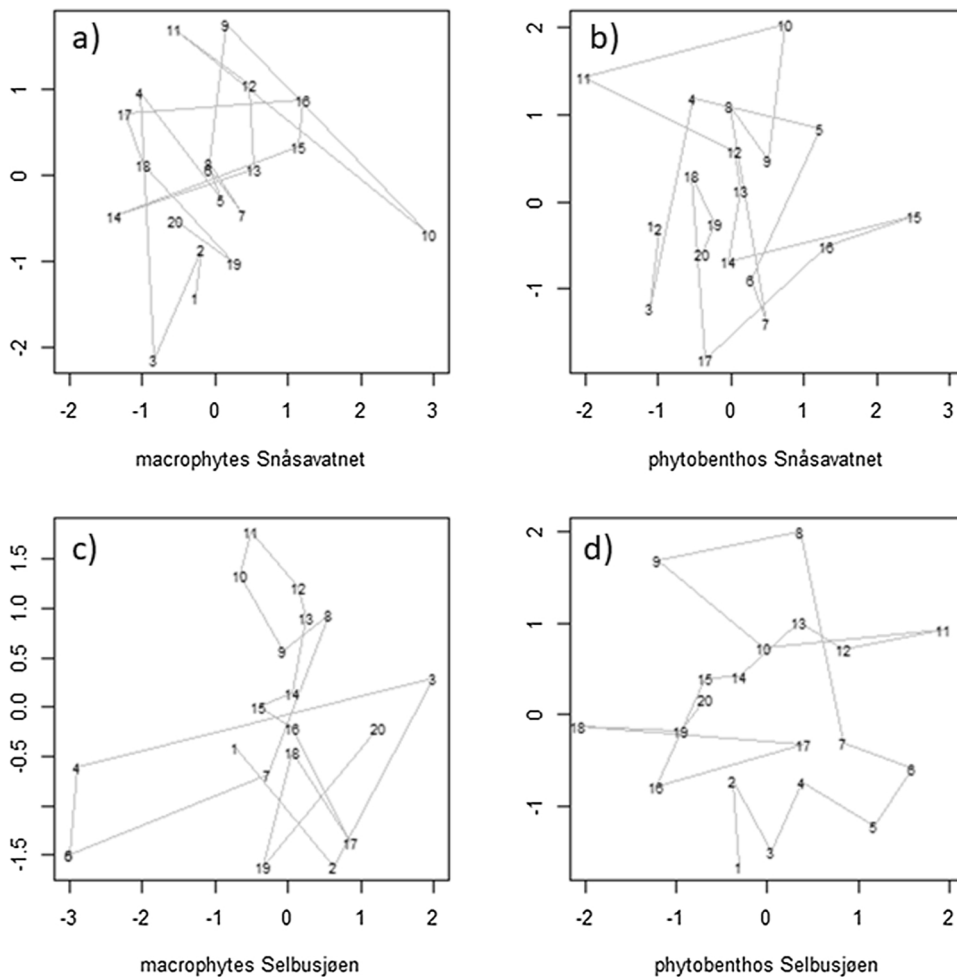


Fig. 2. NMDS based on the complete benthic algal taxon lists, as well as for macrophytes, separate for Selbusjøen and Snåsavatnet; no macrophytes occurred at sites 5 and 6 in Selbusjøen, such that dissimilarity could not be calculated; to depict these sites on the plot, site scores were manually assigned, with similar but more extreme values than site 4, which was the only site where only one macrophyte species occurred in “very rare” abundance. Note that sites 5 and 6 in Selbusjøen lie on top of each other.

of *Isoetes* at this site is unknown. With respect to benthic algal assemblages, sites 10, 11, 15, and partly 17, differed from the other sites (Fig. 2b). At site 10, situated at a campground, *Spirogyra*, *Zygnema* and *Oedogonium* together reached a cover of 70% (Table S1a). Site 11 is located at the north-eastern-most shore of Snåsavatnet, with a sandy beach, and a small village close to the shore. Mats of algae washed ashore were observed during sampling. The phytobenthos assemblage at site 11 differed from the other sites by the presence of *Homoeothrix varians* Geitler, and the absence of the otherwise quite common genus *Bulbochaete* sp. Site 15 is situated close to a railway line. It was the site with the fewest number of phytobenthos taxa in Snåsavatnet, had a high cover of *Spirogyra*, was the only site where *Cosmarium* sp. was absent, and one of two sites where *Gongrosira* sp. Kützing was found in Snåsavatnet. At site 17, cattle could enter the lake. This site was the only site in Snåsavatnet with a high cover of *Phormidium autumnale* Gomont.

In Selbusjøen, sites 4, 5, and 6 stood out by having no or almost no submerged vegetation (Fig. 2c, Table S2b). Phytobenthos taxon richness at sites 5 and 6 was below average (9–10 taxa), but there occurred 15 phytobenthos taxa at site 4 (Table S1b). Sites 4–6 have a steep stony littoral and are located at the northern lake shore underneath steep high cliffs, which probably explains the depauperate macrophyte vegetation. Another group was formed by sites 2, 17 and 19. They have in common that the macrophyte vegetation was depauperate (only 3–4 species occurred per site). In contrast, phytobenthos taxon richness at these sites was 9–16 (Table S1b). The same is true for site 3. At all these sites, the littoral sediment consisted of gravel, and they are obviously a point of access for swimming, bathing, and boating. Sites 8–13 formed a third group. They lie in the easternmost bay of Selbusjøen, and the

macrophyte vegetation differs from most other sites by the occurrence of *Callitriche hamulata* Kützing ex Koch, *Myriophyllum alterniflorum*, and *Potamogeton perfoliatus*. With respect to the phytobenthos assemblages, no groups of sites sharing similar assemblages were obvious (Fig. 2d). The individual sites with benthic algal assemblages most dissimilar from the rest were sites 8, 9, 11, and 18 (Fig. 2d). Sites 8 and 9 are located at the eastern shore of Selbusjøen, and were the only sites where *Stigeoclonium* sp. Kützing occurred. There was some agriculture, and a small factory close to the shoreline, but at both sites there were buffer stripes with woody vegetation towards the lake shore. Site 11 was the only site where *Anabaena* sp. Bory de Saint-Vincent & Flahault, *Chaetophora elegans* (Roth) C. Agardh and *Tetraspora gelatinosa* (Vaucher) Desvaux occurred. This site is situated in front of a wood-processing factory, and the littoral sediment was covered with wood chips. Site 18 is located at a small valley between deep cliffs. The valley ground is used for agriculture, and there also is a small inflow to Selbusjøen. The phytobenthos assemblage differed from the other sites by the occurrence of *Phormidium corium* Gomont ex Gomont, *Microspora lauterbornii* Schmidle and a broad form of *Mougeotia* (Table S1b).

The average number of macrophyte species per site was 11 in Snåsavatnet (range: 4–16), while an average of 18 phytobenthos taxa was recorded per site (range: 10–22). Selbusjøen was slightly species poorer, with an average of 6 macrophyte (range 0–13), and 15 phytobenthos taxa per site (range 9–23). At most sites, there occurred fewer macrophyte than phytobenthos taxa (Fig. 3). This was also by and large true when only strictly benthic algal taxa were considered (Fig. 3b). Although the explained variance was low, benthic algal taxon richness was significantly correlated with macrophyte taxon richness (Pearson $r = 0.33$; $p = 0.04$), but only for the complete taxon list, not for the

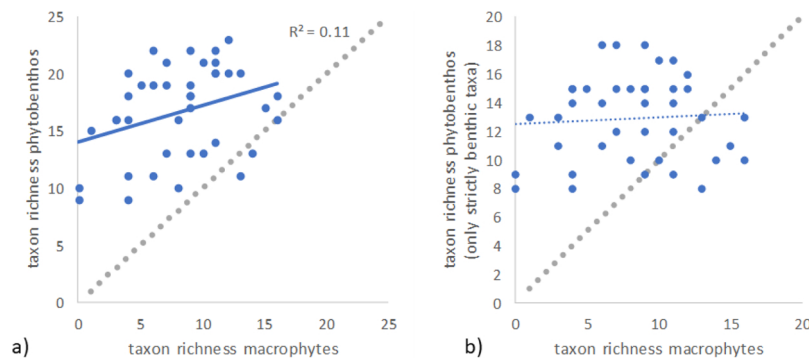


Fig. 3. Benthic algal taxon richness plotted against macrophyte taxon richness; a) complete phytobenthos taxon list, b) only strictly benthic algal taxa; the grey dotted line depicts a 1:1 relationship.

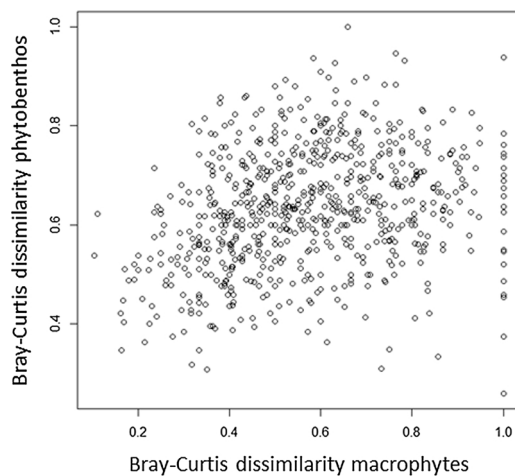


Fig. 4. Bray-Curtis dissimilarities among site-specific macrophyte assemblages, plotted against dissimilarities among site-specific phytobenthos assemblages; 5-scale abundance classes were used for calculating dissimilarities. Mantel test statistics: $r = 0.27$, $p = 0.001$.

strictly benthic taxon list.

Despite considerable variation, dissimilarities in phytobenthos assemblages among sites were significantly correlated with dissimilarities in macrophyte assemblages among sites (Fig. 4). This was true for all four different ways of calculating dissimilarities: using presence-absence data as well as 5-scale abundance classes, for the complete as well as for the “strictly benthic” algal taxon list (Mantel test statistics: complete phytobenthos taxon list, abundance classes: $r = 0.27$, $p = 0.001$; strictly benthic algal taxon list, abundance classes: $r = 0.18$, $p = 0.013$; complete phytobenthos taxon list, presence-absence: $r = 0.27$, $p = 0.001$; strictly benthic algal taxon list, presence-absence: $r = 0.19$, $p = 0.016$).

3.3. Site specific eutrophication effects

To analyze if macrophytes and phytobenthos provided comparable information on local hotspots of nutrient input, we tested if site-specific values of the eutrophication indices for macrophytes and phytobenthos (Tic and PIT, respectively) were correlated with each other. This was not the case, however, even though by and large, high Tic values (indicating oligotrophic conditions) were associated with low PIT values (also indicating oligotrophic conditions; Fig. 5).

Land use adjacent to the sampling sites was dominated by agriculture and coniferous forest in both lakes (Table S3). However, some sites also were close to urban areas. We estimated P-export from 2.5 ha adjacent to each site based on CORINE land cover and AR50 site index classes (Table S3). Average P-export per site estimated from CORINE

was 7.5 kg/year in Snåsavatnet (range: 0.5–11.6), and 6.1 kg/year in Selbusjøen (range: 0.8–12.2). Average P-export estimated from AR50 site indices was 3.9 kg/year in Snåsavatnet (range: 0.9–12.2), and 1.8 kg/year in Selbusjøen (range: 0.6–12.2).

To test if local phosphorus export affected site-specific macrophyte and phytobenthos assemblages, we tested for correlations between P-export, and macrophyte and phytobenthos response parameters, separately for Snåsavatnet, Selbusjøen, and the combined dataset (Table S4). The only significant correlation occurred between P-export and the taxon number of macrophytes in the combined dataset, indicating that more macrophyte taxa occurred at sites with a higher nutrient export. However, the explained variance was low (Table S4, Fig. 6).

To test if different land use categories adjacent to the sampling sites affected macrophyte and phytobenthos assemblages, we modelled the response parameters from CORINE land cover and AR50 site index classes, separately for each lake and the combined dataset. Three consistent relationships (i.e. informative and similar models for each lake, as well as for the combined dataset) were detected (Table 2). (i) Increased algal cover occurred at sites with low index coniferous forest, sparsely vegetated and urban areas, while decreased algal cover occurred at sites with medium index coniferous forest and agricultural areas (quantified from AR50). (ii) Sites located adjacent to wetlands and agricultural areas had a lower (more eutrophic) macrophyte index Tic, while sites located adjacent to sparsely vegetated areas or medium index coniferous forest (quantified from AR50) had a higher (more oligotrophic) Tic. (iii) Sites located adjacent to urban areas and areas with complex cultivation patterns (quantified from CORINE) had a higher (more eutrophic) PIT index.

4. Discussion

4.1. Overall assessment of lake ecological status

Macrophytes and phytoplankton are routinely used in lake status assessment and, in this study, we asked if phytobenthic algae could provide additional important information to these two biological quality elements. There is no a priori reason why phytobenthos has to be the third choice, but this is the situation that has developed in most of Europe (Kelly et al., 2016). If phytobenthic algae indicated a worse ecological status than macrophytes or phytoplankton, then this would suggest that they are more sensitive to the relevant pressures. This would be a strong argument for including phytobenthos in surveillance monitoring. However, this was not the case for the two lakes of our case study. In Snåsavatnet, phytoplankton was “worst” by indicating “good” ecological status, while macrophytes and phytobenthos indicated “high”. In Selbusjøen, macrophytes were “worst” by indicating “good” ecological status, while phytoplankton and phytobenthos indicated “high”. While the macrophyte and phytoplankton assessment methods used in Norway are officially accepted and intercalibrated with

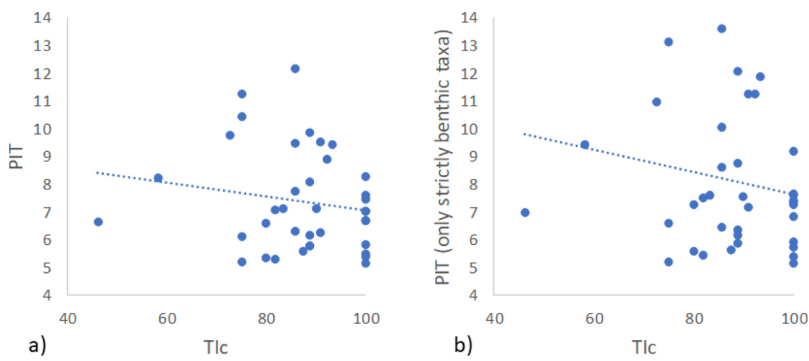


Fig. 5. Macrophyte trophic index (Tic) plotted against the Periphyton Index of Trophic status (PIT), at the 38 sites in Selbusjøen and Snåsavatnet for which both Tic and PIT could be calculated; a) complete benthic algal taxon list; b) only strictly benthic algal taxa.

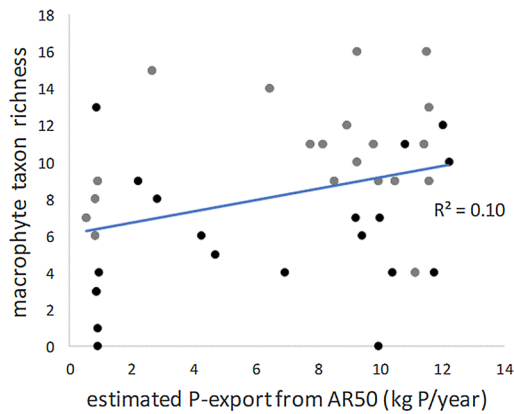


Fig. 6. Macrophyte taxon richness at 20 sites in Selbusjøen (black) and 20 sites in Snåsavatnet (grey), plotted against estimated P-input from the nearshore area (from AR50 site index classes). The correlation line is derived from the combined dataset. Note that the correlation is only significant for the combined dataset, while there is no significant correlation within each lake.

assessment methods of neighboring countries, no phytobenthos method exists for lakes in Norway. To compare phytobenthos with macrophyte and phytoplankton assessment, we therefore applied the Periphyton Index of Trophic status PIT (Schneider and Lindstrøm, 2011), which was developed and intercalibrated for use in Norwegian rivers and streams, and applied the same status class boundaries as for rivers and streams with a corresponding water chemistry to the study lakes (i.e. the boundaries given for rivers with a Ca concentration > 1 mg/l). Our decision to use indicator values developed for lotic waters also in lakes was based on earlier experience from macrophytes, which showed that species-specific eutrophication scores between independently developed stream and lake metrics were correlated with each other (Schneider, 2007). Generally, the similarities between rivers and the shallow part of lake littoral zones mean that concepts should be transferable (DeNicola and Kelly, 2014). We therefore feel confident that the overall assessment of a lake using a river eutrophication metric is not far from the truth. There is however, without doubt, some uncertainty in the status class boundary values, as well as in individual species scores. However, phytoplankton (Snåsavatnet) and macrophytes (Selbusjøen) indicated “good” ecological status, respectively. This means that to be more sensitive than macrophytes and phytoplankton, phytobenthos would have to indicate “moderate” ecological status. However, 83% of the phytobenthos taxa found are associated with very low nutrient concentrations in Norwegian lotic waters (i.e. indicating “high” status). Despite the uncertainties related to the application of a river index to a lake, the strong dominance of taxa associated with very nutrient poor conditions makes it highly unlikely that the “true” phytobenthos status is “moderate”, i.e. worse than macrophytes and phytoplankton. We therefore conclude that in our two study lakes phytobenthos did not provide additional important

information to macrophytes and phytoplankton for surveillance monitoring. This is despite the fact that in both lakes there were potential local sources of nutrient input, which phytobenthos is assumed to react faster to than macrophytes (DeNicola and Kelly, 2014, and literature cited therein). Differences between the different ways of calculating an overall status class from site-specific results (averaging sites versus creating an overall lake taxon list, complete versus “strictly benthic” taxon lists) were minor and did not affect status classification.

Phytoplankton, macrophytes and phytobenthos are all sensitive to eutrophication. However, they differ in reaction time and in access to sediment nutrients. Macrophytes have a longer life span and persistent propagules, and therefore respond more slowly than phytoplankton or phytobenthos to changing environmental conditions (Schneider et al., 2012; Eigemann et al., 2016). Macrophytes can take up nutrients from both water and sediment (Madsen and Cedergreen, 2002), while phytoplankton and phytobenthic algae have direct access only to water nutrients (although epipelagic periphyton may play a role in the release of sediment-associated phosphorus; Lowe, 1996). The main nutrient source for phytoplankton therefore is the pelagic (phytoplankton samples are generally taken in the middle of the lake), for phytobenthos the littoral water, and for macrophytes the littoral sediment in addition to the water. In Snåsavatnet, phytoplankton indicated slightly worse ecological status than macrophytes and phytobenthos (Table 1). This may indicate a slightly enhanced availability of nutrients from the pelagic compared to the littoral. Indeed, Snåsavatnet has slightly humic water (4 mg/l TOC) while the littoral sediment mainly consists of gravel and sand (pers. obs.). Dissolved organic matter forms a large potential source of nutrients and carbon for phytoplankton (Vahatalo et al., 2003). In contrast, macrophytes indicated slightly worse ecological status than phytoplankton and phytobenthos in Selbusjøen (Table 1). This indicates an enhanced nutrient availability from the littoral sediment and may possibly be explained by the water level fluctuations caused by regulation (6.3 m between upper and lower regulation level), because re-wetting of desiccated sediments results in an initial flush of available nutrients, which can be incorporated into macrophyte biomass (Baldwin and Mitchell, 2000).

4.2. Richness and diversity patterns

There were generally more phytobenthos than macrophyte taxa per site (Fig. 3). This meets expectations, particularly in lakes subject to water level fluctuations, such as Snåsavatnet and Selbusjøen. In lakes subject to hydrological pressure, the frequency of occurrence of many macrophyte species may decline (Boschilia et al., 2012). Likewise, the observation that fewer species occurred per site in Selbusjøen than in Snåsavatnet (both for macrophytes and phytobenthos) is likely explained by the greater water level fluctuations in Selbusjøen (the difference between upper and lower regulation level is 6.3 m in Selbusjøen, compared to 1.4 m in Snåsavatnet). Our data therefore support the assumption that water level fluctuations lead to a depauperate littoral vegetation. This notwithstanding, the macrophyte vegetation was

Table 2
 Summary of significant ($p < 0.05$) fitted regression models. Models in bold and italic have an adjusted $R^2 > 0.2$, and contain maximum three model parameters (i.e. do not obviously suffer from overfitting). Response parameters with the extension 2 were calculated from the strictly benthic taxon list. Abbreviations for model parameters are explained in Table S3.

ARS0 site index classes	response	best model				interpretation		
		complete dataset	Adj. R^2	Selbusjøen	Adj. R^2		Snåsavatnet	Adj. R^2
ARS0 site index classes	log PIT	no model significant		no model significant		no model significant	0.37	no clear pattern
	log PIT2	8.37e-01 + 2.62e-05*bland_high + 4.59e-06*jordbruk - 5.24e-05*lauv_high - 3.16e-05*snaumark	0.2	no model significant		no model significant		no clear pattern
	log algal cover	<i>-2.18e-01 + 1.06e-04*bar_low - 4.1e-05*bar_med</i>	0.33	<i>-2.17e-01 + 4.21e-05*bygg - 3.94e-05*bar_med - 2.78e-05*jordbruk</i>	0.34	<i>-8.80e-02 + 9.57e-05*bar_low + 1.07e-03*snaumark</i>	0.36	increased cover at sites with low index class coniferous forest, sparsely vegetated and urban areas; decreased cover at sites with medium index class coniferous forest and agricultural areas
	log cover cyanobacteria	no model significant		<i>-1.23e+00 - 1.91e-01*beite + 6.48e-05*bygg</i>	0.23	no model significant		no clear pattern
	log cover algae	<i>-5.68e-01 + 1.19e-04*bar_low - 4.26e-05*bar_med - 6.27e-04*lauv_med</i>	0.31	no model significant		<i>-3.83e-01 + 1.05e-04*bar_low + 1.23e-03*snaumark</i>	0.35	similar to total algal cover
	sqrt TIC	<i>9.65 - 5.46e-04*myr - 2.25e-05*jordbruk + 2.3e-04*snaumark</i>	0.46	<i>9.66 - 4.44e-04*myr - 2.23e-05*jordbruk</i>	0.47	no model significant (but a model with $p = 0.06$ indicates that bar_low and snaumark lead to a higher TIC)		wetlands and agriculture decrease the TIC, sparsely vegetated areas and low quality coniferous forest increase the TIC
	taxon richness macrophytes	9.79 - 3.17e-04*bar_med - 3.86e-03*lauv_med - 2.18e-04*bar_high + 3.38e-04*bland_high	0.44	7.93 - 2.51e-04*bar_med - 3.12e-03*lauv_med - 2.24e-04*bar_high + 7.93e-04*myr	0.64	8.62 - 4.0e-03*snaumark - 7.24e-05*bar_low + 3.65e-03*bland_high - 1.4e-02*lauv_high + 2.74e-04*bygg + 1.14e-04*jordbruk	0.6	no consistent pattern; results suffer from model overfitting
	taxon richness cyanobacteria	no model significant		3.24 + 1.02e-04*bar_med + 3.81e-04*bland_imp + 1.13e-02*bar_imp + 9.03e-05*bar_high	0.3	no model significant		no clear pattern
	taxon richness algae	1.27e+01 - 1.47e-03*snaumark - 1.41e-04*bar_med - 2.27e-03*lauv_med	0.18	no model significant		no model significant		no clear pattern
	taxon richness benthic algae	17.21 - 0.0016*snaumark - 0.0032*lauv_medium	0.16	no model significant		no model significant		no clear pattern
	taxon richness cyanobacteria2	no model significant		3.120 + 1.0e-04*bar_med - 6.42e-04*myr + 3.97e-04*bland_imp + 6.85e-04*snaumark + 6.56e-05*bar_high	0.5	no model significant		no clear pattern
	taxon richness algae2	no model significant		no model significant		no model significant		no clear pattern
	taxon richness benthic algae2	13.02 - 0.00078*myr - 0.0019*lauv_med + 0.0047*bar_imp	0.13	no model significant		no model significant		no clear pattern
	complete dataset	complete dataset	Adj. R^2	Selbusjøen	Adj. R^2	Snåsavatnet	Adj. R^2	

(continued on next page)

Table 2 (continued)

CORINE	response	best model				interpretation		
		8.28e-01 + 1.40e-05*disc.urb + 3.93e-06*compl.cult	0.25	7.92e-01 + 5.41e-05*disc.urb	0.27		8.69e-01 + 1.1e-05*disc.urb + 3.19e-06*compl.cult	0.21
	log PIT	8.57e-01 + 1.49e-05*disc.urb + 5.09e-06*compl.cult	0.2	8.08e-01 + 5.71e-05*disc.urb	0.21	3.21e-01 - 3.93e-05*compl.cult + 3.46e-05*conf.forest	0.22	higher (more eutrophic) PIT at sites close to urban and cultivated areas similar to PIT, but less clear
	log PIT2	-2.48e-01 - 4.48e-05*shrub + 2.84e-05*conf.forest	0.11	no model significant		no model significant		no clear pattern
	log algal cover	no model significant		no model significant		no model significant		no pattern
	log cover cyanobacteria	no model significant		no model significant		no model significant		no pattern
	log cover green algae	no model significant		no model significant		no model significant		no pattern
	sqrt TIC	no model significant		no model significant		no model significant		no pattern
	taxon richness macrophytes	9.18e+00 - 3.19e-04*shrub - 8.51e-05*conf.forest	0.21	7.31 - 2.37e-04*shrub - 1.50e-04*conf.forest	0.28	no model significant		fewer macrophyte species at sites with coniferous forest and transitional woodland-shrubs, but Snåsavatnet did not show such a relationship
	taxon richness cyanobacteria	no model significant		no model significant		no model significant		no pattern
	taxon richness green algae	no model significant		no model significant		no model significant		no pattern
	taxon richness benthic algae	no model significant		no model significant		no model significant		no pattern
	taxon richness cyanobacteria2	no model significant		no model significant		no model significant		no pattern
	taxon richness algae2	no model significant		no model significant		no model significant		no pattern
	taxon richness benthic algae2	no model significant		no model significant		no model significant		no pattern

not seriously affected by regulation in either of the two lakes. This is evidenced by the macrophyte index WIC, which was developed to indicate water level drawdown (Mjelde et al., 2013). The WIC indicated “high” status for both Snåsavatnet and Selbusjøen (Lyche Solheim et al., 2017). Although the WIC is still regarded as preliminary for some lake types and regulation regimes, it indicates that the water level fluctuations did not lead to a seriously depauperate macrophyte vegetation in any of the two lakes.

There was a weak correlation between macrophytic and phytobenthic taxon richness across the sites (Fig. 3a). However, the correlation did not occur when phytobenthic taxa lacking hold-fast structures were excluded (Fig. 3b). This is consistent with the observation that many phytobenthos taxa (e.g. *Closterium* species) occur loosely attached to submerged macrophytes (John et al., 2011). They may, however, be dislodged by wave action, such that their occurrence at a particular site may not necessarily be related to site-specific pressures. Taxon richness is generally affected by habitat complexity, disturbance, and chemical conditions such as nutrient concentrations (Biggs and Smith, 2002; Casartelli and Ferragut, 2018). The main difference in the exposure of macrophytes and phytobenthos to habitat complexity, disturbance, and chemical conditions at each sampling site is related to the depth where the samples were taken. While phytobenthos was sampled from the shallow littoral, the entire littoral down to the lower vegetation depth was taken into account for macrophytes. The lower vegetation depth was around 5 m in both lakes (Lyche Solheim et al., 2017). This means that within site variability in habitat complexity, disturbance and water chemistry between 1 and 5 m depth has affected macrophyte, but not phytobenthos richness. This likely explains the absence of a correlation between macrophyte and phytobenthos richness, when only strictly benthic algal taxa were considered (Fig. 3b).

The different sampling depth likely also explains why phytobenthos but not macrophyte assemblages at sites 11, 15 and 17 in Snåsavatnet were different from the other sites (Fig. 2a and b). These sites are affected by trampling (11), artificial substrate (15), as well as trampling combined with pollution from cattle (site 17). These disturbances mainly affect the shallow littoral, which explains why phytobenthos assemblages exclusively taken in the shallow littoral react most. In Norway, macrophyte vegetation is not recorded in “depth zones”, such as for example in Germany (Schaumburg et al., 2004). It is possible that the shallow littoral macrophyte vegetation would have indicated these stressors, but our data do not allow such an analysis. In Snåsavatnet, site 10 is situated at a camp-ground, which implies that the site likely is affected by recreational activities such as swimming, fishing, and boating. These disturbances led to differing macrophyte as well as phytobenthos assemblages (Fig. 2a and b).

While taxon richness only considers how many taxa occur at a site, assemblage similarities additionally consider taxon identities and abundances. Dissimilarities in phytobenthos assemblages among sites were significantly correlated with dissimilarities in macrophyte assemblages, although the explained variance was low (Fig. 4). This means that sites having a similar macrophyte assemblage are likely to also have a similar phytobenthos assemblage, and indicates that macrophytes and phytobenthos give partly redundant information. In accordance with the results from the taxon richness patterns, the correlation was stronger when all phytobenthic algae (including those that lack hold-fast structures) were taken into account. This does not, however, exclude the possibility that some additional important information is contained. In Selbusjøen, 7 out of 20 sites had depauperate or absent macrophyte vegetation. This likely is related to natural conditions (steep and stony sediment) combined with water level fluctuations caused by regulation. At the same time, phytobenthos assemblages were not particularly species poor. This suggests that phytobenthos assemblages may generally be useful indicators at sites with little or no macrophyte vegetation.

4.3. Site-specific eutrophication effects

Macrophytes and phytobenthos commonly react to increased nutrient input by increased growth, as well as shifts in species composition towards taxa thriving in nutrient rich conditions (Lowe, 1996; Egertson et al., 2004). In the field, this is often immediately visible as increased algal cover, often in form of green algae or cyanobacteria (Hart et al., 2013), or as macrophyte assemblages dominated by taxa known to thrive in nutrient rich conditions (e.g. *Elodea canadensis*, *Sparganium emersum*; Melzer, 1999; DG, 2013). Taxon specific indices, such as the TIC and the PIT, are a convenient way to quantify shifts in macrophyte and phytobenthos assemblages related to nutrient inputs. Nutrient input into lakes can principally occur via aerial deposition, or from surface or subsurface inflow (Lowe, 1996). While aerial deposition should affect the entire lake more or less homogeneously, surface and subsurface inflow is more local, and therefore primarily leads to local changes in macrophyte and phytobenthos biomass and species composition. Such changes have long been used to detect local sources of nutrients around the shoreline of lakes (Melzer, 1999). Although the macrophyte index TIC applied in Norway is not designed to indicate site-specific differences in nutrient input, changes in macrophyte species composition in response to local nutrient input should nevertheless be reflected in the TIC. Likewise, changes in the phytobenthic algal species composition towards species that thrive in nutrient-rich conditions should be reflected in the PIT. In the absence of site-specific water chemistry data, we estimated site-specific P-export from CORINE land cover and AR50 site index classes. This is a common method which has been successfully applied before to infer water quality data, albeit at a larger scale (lake scale, instead of site-scale; e.g. Smith et al., 2005; Uuemaa et al., 2005). However, site-specific P-export was uncorrelated to any of the macrophyte or phytobenthos response parameters, with the exception that more macrophyte taxa occurred at sites with a higher nutrient export (Fig. 6). The result that increased nutrient input is related to increased macrophyte species richness is consistent with the hump-shaped relationship between productivity and species-richness: species richness generally peaks at intermediate levels of productivity (Dodson et al., 2000). Oligotrophic Scandinavian softwater lakes, such as Snåsavatnet and Selbusjøen, generally have low macrophyte species richness (Murphy, 2002), and this means that an increase in nutrient input will lead to an increase in species richness. The absence of correlations between estimated P-export and any of the other macrophyte and phytobenthos response parameters likely is explained by the overriding effect of other natural conditions and stressors, which also affect macrophytes and phytobenthos, such as substrate type, sediment slope, grazing, turbidity, temperature, and mechanical disturbance by e.g. wave action (Lowe, 1996; Weisner et al., 1997; Lehmann, 1998), combined with uncertainties in the estimation of P-export (Smith et al., 2005).

Another factor which likely contributed to the absence of correlations between P-export and macrophyte and phytobenthos assemblages, is the relatively short gradient covered by the eutrophication indices TIC and PIT in our data. The TIC index generally ranges from -100 to +100 (DG, 2013), but among the 40 sites sampled in Snåsavatnet and Selbusjøen, all sites except 2 had values above 70 (Fig. 5). Likewise, the PIT can take values between 2 and 68 (Schneider and Lindström, 2011), but the 40 sites in our dataset all had values below 14 (Fig. 5). Short gradients can prevent the detection of relationships between stressors and responses (Feld et al., 2016). The short gradients likely also explain the absence of a correlation between site specific TIC and PIT values.

In order to explore the data in greater detail, we also tested if macrophyte and phytobenthos response parameters could be explained by land use adjacent to the sampling sites (Table 2). Although phytobenthos cover was consistently related to land use (Table 2), our results indicate that using algal cover as an indicator of site-specific nutrient input is overly simplistic. Increased algal cover was associated with urban areas as well as with coniferous forest, while agriculture seemed

to decrease algal cover (Table 2). Poor relationships between phyto-benthos biomass and lake trophic status have been observed before (Lowe, 1996), and are explained by the many natural conditions and stressors which interact to affect phyto-benthos biomass. In addition, the most common and prevalent taxa in both Snåsavatnet and Selbusjøen (*Cosmarium*, *Mougeotia*, *Spirogyra*, *Zygnema*, *Oedogonium*) are known to occur in a wide range of habitats, including nutrient-poor as well as nutrient-rich waters (John et al., 2011).

Wetlands and agricultural areas tended to be related to a more eutrophic macrophyte index TIC. While the influence of agriculture is obviously caused by nutrient run-off from fertilized land, a eutrophying impact of wetlands may seem counter-intuitive. However, organic sediments generally contain more nutrients than mineral sediments (Schneider and Melzer, 2004), which often dominated in Snåsavatnet and Selbusjøen (pers. obs.). Since sediment nutrients are available to macrophytes (Madsen and Cedergreen, 2002), more “eutrophic” macrophyte assemblages may occur on organic sediments in generally nutrient poor lakes. More data are needed before the effects of naturally increased sediment nutrients can be differentiated from anthropogenically increased nutrient input, but our data indicate that the TIC is a promising tool which could be improved to detect site-specific nutrient input. Likewise, urban and cultivated areas were associated with a more eutrophic PIT index. Both urban and cultivated areas are known to have a higher nutrient export than natural vegetation (Smith et al., 2005). Although the explained variance was low (Table 2), this indicates that the PIT may be a promising tool to detect site specific nutrient input.

5. Conclusions

Both Snåsavatnet and Selbusjøen are generally nutrient poor, but there nevertheless are some potential local sources of nutrients (villages, agriculture, campground) around the shore. In addition, both lakes are regulated, albeit to a varying degree. In such a situation, phyto-benthos is expected to be potentially more informative with respect to eutrophication than macrophytes and phytoplankton (DeNicola and Kelly, 2014). However, our data provided no indication that phyto-benthos would indicate a worse overall ecological status than macrophytes and phytoplankton. Therefore, although we only have data from two lakes, our results provide no support for generally including phyto-benthos into surveillance monitoring of lakes in Norway.

Overall, site-specific taxon richness and assemblage similarities were loosely correlated between macrophytes and phyto-benthos. This indicates that similar natural conditions and stressors affected macrophytes and phyto-benthos at the sampling sites, and means that macrophytes and phyto-benthos do give partly redundant information. Since the explained variance was low, however, the results could actually provide some useful additional information. The additional information provided by phyto-benthos likely is most relevant at sites with little or no macrophyte vegetation. In Selbusjøen, 7 out of 20 sites had depauperate or absent macrophyte vegetation (≤ 4 macrophyte taxa occurred). In contrast, the phyto-benthos assemblages at these sites contained between 9 and 18 taxa. This indicates that phyto-benthos may generally be useful in lakes with little or no macrophyte vegetation.

We found that using algal cover as an indicator of site-specific nutrient input is overly simplistic. This was because increased algal cover was associated with potentially high (urban areas) as well as with potentially low sources of phosphorus (coniferous forest) to water bodies, while other potential high sources of phosphorus (agriculture) were related to decreased algal cover. Wetlands and agricultural areas tended to be related to a more eutrophic macrophyte index TIC. Our data therefore indicate that the macrophyte index TIC may be developed into a tool for detecting site-specific nutrient input. This requires more data, however, such that the effects of naturally increased sediment nutrients can be differentiated from anthropogenically increased nutrient levels. Urban and cultivated areas were associated with a more eutrophic

phyto-benthos index PIT. This means that the PIT, despite being developed for lotic waters, may be used to detect site specific nutrient input, and indicates that the PIT may be useful for operational and investigative monitoring. More data are required, however, for setting status class boundaries.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at <https://doi.org/10.1016/j.limno.2019.02.001>.

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