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Abstract

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PRIMARY RESEARCH PAPER

Coherence of phytoplankton and attached diatom-based ecological status assessment in Lake Balaton

Luciane O. Crossetti · Csilla Stenger-Kovács · Judit Padisák

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Abstract Coherence between ecological status assessment by phytoplankton and attached diatoms was analyzed in the littoral zone of Lake Balaton. Sampling of periphytic diatoms, phytoplankton, and water were carried out at ten different littoral sites in the northern and southern shores of the lake for a year. Phytoplankton species were sorted into functional groups and ecological status was assessed by means of the phytoplankton assemblage Q index. The index TDIL was calculated using quantitative attached diatom data. Significant differences were found between the ecological assessments based on phytoplankton and phytobenthos metrics, both seasonally and spatially. The Q index indicated ecological states varying from bad to good, while the average of diatom indices varied from moderate to high conditions. The Q index provided more realistic ecological status of Lake Balaton, compared with trophic status based on TP values, especially in the summer period. Differences in the response-time indication of phytoplankton and attached diatoms suggest that lack of coherence should also be expected between the responses of other BQEs.

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Introduction

Algae are small, live short lives and provide fast response to environmental pressures. These simple features make them suitable for testing ecological hypotheses. The first competition models and their experimental tests used phytoplankton species as model organisms (Tilman, 1977). Natural phytoplankton assemblages proved to be very useful in testing ecological concepts like the Intermediate Disturbance Hypothesis (Sommer et al., 1993; Reynolds et al., 1993) or the Equilibrium Concept (Naselli-Flores et al., 2003).

Short generation times of phytoplankton species make them responsive indicators of environmental changes. Attached diatoms were less frequently used for hypothesis testing although, in theory, their assemblages might be described using similar models. The first attempts (Passy, 2007) of using attached diatom functional groups are promising but test studies are rare (Rimet & Bouchez, 2011; Stenger-Kovács et al., 2012). Regarding phytoplankton, the functional groups are based on the morphological, physiological, and ecological similarities of the species (Reynolds et al., 2002), being widely used (Padisák et al., 2009).

Since freshwater quality in the industrialized world has begun to degrade, an increasing need to use biota as water quality indicators has emerged. The saprobic system was developed in the early twentieth century and has been still applied despite its recognized weaknesses. Heavy organic pollution can indeed be monitored by the saprobic system but it is unable to distinguish between effects of pollution and autochthonous organic matter content. When eutrophication became a central issue in limnology, proxy variables (like chlorophyll-*a* concentration or Secchi transparency in deep lakes) of water quality assessment gained overall acceptance. The weaknesses of the saprobic system and the simple metrics are that they do not take into account differences in stream or lake types. As an extreme case, the same metrics and scales were used to assess ultraoligotrophic lakes and old, pristine oxbows and other naturally eutrophic lakes and ponds (Borics et al., 2012).

Landscape-oriented ecological concepts such as habitat fragmentation were developed relatively early to explain terrestrial biodiversity homogenization and degradation of ecosystems. Curiously, such concepts began penetrating into aquatic ecology and water quality assessment with a considerable delay. At present, it is becoming widely accepted that degradation of surface waters is attributable to a large extent to non-pollution originated human impacts like canalization of river basins, shore-line or bank protection constructions in lakes, water stand or flow regulations, etc.

The first coherent concept that accepts these hydromorphological modifications and tries to handle them at a similar rank with organic/inorganic pollution is the Water Framework Directive (WFD) which was issued by the EC in 2000 (Directive, 2000). Thereby, ecoregions were established and stream and lake types were defined on the basis of some robust parameters (i.e., geology, catchment area or surface area, altitude, depth, etc.). According to the WFD ecological status is 'an expression of the quality of the structure and functioning of aquatic ecosystems associated with surface waters' (Directive, 2000: Article 2: 21). Despite the fact that WFD allows for an incomparably more ecology-orientated assessment of ecosystems ecological status (type specificity, species-based indices, etc., see Padisák et al., 2006), it raises an unrealistic target in requiring a "one out, all out" assessment (Nõges et al., 2009). It means that status is assigned as the lowest of that achieved by one of five Biological Quality Elements (BQEs: fish, macroinvertebrates, macrophytes, phytobenthos, and phytoplankton). However, the BQEs may not respond equally to the same stressors, leading to different ecological status assessment, and, in this sense, the one out all out concept may downgrade sites unjustifiably (Moss, 2007). Because of its serious ecological limitations (for example, disregarding scale-dependence) the "one out, all out" concept needs to be evaluated.

A number of studies consider either phytoplankton or phytobenthos in the context with the WFD guidelines. Indices for both groups were separately proposed and/or applied aiming at the assessment of ecological status of lakes and rivers (e.g., Schaumburg et al., 2004; Ács et al., 2004; Padisák et al., 2006; Borics et al., 2007; Stenger-Kovács et al., 2007; Ptacnik et al., 2009; Nõges et al., 2010; Abonyi et al., 2012; Stanković et al., 2012). Nonetheless, there has been an overall lack of integrative studies comparing the efficiency of such methods in the determination of the ecological status in surface waters (Blanco et al., 2007). Recognizing this fact, some research focused on using different groups of biota included in the WFD to access the ecological status, such as epilithic diatoms and macroinvertebtates (Blanco et al., 2007); diatoms, macroinvertebrates, and macrophytes (Triest et al., 2004); macrophytes and diatoms (Furse et al., 2006); fish, macrophytes, benthic diatoms, and macroinvertebrates (Johnson et al., 2006); invertebrates, oligochaetes, diatoms, and fishes (Lafont et al., 2001); phytoplankton and macrophytobenthos (Sagert et al., 2005); phytoplankton, phytobenthos, and macrophytes (Cellamare et al., 2011); and phytoplankton, macroalgae, and macroinvertebrates (Simboura et al., 2005), sometimes indicating substantial dissimilarities between the BQEs' responses.

Phytoplankton and phytobenthos share a common feature: species of both types have short generation times. However, they are not expected to respond equally to the same environmental impacts. Phytoplankton tends to respond faster to environmental changes since it drifts freely in horizontal water currents of lakes and its biomass doubling time is approximately twice as fast as that of periphyton

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(~3 days against about a week; Padisák, 1994). On the other hand, phytobenthos does not drift in adverse conditions due to its sessile life form (Stevenson, 1996), increasing the ability to hold information about previous environmental events in its structure, being influenced by both properties of the open water and local impacts from the shore. So, we hypothesize that the ecological status of Lake Balaton assessed using phytoplankton and phytobenthos might be different since the responses of both communities to environmental conditions must be different in time. Then, one of these communities must provide a more realistic ecological status of Lake Balaton. In this sense, the objective of the present study was to analyze the coherence between ecological status assessment by phytoplankton and attached diatoms in the littoral zone of Lake Balaton, Europe's largest shallow lake, focusing on the following questions: (1) What is the ecological status of Lake Balaton using phytoplankton and phytobenthos based indexes? (2) Is there coherence between the classifications given by each of these biological quality elements?

Materials and methods

Lake Balaton (43°3′50″–46°42′6″N, 17°14′58″–18°10′28″E, 104.5 m a.s.l.) is the largest shallow lake in Central Europe. It has a surface area of 593 km^2 and is 77.9 km long and 9 km wide on average (maximum width 15 km). The lake has a mean depth of 3.14 m (Z_{max} = 11 m) and its theoretical retention time is 3–8 years (Padisák, 1992). Several studies have been published about the lake's phytoplankton and phytobenthos and its eutrophication and restoration histories (Padisák & Reynolds, 1998; Istvánovics et al., 2007). Differences in the ecological status of Lake Balaton's eastern and western basins have been described in previous studies and have been found to originate from ontogenetic reasons and catchment morphology (Padisák et al., 2006). This study took place at 10 sampling stations in the littoral region of Lake Balaton (Fig. 1): Balatonkenese (D1), Siófok (D2), Szántód Rév (D3), Balatonlelle (D4), Keresztúr (D5), Vonyarcvashegy (E1), Badacsony (E2), Szepezdfürdő (E3), Tihany (E4), Csopak (E5).

Water samples were taken with plastic bottles at approximately 10 cm below the water surface, monthly from March 2006 to February 2007. Temperature, pH, conductivity, dissolved oxygen, and oxygen saturation were measured using standard electrodes in the field. The following variables were determined in the laboratory: chemical oxygen demand (COD), soluble reactive phosphorus (SRP), total phosphorus (TP), nitrite (NO_2^--N), nitrate (NO_3^--N), ammonium (NH_4^+-N), total nitrogen (TN), and soluble reactive silica (SRSi) (APHA, 1995). Abiotic data were compared with those found in the monitoring service of Lake Balaton (North Transdanubian District Water Authority—Székesfehérvár; non-published data) to ensure the reliability of the observed values.

Phytoplankton samples for quantitative study were taken at approximately 10 cm below the water surface with a 200-ml flasks and immediately fixed with acetic lugol 1% in the littoral zone adjacent to open water (approximately 10–15 m from the shore), avoiding metaphyton sampling as much as was possible. The

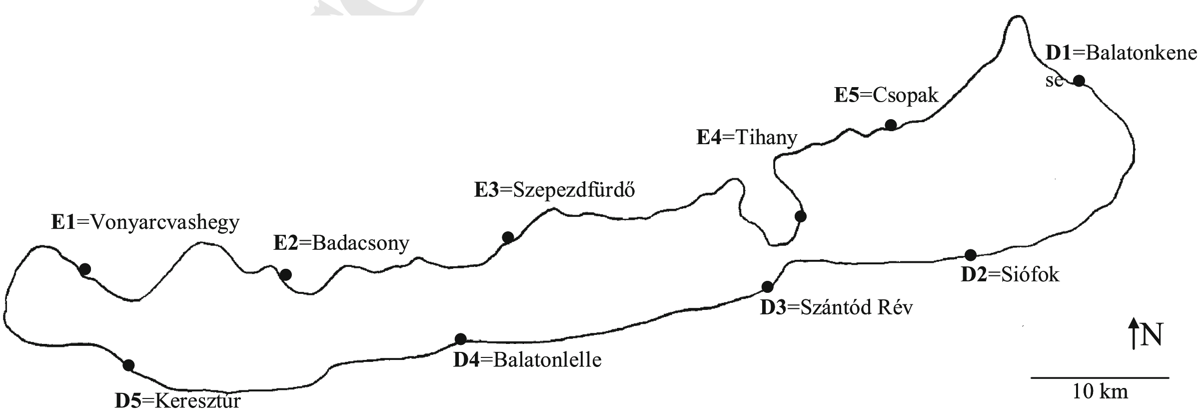

Fig. 1 Sampling stations on the northern (E1, E2, E3, E4, E5) and southern (D1, D2, D3, D4, D5) shorelines of Lake Balaton

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analysis followed Utermöhl's (1958) method. At least 400 settling units were enumerated in every sample giving a counting accuracy of ±10% (Lund et al., 1958). Biomass (mg l^{-1}) was estimated using geometric approximation and the OPTICOUNT (2008) software was applied for counting and calculations. Phytoplankton biomass values were compared with the ones found in the literature (Padisák et al., 2010) to ensure the reliability of the data found in Lake Balaton. Phytoplankton identification was based on specific literature (e.g., Huber-Pestalozzi, 1955; Komárek & Fott, 1983; Popovski & Pfiester, 1990; Komárek & Anagnostidis, 1999, 2005; Komárek & Komárková, 2004, 2006; Komárek & Zapomělová, 2007, 2008).

Attached diatoms were removed from five randomly selected reed stems with brushes. Identification of diatoms followed the peroxide method (Ács & Kiss, 2004) with embedding in Zrax©. At least 400 valves were counted under a light microscope to estimate relative abundances. Additionally, SEM investigations (Kiss, 1986) were applied whenever it was necessary. Diatoms identification was based on specific literature (e.g., Krammer & Lange-Bertalot, 1991–2000; Krammer, 2002; Lange-Bertalot, 1995–2002).

The functional group approach (Reynolds et al., 2002; Padisák et al., 2009) was applied to the phytoplankton species. Due to the high occurrence of non-planktonic diatoms eventually found in offshore phytoplankton samples (**MP** codon according to Padisák et al., 2009) they were not included in the phytoplankton analysis in order to avoid noise and autocorrelations between phytoplankton and phytobenthos ecological assessment, considering the objective of the present study. The **MP** species represented an average of 62% of phytoplankton biomass in the open water samples and their actual share depends on the absolute value of phytoplankton biomass (the less total biomass the higher share of **MP** diatom) and intensity of storm activity (Padisák et al., 1988, 1990). The ecological status assessment from the phytoplankton data was based on the Q index (Padisák et al., 2006), which takes into account the relative contribution of phytoplankton functional groups in the total biomass, as well as a factor number determined for each group according to the type of water body. The factor numbers considered were the ones determined in Padisák et al. (2006) according to the typology of Hungarian lakes (Lake Balaton = type 1). Concerning diatom data, ecological status assessment was performed by the Trophic Diatom Index for Lakes (TDIL) (Stenger-Kovács et al., 2007), developed for Hungarian lakes, which is based on weighted average method, and takes into account a TP model, the relative abundance, the sensibility and the trophic indicator value of the species. Recently, TDIL, along with Indice Biologique Diatomees (IBD, Lenoir & Coste, 1996) were recommended for ecological status assessment based on benthic diatoms in Lake Balaton (Bolla et al., 2010). In the present study, only TDIL, which is type specific, was considered. IBD was specifically designed as an index of general water quality (Lenoir & Coste, 1996) and was not adapted to the region and the type of lake, as recommended by WFD intercalibration protocols (Pollard & van De Bund, 2005). The Q index and TDIL separated the ecological assessment into five classes: 0–1: bad; 1–2: poor; 2–3: moderate; 3–4: good; and 4–5: high. Boundary data in this study were set as described in Padisák et al. (2006) and Stenger-Kovács et al. (2007). Lake Balaton represents the Type 1 in Hungarian typology of lakes. There is no other lake in this category, therefore cross-testing of metrics and their boundary layers is impossible. The Q index, during its development, was tested against phytoplankton biomass and % of alien species (mostly cyanobacteria) as demonstrated in Padisák et al. (2006). Later, historical phytoplankton data were used to set reference state for the phytoplankton metrics (Hajnal & Padisák, 2008). In all these comparisons the Q index proved to be a useful tool to trace changes in ecological status of the lake both in eutrophication and restoration period. It is important to note here that the philosophy of the Q index is different from other metrics. While most others strongly rely on trophic status and TP as pressure variables (see Carvalho et al., 2013), the Q index measures the deviation from the phytoplankton composition that is considered optimal with no regards of the pressure variable which, apart the changes in trophic status, could be salinity change or change in dissolved organic materials. This provides a flexibility of the Q index though does not ease its testability. Additionally, no long-term TP data on Lake Balaton are available; in studies focusing on eutrophication history of the lake, Biologically Available Phosphorus (BAP) load in used as deriving force (Istvánovics et al., 2007), moreover, phytoplankton composition

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was proved to be sensitive to BAP-load and loading N/P ratio (Padisák & Istvánovics, 1997).

The TDIL was developed on data-base from 83 Hungarian shallow lakes using diatom and TP data with high statistical accuracy (r^2: 0.96; Stenger-Kovács et al., 2007).

For comparisons among phytoplankton- and phytobenthos-based ecological status assessments, two sample t tests were employed. Multiple regressions were applied to verify the accuracy of the metrics of the BQEs concerning the variation of environmental variables (SRP, nitrite, TP, temperature, and conductivity). These analyses were performed with the software Systat version 12, for Windows (Systat, 2007). Environmental data were used to support the quality assessment and explored by means of multivariate descriptive analysis and comparisons of TP values with the trophic state index boundaries of the OECD (1982), namely ultra-oligotrophic (TP < 4 μg l^{-1}), oligotrophic (10 < TP > 4 μg l^{-1}), mesotrophic (35 < TP > 10 μg l^{-1}), eutrophic (100 < TP > 35 μg l^{-1}), hypereutrophic (TP > 100 μg l^{-1}). A principal component analysis (PCA) was carried out to verify the main tendencies of the abiotic data variability in the studied period, to a covariance matrix with data transformed by 'ranging'. Integrated analysis between phytoplankton, periphytic diatoms and abiotic data was performed by the ordination method canonical correspondence analysis (CCA) to determine the ordination of phytoplankton functional groups and phytobenthos species depending on the main environmental variables. For the analysis, abiotic variables with strongest correlations in the PCA were chosen as well as attached diatom species and phytoplankton functional groups which contributed with more than 1% of total relative abundance and to the total biomass, respectively. The statistical significance was tested by a Monte Carlo permutation test. For the CCA, abiotic and biological data were log-transformed (x + 1). PCORD version 6 (McCune & Mefford, 2011) was used for the multivariate analyses.

Results

Environmental variables

Descriptive analysis of abiotic variables of Lake Balaton during the study is given in Table 1. High variability of abiotic data was observed in the littoral region of Lake Balaton due to the habitat inhomogeneity and its exposure to point-like temporary events. PCA performed with nine environmental parameters explained 60.1% of the total variance in its two first axes (Fig. 2). Temperature (r = 0.985) and dissolved oxygen (−0.782) were the most important variables in the ordination of the first axis, while nitrite (−0.754), conductivity (−0.630), COD (−0.574), and SRP (−0.551) were the main variables in the second axis' ordination. Clear seasonal gradients were observed over axis one. To the positive side of this axis, all the sample units of summer and spring were correlated to the higher temperature values. In contrast, in the negative side of axis one, sample units of winter and autumn were correlated to the higher concentration of dissolved oxygen. The second axis showed a spatial gradient. On its positive side, sample units of northern shore of Balaton were associated with higher levels of pH, and on its negative side southern samples were ordinate towards the higher values of all dissolved nutrients, conductivity, and COD.

The largest range of TP values were observed in the points E1, E3, E5, and D4 (Fig. 3). The medians showed much variation in the values of TP, classifying Lake Balaton between mesotrophic and eutrophic during the studied period. Higher concentrations of TP were observed in the summer, especially August, at various sampling points in the northern and southern shorelines of Lake Balaton, reaching the boundaries of hypertrophic condition, such as observed at E1 (550 μg l^{-1}), E2 (2925 μg l^{-1}), E3 (139 μg l^{-1}), E5 (166 μg l^{-1}), D1 (2246 μg l^{-1}), D2 (407 μg l^{-1}), and D4 (104 μg l^{-1}).

Phytoplankton and phytobenthos integrated analysis

The results of the CCA ordination with main attached diatom species with respect to environmental variables accounted for eigenvalues of 0.068 and 0.037 for the first two axes, while with phytoplankton functional groups it accounted for eigenvalues of 0.297 and 0.125. The Pearson correlation of environment-species for the first two axes of both analyses (0.718, 0.512 and 0.656, 0.521, respectively) indicated strong correlation between abiotic variables and algal species. The Monte Carlo test (99 permutations; $P < 0.05$) demonstrated that the ordination of axes 1

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Table 1 Minimum (min), maximum (max), mean values, standard error (SE), and variation coefficient (VC; %) of abiotic and biological variables on the northern (D1–D5; $n = 90$; * $n = 50$) and southern (E1–E5; $n = 90$) shorelines of Lake Balaton

Variables	North Balaton					South Balaton				
	Min	Max	Mean	SE	VC	Min	Max	Mean	SE	VC
Temperature (°C)	1.0	29.4	13.8	0.9	63	1.5	29.4	13.8	0.9	61
pH	6.5	9.4	8.0	0.1	6	5.8	9.7	8.0	0.1	6.4
Conductivity (μS)	270	1,280	714	19	26	230	1,100	700	17	22
DO (mg l^{-1})	2.0	26.9	10.2	0.5	45	2.0	16.9	9.7	0.4	37.8
SRP (μg l^{-1})	5.8	68.3	15.7	1.3	78	5.8	330.0	30.1	5.3	167
TP (μg l^{-1})	5	3,390	90	37	394	5	3,130	206	53	244
NO_3^--N (μg l^{-1})	20	14,354	677	164	230	20	1,665	406	35	83
NO_2^--N(μg l^{-1})	3	1,780	200	26	125	3	1,218	177	16	83
NH_4^+-N (μg l^{-1})	37	418	57	5	89	10	738	90	12	129
COD (mg l^{-1})	5	41	11	0	40	5	40	10	0	35
Silicate (mg l^{-1})*	2	18	7	1	65	1	16	6	1	65
Phytoplankton biomass (mg l^{-1})	0.0002	11	1.8	0.3	121	0.02	9.9	1.5	0.3	133

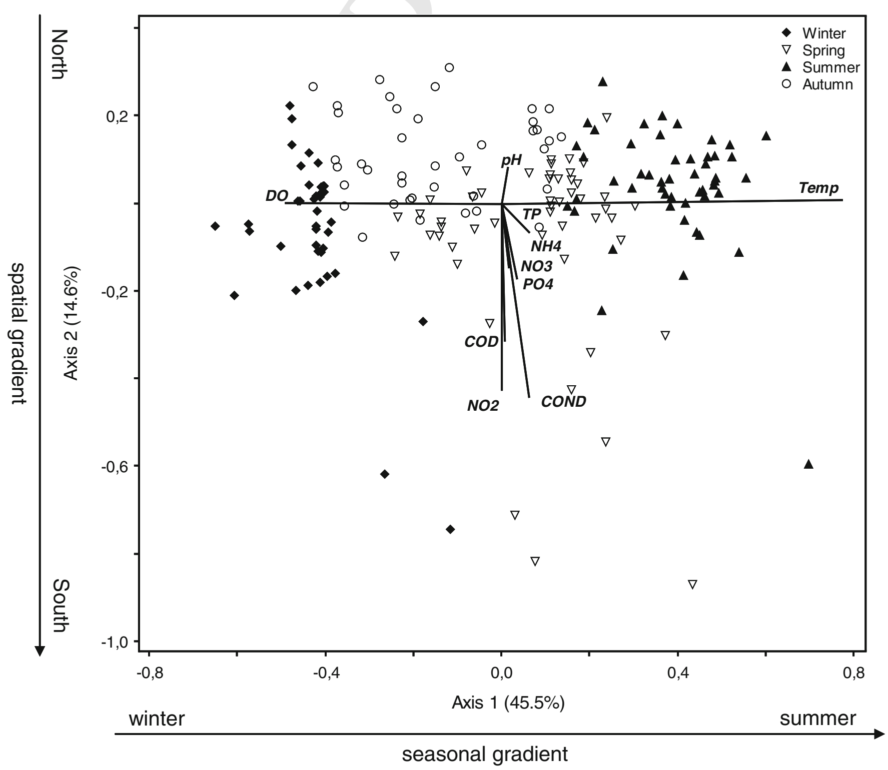

Fig. 2 Biplot of PCA for the mean values of abiotic variables in Lake Balaton ($n = 180$). *COND* conductivity, *Temp* temperature, *DO* dissolved oxygen, *pH* pH, *TP* total phosphorus, *SRP* soluble reactive phosphorus, *NH4* ammonium, *NO2* nitrite, *NO3* nitrate, *COD* chemical oxygen demand

and 2 was statistically significant ($P = 0.001$) for both BQEs (Fig. 4).

The analysis with the main attached diatom species showed through the canonical coefficients that temperature (1.013) was the most important variable to axis one ordination and SRP (0.600) and conductivity (0.407) to the ordination of axis two. Representing possible correlations between the abiotic variables and their ordination with that axis, but retaining the dependence relation between biotic and abiotic

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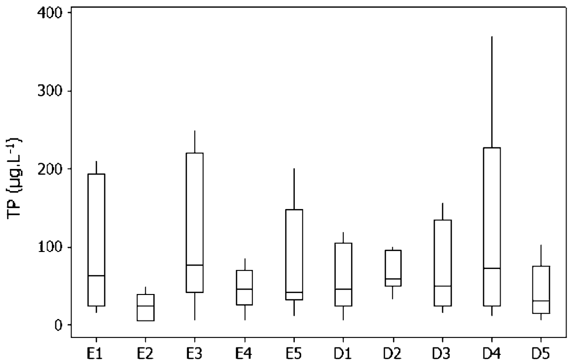

Fig. 3 Boxplot (variation of the data, median, interquartile ranges, $n = 12$) of total phosphorus (TP, µg l^{-1}) in South (D) and North (E) basins of Lake Balaton. Outliers were removed

variables, the intra-set correlations pointed out temperature (0.950) and dissolved oxygen (−0.709) as the most important variables for axis one while SRP (0.834) and conductivity (0.689) were highlighted for axis two (Fig. 4). As showed in the CCA with attached diatoms, the analysis with phytoplankton functional groups showed through the canonical coefficients that temperature (1.294) was the most important variable to axis one. For axis two, nitrite (−0.824) and dissolved oxygen (−0.646) presented the higher canonical coefficients. Considering the intra-set correlations for axis one, temperature (−0.871) was the most important variable, while dissolved oxygen (−0.687) and conductivity (0.578) were highlighted for axis two (Fig. 4).

As showed by the PCA, the diagrams of both CCAs displayed a seasonal gradient over axis one and a spatial gradient (South–North) over axis two, with higher concentrations of nutrients, especially nitrite and conductivity on the southern shore (Fig. 4). The phytobenthos species *Nitzschia palea*, *N. fonticola*, *N. paleacea* were ordinated on the positive side of the first axis associated with higher nutrient availability in western basin in the summer. *Fragilaria capucina* var. *gracilis* was associated to the higher concentrations of nitrite and SRP, in winter, in the southern shore. On the negative side of axis two, another guild was associated with lower nutrient concentrations especially in the summer period, whereas *Diatoma moniliformis* and *Gomphonema olivaceum* were associated in the winter with lower availability of nutrients and higher dissolved oxygen concentrations.

Regarding phytoplankton, CCA diagram evidenced the functional groups **H1** and $\mathbf{S_N}$ associated with higher temperature in western basin and lower values of dissolved oxygen. Higher concentration of nitrite in summer ordinate the groups $\mathbf{L_O}$ and **E**, closest to the southern shore. Most of the phytoplankton functional groups (**J**, **D**, **P**, **S1**) were closely related to the higher SRP concentrations in the spring. Higher

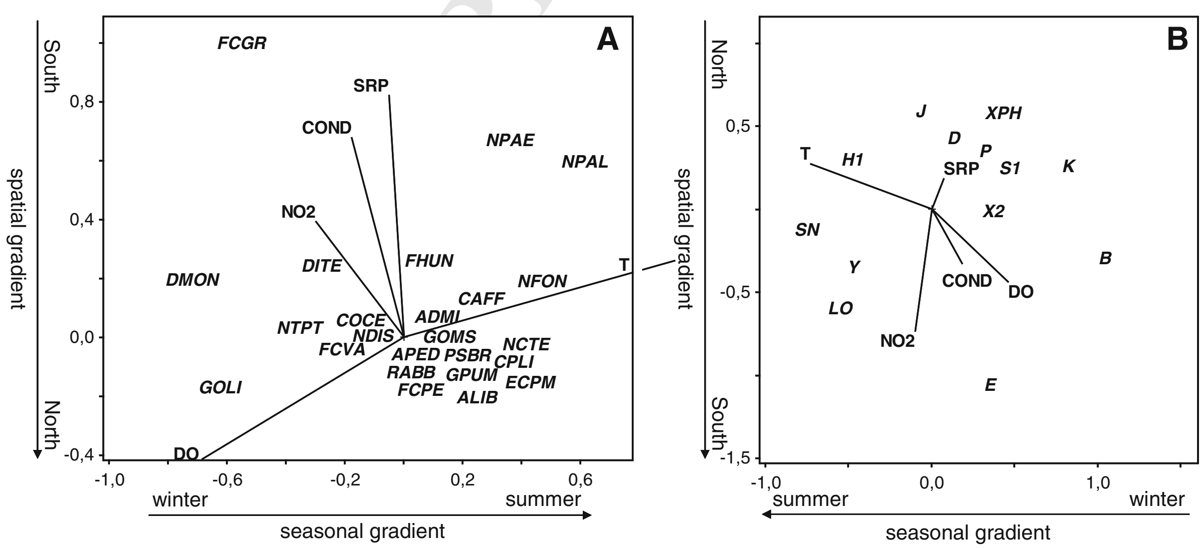

Fig. 4 CCA diagram for littoral zone of Lake Balaton, including limnological variables (explanatory variables) and benthic diatom species (**A**) and phytoplankton functional groups (**B**) (dependent variables). *DO* dissolved oxygen, *T* water temperature, NO_2 nitrite, *COND* conductivity, *SRP* soluble reactive phosphorus. Species legends are given in Table 2

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concentrations of dissolved oxygen found in winter time ordinate specifically with the functional group **B**.

Phytoplankton and phytobenthos ecological assessments

Littoral phytoplankton total biomass was mostly composed of non-planktonic diatoms (62%). Of the euplanktonic species, about 39% of total biomass belonged to Cyanobacteria, followed by high contributions of planktonic diatoms (32%) and green algae (17%). Twenty two functional groups were found, the most representative coda were: **B**, **D**, **E**, **H1**, **J**, **K**, $\mathbf{L_O}$, **P**, **S1**, $\mathbf{S_N}$, **X2**, $\mathbf{X_{PH}}$, and **Y** (Table 2).

Ecological assessment based on the Q index indicated ecological status varying from bad to good on both northern and southern shorelines of Lake Balaton (Fig. 5). Table 2 shows the most important attached diatom species and their relative contribution in the littoral region of Lake Balaton. The ecological assessment based on TDIL indicated ecological status which varied from moderate to high in both the southern and northern shorelines of Lake Balaton. Higher ecological statuses were verified in the eastern basin (sampling points E4, E5, D1, D2, and D3) (Fig. 5). TDIL and Q Index varied significantly according to a set of environmental variables ($r^2 = 13\%$, $F = 3.2$; $P = 0.01$ and $r^2 = 16\%$, $F = 4.0$; $P = 0.002$, respectively).

Significant differences were found between the ecological assessment based on the two assemblage types ($t = 8.991$; $P = 0.001$) used in this study. Figure 6 shows the Ecological Quality Ratio (EQR) based on phytoplankton and benthic diatoms indices. Discrepancies between the ecological statuses were observed. The Q index indicated worse conditions in Lake Balaton, indicating ecological status varying from bad to good (Fig. 6) while the average diatom index indicated a variation from moderate to good conditions.

Regarding seasonality, the ecological status assessed by both community indices was different in each seasonal period ($t = 3.5$—winter; $t = 4.7$—spring; $t = 6.2$—summer; $t = 4.5$—autumn; $P = 0.000$).

Spatially, the ecological status provided by the Q index and benthic diatom average index did not differ in the western → eastern ($t = 7.8$, $t = 4.8$; $P = 0.000$) or the northern → southern gradients ($t = 5.1$; $t = 7.3$; $P = 0.000$).

Discussion

Phytoplankton and phytobenthos assessments in this study provided different results of ecological status of Lake Balaton.

Integrated classification has been one of the major issues addressed in the context of WFD guidelines. Phytoplankton and attached diatoms are the first groups of organisms to respond to nutrient enrichment with excessive growth and shifts in community composition resulting in several changes in the aquatic ecosystem. Until now, the studies supporting the implementation of the WFD have targeted the development of individual indicators at the level of the BQEs or single parameters within a BQE. Many studies focusing on single BQE have appeared in the scientific literature in recent years (Nõges et al., 2009, Aguiar et al., 2011; Beltrami et al., 2012). Intercalibration exercises have also been done at BQE level, considering national approaches within the same geographical area (Kelly et al., 2009).

One of the crucial points of discussion is the confidence level of the metrics used. The index used for phytoplankton and phytobenthos assessment were previously tested and showed reliable results. In the original proposal, the phytoplankton assemblage Q index was tested not only in Lake Balaton, but also in Lake Fertő on the Austrian-Hungarian border, some alkaline lakes and Hungarian oxbows (Padisák et al., 2006). Recent studies have shown the applicability of this tool in tropical, subtropical, mediterranean, and temperate environments, after the appropriate calibration of the phytoplankton functional weights (F factor) (Crossetti & Bicudo, 2008; Becker et al., 2009; Becker et al., 2010; Nõges et al., 2010; Pasztaleniec & Poniewozik, 2010). The index selected for the diatom approach was successfully tested in 83 Hungarian lakes (Stenger-Kovács et al., 2007) and recently recommended as one of the most appropriate metrics for ecological assessment through benthic diatoms in Lake Balaton (Bolla et al., 2010).

Comparison of ecological evaluations by different communities requires caution. Disparities shown by the assessment of these communities may be assigned to their structural and dynamic dissimilarities. An important difference between planktonic and benthic algae is the spatial organization of the algal community. Phytoplankton are separate entities suspended in the water column with nutrients available from any

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Table 2 Main phytoplankton species, functional groups (FG) and contribution to the total biomass (%; $n = 120$) and main phytobenthos species, codes, and relative abundance (%; $n = 120$) in Lake Balaton

Codes	Phytobenthos species	%
ADMI	*Achnanthidium minutissimum* (Kütz.) Czarn.	16
APED	*Amphora pediculus* (Kütz.) Grun.	7
FHUN	*Fragilaria hungarica* Pant.	5
CPLI	*Cocconeis placentula* Ehr. var .*lineata* (Ehr.) Van Heurck	5
GOMS	*Gomphonema* sp.	5
FCVA	*Fragilaria capucina* Desm. var. *vaucheriae* (Kütz.) Lange-Bert.	4
RABB	*Rhoicosphenia abbreviata* (C.Agardh) Lange-Bert.	3
DMON	*Diatoma moniliformis* Kütz.	3
NCTE	*Navicula cryptotenella* Lange-Bert.	3
GOLI	*Gomphonema olivaceum* (Horn.) Bréb.	3
COCE	*Cyclotella ocellata* Pant.	3
CAFF	*Cymbella excisa* Kütz.	3
PSBR	*Pseudostaurosira brevistriata* (Grun. in Van Heurck) Williams & Round	3
NTPT	*Navicula tripunctata* (O. F. Müller) Bory	2
NPAE	*Nitzschia paleacea* (Grunow) Grunow in van Heurck	2
GPUM	*Gomphonema pumilum* (Grunow) Reichardt & Lange-Bertalot	2
ECPM	*Encyonopsis minuta* Krammer & Reichardt	2
ALIB	*Amphora libyca* Ehr.	2
NDIS	*Nitzschia dissipata* (Kütz.) Grun.	1
FCPE	*Fragilaria capucina* Desm. var. *perminuta* (Grun.) Lange-Bert.	1
FCGR	*Fragilaria capucina* Desmazieres var. *gracilis* (Oestrup) Hustedt	1
NPAL	*Nitzschia palea* (Kütz.) W. Smith	1
DITE	*Diatoma tenuis* Agardh	1
NFON	*Nitzschia fonticola* Grunow in Cleve et Müller	1
FG	Phytoplankton species	%
B	*Cyclotella comta* (Ehr.) Kütz.	16
	Cyclotella ocellata Pant.	2
D	*Fragillaria ulna* (Nitzsch) Lange-Bert.	11
E	*Dinobryon sociale* Ehr.	1
H1	*Aphanizomenon klebahnii* (Elenk.) Pechar et Kalina	3
J	*Pediastrum duplex* Meyen	3
	Pediastrum boryanum (Turpin) Menegh.	3
K	*Synechococcus nidulans* (Pringsheim) Komárek	1
LO	*Ceratium hirundinella* (O. F. Müller) Bergh	2
	Peridinium cinctum (O. F. Müller) Ehr.	1
P	*Aulacoseira granulata* (Ehr.) Simon.	2
S1	*Planktolyngbya limnetica* (Lemm.) Kom.-Legn. & Cronb.	1
$\mathbf{S_N}$	*Cylindrospermopsis raciborskii* (Wolosz.) Seen. & Subba-Raj.	28
X2	*Rhodomonas lacustris* Pascher & Ruttner	1
$\mathbf{X_{PH}}$	*Phacotus lenticularis* (Ehr.) Stein	3
Y	*Cryptomonas erosa* Ehr.	2

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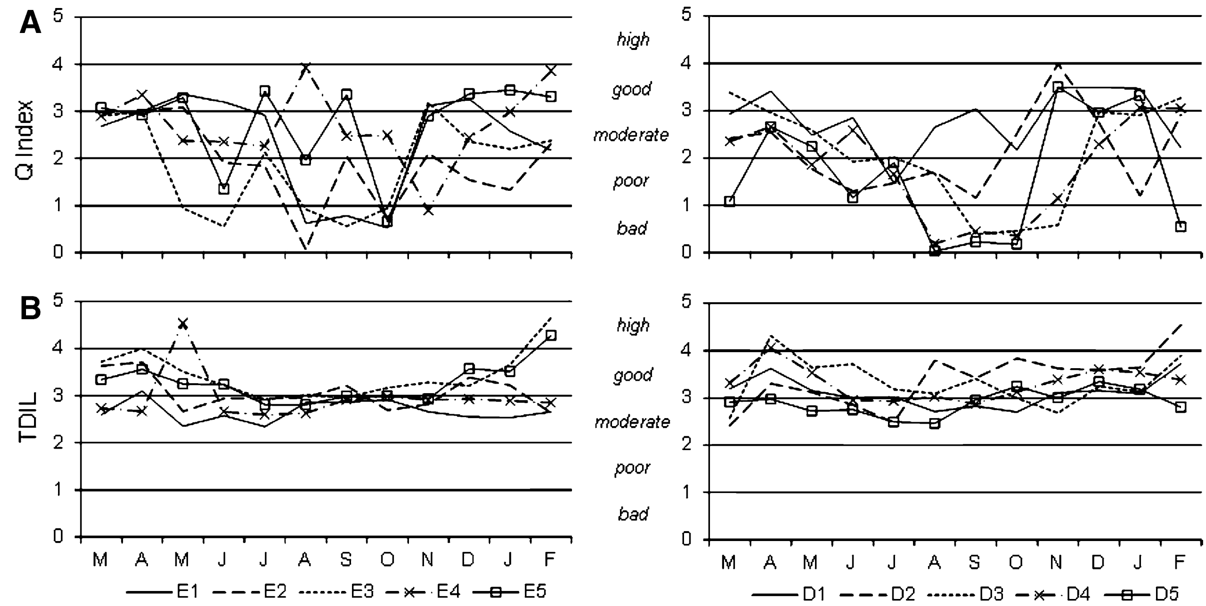

Fig. 5 Ecological status assessment (1–5) based on phytoplankton Q index (**A**) and phytobenthos TDIL (**B**) in southern (D1, D2, D3, D4 and D5) and northern (E1, E2, E3, E4, E5) shorelines of Lake Balaton

direction. Benthic algae, on the other hand, often create mats on the substratum that are many cells thick (Borchardt, 1996), presenting a unique and complex relation with nutrient availability and cycling within the mats. Considering the trophic status based on TP values, the worst classification of Lake Balaton was observed in August. In that period, Q index pointed out the worst ecological status of Lake Balaton (poor) for the majority of the sampling points, in comparison the TDIL values indicated a moderate–good ecological status. In addition, after integrating the biological information through the CCA, the output showed that the responses of phytoplankton functional groups and the benthic diatoms species to the environmental variability were dissimilar. The majority of phytoplankton functional groups were associated with the higher concentration of nutrients, while it was possible to verify the strongest association of phytobenthos species to the lower concentrations of nutrients, especially in the eastern basin of Lake Balaton. Benthic algae may respond weakly to water column nutrients (Hansson, 1988). The lack of a strong direct response from periphyton to nutrient increases in water column may be a function of the compactness of the periphyton community and steep resource gradients within the community (Lowe, 1996).

Beside nutrients, both shallow lake phytoplankton and phytobenthos, especially in shoreline regions, are intermittently influenced by several factors related to the kinetic behavior of the medium. Such factors include wave action, turbulence (Tóth et al., 2011) and the consequent changes in light penetration and nutrient availability. As shown by Padisák et al. (1988, 1990; see also Padisák, 1993), phytoplankton of Lake Balaton is influenced by the stirring up effect of the wind which may reintroduce meroplanktonic species (Padisák & Dokulil, 1994) in the plankton and abruptly change the phytoplankton structure and dynamics providing competitive advantage to small-sized, *r*-strategists. A similar environmental scenario may disturb the developed canopy of attached diatom communities by detaching the loosely adherent species and simultaneously providing an opportunity to good colonizers, as described by Passy (2007) for stream diatoms. However, colonization needs time and therefore might be delayed. These differences are intrinsic to the considered communities and differences in specific response times to equal environmental forces are expected to asynchronize dynamic responses. Net growth rate for phytoplankton is considered as approximately 3 days (Padisák, 1994) while for attached diatoms the rate is about 6 days (Ács

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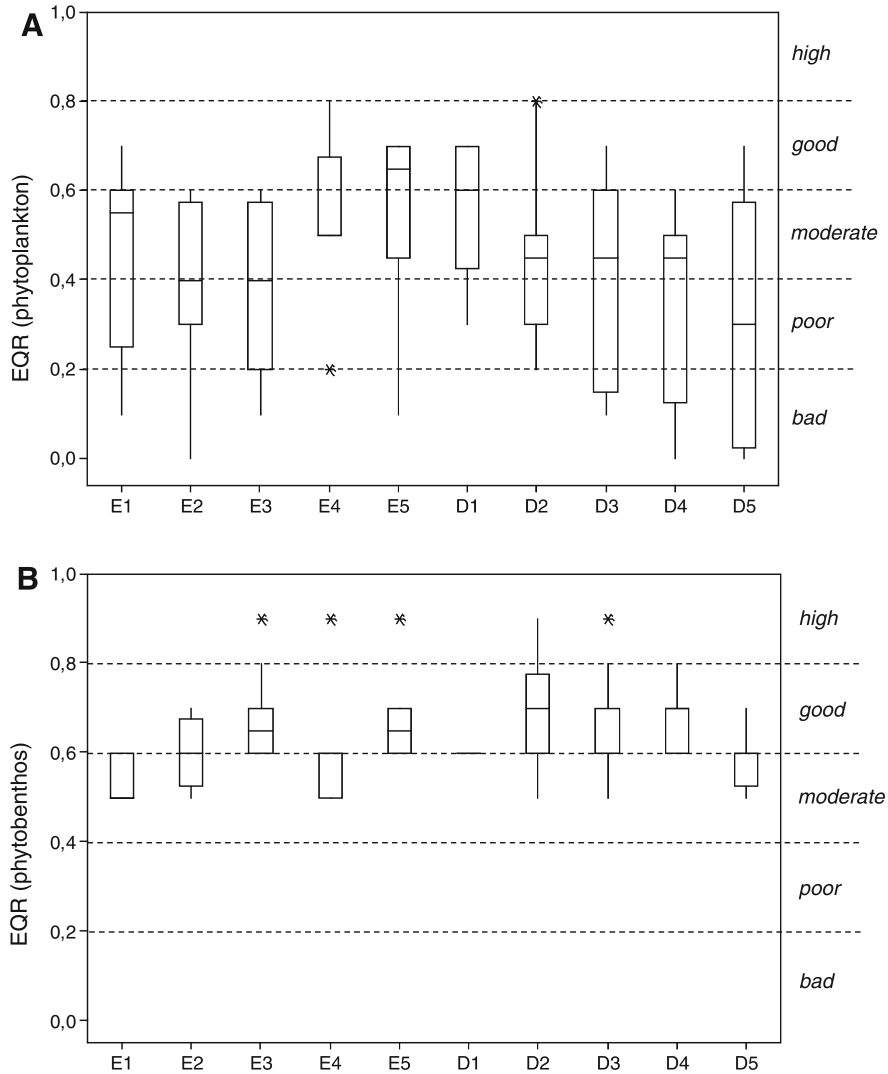

Fig. 6 Boxplot (variation of the data, median, interquartile ranges, outliers, $n = 12$) of Ecological Quality Ratio (EQR, 0–1) based on Q index (**A**) and TDIL (**B**) in South (D) and North (E) basins of Lake Balaton

& Kiss, 1993). Characteristic lags were recently explored in response of diatoms, non-diatom algae, and macrophytes in a data-set from 24 Polish rivers (Schneider et al., 2012).

The WFD "one out–all out" recommendation states that the BQE showing the worst classification determines the ecological status. According to some experts (Moss, 2007), this principle tends to downgrade sites unjustifiably depending on the metrics included in the assessment. Johnson et al. (2006) showed that errors within the individual quality elements and metrics tended to show considerable variation studying invertebrates, diatoms, fishes and macrophytes in 162 European streams. Recently Rask et al. (2011) discussed the discrepancy between the Finish national classification system, which provided the more realistic responses, and the "one out, all out" approach. Following this WFD recommendation, the dissimilarities found in Lake Balaton in the present study through phytoplankton and phytobenthos analysis would, especially in summer, downgrade the ecological status of this ecosystem based on the poor ecological status determined by Q index. Some ideas were already discussed to avoid wrong determinations in the ecological statuses of aquatic bodies, such as the calculation of multimetric indices aggregating several BQEs (Van de Bund & Solimini, 2006) or even the development of alternative approaches for dealing

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with the heterogeneity of the biological information and uncertainty of the "one out all out" principle (Gottardo et al., 2011).

Although some authors indicate the best period of sampling for ecological assessment purposes (e.g., Padisák et al., 2006) for some biological indicators, the results shown in the present study point out that there is no guarantee that the classifications given by different biological communities, as recommended by WFD, will match. This might seriously influence the agreement of different BQEs and the overall EQR of European aquatic ecosystems. The "one out, all out" WFD recommendation disregards at least two basic ecological concepts: one is simply the scale-dependence and the other is the different habitat requirements by different BQEs.

In summary, the ecological status assessments of Lake Balaton based on phytoplankton and phytobenthos were different, ranging from bad to good and from moderate to high, respectively. No reliable concordance of ecological status assessment based on phytoplankton and attached diatoms were demonstrated in a year in the littoral region of Lake Balaton. The Q Index provided more realistic ecological status of Lake Balaton, comparing with trophic status based on TP values, especially in the summer. Phytoplankton variability was higher and response tends to be faster and more immediate for a specific site in the case of sensitive periods (for example, the high tourist season), while benthic communities preserve the history from several weeks prior to investigations. Phytobenthos are attached and less susceptible to short-scale temporal variations. Since response-time differences were traced regarding phytoplankton-attached diatom indication, differences should also be expected in the context of these to any other BQEs since their characteristic response-time differences are even longer.

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