Morphological Plasticity of Dominant Species in Response to Nutrients Dynamics in Bidighinzu Reservoir of Sardinia, Italy

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Abstract

The aim of this study was to describe the effect of environmental factors on the variation in population structure of phytoplankton in a hypertrophic reservoir and to analyse the size variability of dominant species, and finally to try to find a linkage between environmental factors and phytoplankton size structure.

Species composition and dynamics of phytoplankton were typical of highly trophic conditions and were frequently characterized by the dominance of cyanobacteria, *Microcystis, Anabaena, Aphanizomenon*. Some environmental parameters play an important role in determining the phytoplankton community succession and their size structure, favouring or limiting the growth of the different groups of the phytoplankton. The results indicate that overall nutrient concentrations in Bidighinzu Lake were not limiting the phytoplankton growth in general. On the contrary, concentrations of nutrient were always above the required levels for phytoplankton growth. The results show that phytoplankton community succession and their seasonal variability of size structure (consequently cell surface area, cell volume and surface area to volume ratio) were mostly influenced by the temperature and nutrients. However, these affecting factors (especially nutrients) on seasonal variability of size structure of common species in Lake Bidighinzu showed difference among the species.

Keywords: Phytoplankton, Surface-volume Ratio, C-R-S strategies, Environmental factor.

Introduction

Most aquatic ecosystems have suffered from eutrophication phenomenon during the last decades and (Köhler Hoeg, 2000). Differences in phytoplankton biomass and species composition have been found between reservoirs of different trophic status (Negro et al., 2000; Naselli-Flores and Barone, 2003). Freshwater phytoplankton populations are seasonally variable (Hutcinson, 1967) and are regulated by both chemical and physical environmental factors. Growth of each planktonic species is supported within specific ranges of physical and chemical parameters. Many nutrients may potentially be limited factors for algal growth and tend to accumulate in aquatic systems (Fisher et al., 1999). Nutrients affect not only phytoplankton abundance and growth but also morphological structure.

Bidighinzu Reservoir (Figure 1) located in the North-Eastern Sardinia (40°00' N, 9°00' E), Italy, was built in 1958 for supplying drinking water. The volume of the lake is $11 \times 10^6 \text{ m}^3$ and its area is $1.5 \times 10^6 \text{ m}^3$ 10^6 m^2 , together with the mean depth of 7.3 m and the maximum depth of 30 m. Problems of potabilization have arisen since early years of the reservoir's use, particularly in summer-autumn, because of hypolimnic deoxygenation and the excessive presence of algae in the epilimnion. In 1978, its hypertrophic status was determined and these data were recently confirmed (Sechi and Cossu, 1979; Luglié and Sechi,

1993; Luglié et al., 2001).

The progressive decrease in water quality has been accompanied by modifications in both phytoplankton shape and size (Morabito et al., 2007; Naselli-Flores and Barone, 2007). Morphological structure of microalgal cells (linear dimension, surface area and biovolume) shows variability throughout the year and from different sites. Morphometric diversity may be related to physiological adaptation to varying light and nutrient (Margalef, 1958). A theoretical availability framework for which morphological - functional (m/f) groups should dominate under varying conditions has been developed by Reynolds (1988, 1997) over the past decade (Huszar and Caraco, 1998).

The aim of this study was to describe the effect of environmental factors on the variation in population structure of phytoplankton in a hypertrophic lake and to analyse the morphological variability of phytoplankton species in response to the seasonal changes of water quality in natural environment.

Materials and Methods

The samples were collected monthly from December 1996 to November 1997 at one site located about 500 m from the dam by a Niskin bottle (surface, 1 m, 2.5 m, 5 m, 7.5 m, 10 m, 15 m, 20 m). Dissolved nutrient concentrations were analyzed after filtering

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Figure 1. Location of Bidighinzu Lake and sampling station.

water through Whatman GF/C filter paper. Ammonium nitrogen (Fresenius et al., 1988), nitrate nitrogen, nitrite nitrogen, reactive phosphate and reactive silicates were analyzed according to Strickland and Parsons (1968). Alkalinity was measured by the method of Golterman et al. (1978). Water temperature, pH, conductivity, dissolved oxygen and transparency were measured in situ by an IDROMAR multiparameter probe and a Secchi disc, respectively. The euphotic depth was estimated by the following equation: $Z_{eu} = 2.7 \text{ x}$ Secchi disc depth (Reynolds, 1984). Total chlorophyll a was determined according to Golterman et al. (1978). For phytoplankton analyses, samples were preserved in Lugol's solution. Sub-samples were prepared for sedimentation in counting chambers of 5 ml size and allowed settling for 2 hours (Utermöhl, 1958). Standard keys and manuals were used for taxonomic determinations (Dillard, 1989a; 1989b; Huber-Pestalozzi, 1961; 1969; 1972; 1983; Hustedt, 1985; Kramer and Lange-Bertalot, 1986, 1988, 1991a, 1991b; Komarek and Anagnostidis, 1999). Biomass of phytoplankton species was estimated from biovolume, by geometrical approximations according to Hillebrand et al. (1999). Moreover, during the study period surface area, volume and surface area/volume ratios were calculated for common species. For colonial organisms, cell shape and measures were taken into consideration. Morphological structure similarity was evaluated by using theoretical framework according to Reynolds (1997). The effect of environmental factors on algal growth and size structure was evaluated separately for every species.

Results were analyzed with Student t test using the SPSS 11 program. Spearman rank correlations were used to evaluate relations between species and environmental variables. Bray-Curtis similarity index was used for species groups.

Results and Discussion

Physical and Chemical Characters

Seasonal and vertical changes of the environmental factors in the reservoir water are given in Figure 2. The lowest temperatures (9°C) were measured in August 1997, while the highest (26°C) were measured in January 1997. A vertical thermal uniformity was observed up to May, when stratification started, until September. Thermocline formed between 5-7.5 m. From September to November, temperature decreased and it was already homogenous in the water column in December.

Transparency was relatively low, Secchi depth varied between 50 and 100 cm. The annual mean of the euphotic depth of the lake was 2.2 m. Euphotic depth showed negative correlation with total chlorophyll a and algal growth (P<0.05). The euphotic depth was strongly influenced by the algal biomass. The total increase of phytoplankton density and biomass in summer and early autumn period causes unfavourable light conditions (Figure 3).

Dissolved oxygen in surface water was generally oversaturated due to algal productivity. Significant differences were recorded in vertical distribution of dissolved oxygen, especially during thermal stratification periods. Minimum value of dissolved oxygen (0 %) was obtained at surface in May and the maximum concentration (149 %) was recorded at the depth of 10 and 15 m (near the bottom) in September.

The vertical distribution of pH values changed considerably during thermal stratification period, with higher values in the water above 10 m. The minimum value (7.18) was recorded in September 1997 at 15 m. Alkalinity and conductivity had homogenous vertical



Figure 2. Seasonal variations of environmental factors along the water column.

dynamics. The lowest and the highest values were respectively 1.78 meq L^{-1} in November at 5 m and 3.90 meq L^{-1} in September at 15 m depth, 529 μ S cm⁻¹ in October always at surface and 834 μ S cm⁻¹ in July at surface water.

In general, dissolved nutrient concentrations were extremely high. They were recorded as average 155 ± 54 mg P m⁻³ (surface - 5 m) and 374 ± 278 mg P m⁻³ (7.5-15 m) for reactive phosphate; 230 ± 53

(surface-5m) and 449±281 mg P m⁻³ (7.5-15 m) for total phosphorus; 778±589 mg N m⁻³ (surface - 5 m) and 1273±354 mg N m⁻³ (7.5-15 m) for the inorganic nitrogen (addition of nitrate nitrogen, nitrite nitrogen and ammonium nitrogen). Silicate concentrations were continuously high, with an annual average of 4.5 ± 3.1 mg Si L⁻¹ (surface - 5 m) and 6.7±1.5 mg Si L⁻¹ (7.5-15 m). Vertical distribution of silicate was homogenous from December 1996 to May 1997,



Figure 3. Seasonal changes of euphotic depth and total chlorophyll *a* in the lake.

October and November 1997. During the thermal stratification period, only from May to August 1997 the contents of the first 5 m were low ($\leq 2 \text{ mg Si } L^{-1}$). However, these values did not have a limiting effect on the growth of diatoms. Relatively low nutrient values were recorded during the summer and autumn between surface and 5 m due to excessive phytoplankton development. Nutrient concentrations reached the highest values at the bottom (between 7.5 and 15 meters) with releases from the sediment in summer and autumn. Differences of nutrient concentrations were recorded between surface and bottom waters (ANOVA, P<0.05). Dynamics of dissolved nutrients have been reported previously by Luglié et al. (2001) and Aktan et al. (2005) as more detailed. The ratio of total nitrogen to total phosphorus by mass ratio (TN:TP) was lower than the Redfield ratio throughout the year, confirming its high eutrophic state and underlining the importance of nitrogen on the development and composition of phytoplankton. Lower ratios were usually from May to October (Figure 4). The seasonal changes of TN: TP ratio show that nitrogen was the potentially limiting nutrient due to intensive algal growth (Figure 5), especially late spring, summer and autumn period. Significant correlation (P<0.01) was recorded between TN: TP ratio and phytoplankton abundance. High cyanobacteria concentrations were associated with low TN: TP ratios and very low cyanobacterial abundance corresponded to high TN: TP ratios.

Phytoplankton Abundance and Size Variability

During the study period, more than 104 phytoplanktonic taxa of different classis were identified (unpublished data). The contribution of classis to the total biomass through the study is shown in Figure 5. Bacillariophyceae, Cyanophyceae and Chlorophyceae were the most important groups. Species composition of phytoplankton was typical of highly trophic conditions and was frequently characterized by the dominance of cyanobacteria, especially spring, summer and autumn period. *Cyclotella* spp, *Stephanodiscus hantzschii*, *Aulacoseira granulata* from diatoms; *Aphanizomenon*

flos-aquae, Aphanocapsa sp., Anabaena planctonica, Merismopedia tenuissima, Microcystis aeruginosa, Pseudanabaena mucicola from cyanobacteria; Coelastrum pseudomicroporum, Oocystis spp., Sphaerocystis planctonica and from chlorophytes were the most abundant species. On the other hand, Cyclotella spp, S. hantzschii, A. granulata, M. aeruginosa, Closterium aciculare, Coelastrum pseudomicroporum, Oocystis spp., Scenedesmus quadricauda, S. planctonicus and Crptomonas sp. were the most common species throughout the year.

Significant correlations were found between the phytoplankton abundance and biovolume. They were similar to that of chlorophyll *a*, showing their maxima in August. High values of total phytoplankton biovolume and chlorophyll *a* were produced mainly by large cells (especially *Ceratium hirundinella*). The seasonal variation in total phytoplankton abundance in term of biovolume is given in Figure 5. The highest phytoplankton abundance (27.7 mg L⁻¹) was in the surface water in August, whereas the lowest value (0.2 mg L⁻¹) was in the surface and 10 m in January.

Phytoplankton developments depend on several factors besides physical parameters like temperature and light. Nutrients are important factors among these variables. During the study period, overall nutrient concentrations in Bidighinzu Reservoir never limited the phytoplankton growth. On the contrary, concentrations of nutrient were always above the required levels (Figure 2) for phytoplankton growth (according to Reynolds, 1997).

Seasonal variability of size structure (Figure 6) (consequently cell surface area, cell volume and surface area to volume ratio) and affecting factors on seasonal variability of size structure of common species in Bidighinzu Reservoir showed difference according to species.

For diatom species, SA/V ratios were not variable during the year (Figure 7). The maximum ratios were observed during summer month because of their relatively low volume. In this period, abundance of diatoms was high and cell size was low. Diatoms were dominant group during winter; however, they reached maximum biomass values in the period of autumn. Silicate concentrations, which



Figure 4. The results of TN, TP and TN:TP ratios (as mass) in the lake (TN: Total nitrogen, TP: Total phosphorus): monthly means of depths and their standard deviations.



Figure 5. Seasonal variations of total phytoplankton biomass and contribution of classis to the total biomass.



Figure 6. Seasonal variations size structure (surface area, volume and surface area/volume ratios) of species.



Figure 7. Box plot showing the mean SA/V ratios.

are a crucial nutrients for diatom growth, were negatively correlated (P<0.01) with seasonal distribution total diatom biomass. On the other hand, silica positively affected the SA/V ratios of *Stephanodiscus* and *Aulacoseira*. TP was another important nutrient in terms of size variability among diatoms species. Seasonal distribution of TP concentrations showed positive correlation with SA/V ratios of diatom species, but correlation was not significant for *Aulacoseira* (P<0.01).

Marked similarity was not recorded in seasonal trends of the SA/V ratios of common chlorophytes species and significant correlation (P<0.05) was not observed with environmental factors. However, the SA/V ratios of *Sphaerocystis planctonica* and *Coelastrum pseudomicroporum* decreased in winter and early spring period with higher nitrate concentrations (especially month of March with mean of nitrate concentration as 917 ± 126 mg N m⁻³), despite cell sizes of these species were high, and their densities were low.

Cyanophytes were not common throughout the year due to their high abundance. One common species, Microcystis aeruginosa, had relatively lower ratios of SA/V between May-August. A clear decrease (minimum SA/V ratios) was observed in August when cell size was higher. Increase of cell size caused high values in volume in respect of surface area. Temperature negatively affected SA/V ratios of Microcystis and significant correlation was recorded (P<0.01). On the other hand, the most effective nutrient was dissolved inorganic nutrients (DIN). Significant positive correlation (P<0.01) was observed with Microcystis SA/V ratios. Moreover, TN: TP ratios showed significant positive correlation too and both TN: TP (<7) and SA/V ratios were minimum from May to September. According to N:P ratios, we could say that nitrogen was the potentially limiting nutrient due to intensive algal growth in late spring, summer and autumn period when Microcystis cells had high SA/V ratios (small cell size).

The most common species from Cryptophytes was *Cryptomonas* sp. Minimum SA/V ratios (due to increasing size and biovolume) were recorded during spring and their maximum values were recorded in August. Apart from the reactive and total phosphorus,

SA/V ratios did not show significant correlation with environmental parameters (P < 0.05).

Mean SA/V ratios of dominant species with standard deviations were given in Figure 7. *Stephanodiscus hantzschii* had minimum SA/V ratio (0.37 \pm 0.06) meaning large cell size. *Microcystis aeruginosa* had maximum SA/V ratio (1.94 \pm 0.43) meaning small cell size. It can be concluded that geometric shape and cell size might affect these results.

Species formed two groups according to the SA/V ratios (Figure 8). Reynolds (1997) divides phytoplankton into three major strategies as C- (fast growing small phytoplankton), S- (slow growing large unicells or colonies), and R- (elongated unicells, colonies or filaments) strategist (Huszar and Caraco, 1998). According to morphological-approach developed by Reynolds, small fast growing algae such as Cryptomonas sp. and centric diatoms formed Cspecies. Closterium aciculare, Oocystis spp and Aulocoseira granulata formed R-species. Colonial species with high surface area/volume, such as *Sphaerocystis* planctonica, Scenedesmus quadricauda, Coelastrum pseudomicroporum and Microcystis aeruginosa formed S-species. Moreover, marked deviations were recorded throughout the year in colonial species like Microcystis aeruginosa, Coelastrum pseudomicroporum and Sphaerocystis planctonica. On the contrary, deviation of C-species SA/V ratios did not vary (Figure 8) due to their small size. They could grow quickly but would also be removed rapidly by grazing.

According to Figure 9 and 10, significant positive correlation ($R^2=0.84$) was found between total cell surface area and volume. However, size increase had more effect on biovolume and marked relationship between SA/V and cell volume was negative ($R^2=0.67$). A result of this study shows that size variability was mostly related to volume. On the other hand, except for diatoms we observed that seasonal variability of size structure in some species (consequently cell surface area, cell volume and surface area to volume ratio) was very clear. Affecting factors on seasonal variability of size structure of common species in Lake Bidighinzu showed difference according to species. Correlations



Figure 8. Bray-Curtis similarity index as SA/V ratios of species and morphometric properties of C-, S- and R- species. Taxonomic groups are indicated by \blacksquare =Diatoms (Cyc spp: *Cylotella* spp., Ste han: *Stephanodiscus hanzschii*, Aul gra: *Aulacoseria granulata*); \square =Cryptomonads (Cry spp: *Crytomonas* spp.); O=Chlorophytes (Ooc spp: *Oocystis* spp., Clo aci: *Closterium aciculare*, Coe pse: *Coelastrum pseudomicroporum*, Sce qua: Scenedesmus quadricauda, Sph pla: *Sphaerocystis planktonica*); \blacksquare = Cyanobacteria (Mic aer: *Microcystis aeruginosa*).



Figure 9. The relationships showing between surface area and volume of common phytoplankton in Lake Bidighinzu.



Figure 10. The relationships showing between surface area/volume and volume of common phytoplankton in Lake Bidighinzu.

between every species with environmental factors were not clear. Therefore, we can say that response to environmental factors of every one species was differing toward their own needs.

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