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Phytoplankton and water quality in a Mediterranean drinking-water reservoir (Marathonas Reservoir, Greece)

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Abstract Phytoplankton and water quality of Marathonas drinking-water Reservoir were examined for the first time. During the study period (July–September 2007), phytoplankton composition was indicative of eutrophic conditions although phytoplankton biovolume was low (max. 2.7 mm^3 l^{-1}). Phytoplankton was dominated by cyanobacteria and diatoms, whereas desmids and dinoflagellates contributed with lower biovolume values. Changing flushing rate in the reservoir (up to 0.7% of reservoir's water volume per day) driven by water withdrawal and occurring in pulses for a period of 15–25 days was associated with phytoplankton dynamics. Under flushing pulses: (1) biovolume was low and (2) both 'good' quality species and the tolerant to flushing 'nuisance' cyanobacterium *Microcystis aeruginosa* dominated. According to the Water Framework Directive, the metrics of phytoplankton biovolume and cyanobacterial percentage (%) contribution indicated a moderate ecological water quality. In addition, the total biovolume of cyanobacteria as well as the dominance of the known toxin-producing *M. aeruginosa* in the reservoir's phytoplankton indicated a potential hazard for human health according to the World Health Organization.

Keywords Drinking-water reservoir · Flushing rate · Cyanobacteria · Ecological water quality · Management · WFD

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Introduction

During the last decades, human activities have deteriorated water quality in a significant number of lakes and reservoirs worldwide. This, along with the growing necessity to meet societal demands for both water quantity and water quality (Kennedy 1999), suggests that monitoring the hydrology and water quality of freshwaters is critical for their proper management in the long term. Reservoirs differ considerably from natural lakes in respect of basic physical, chemical and biological processes. One of the most striking differences is their hydrological regime (Straškraba and

Tundisi 1999). Due to an operational management of their water volume, reservoirs experience shorter retention times compared to those of lakes, intense water level fluctuations, as well as periodic pulses of mixing depending on their use (Tundisi et al. 1999). These features have profound effects among others on the phytoplankton community of a reservoir (Naselli-Flores 2000; Naselli-Flores and Barone 2005).

Phytoplankton can be used as a good indicator of water quality changes, given its sensitivity and dynamic responses to changes in the surrounding environment (Padisák et al. 2006). In Europe, after the establishment of the Water Framework Directive (WFD) in 2000 (European Parliament, Council 2000), phytoplankton is one of the four biological elements suggested for assessing the ecological status/potential of surface waters. In addition, the World Health Organization (WHO) has established an Alert Levels Framework for drinking-water supplies based on the cyanobacteria presence in order to determine water quality and human health risk (Bartram et al. 1999).

In Greece, several studies have reported prolonged cyanobacterial dominance (e.g. Moustaka-Gouni et al. 2007) along with the presence of toxin-producing cyanobacterial species (Vardaka et al. 2005) and considerable concentrations of cyanotoxins (Cook et al. 2004; Gkelis et al. 2005a) in several lakes and reservoirs. However, these water systems lack any management regarding cyanobacteria and cyanotoxins (Cook et al. 2005). Knowledge on the phytoplankton of Greek drinking-water reservoirs is scarce (Moustaka-Gouni and Nikolaidis 1992; Chrisostomou et al. 2009). Publications on the phytoplankton of the Marathonas Reservoir are completely lacking despite the fact that it was created in the 1930s in order to be used as a drinking-water supply for Athens, the capital of Greece. Only one study on the phylogenetic diversity of cyanobacteria of the reservoir has recently been published (Lymperopoulou et al. 2010).

This is the first paper on the phytoplankton and water quality of Marathonas Reservoir. The aim of this paper was (a) to examine the phytoplankton compositional diversity and biovolume under a changing flushing rate and (b) to assess the reservoir's water quality according to both WFD and WHO guidelines.

Methods

Study site

Marathonas Reservoir (38°09′59″ N, 23°53′58″ E; Fig. 1) is situated at the northeastern part of the Attica region, Central Greece, at 173 m above sea level. It was put in operation for supplying drinking water to Attica region (including the city of Athens, the capital of Greece) in 1931. The reservoir has a mean surface area of 2.45 km^2, maximum depth 54 m, mean depth 15 m and a catchment area of 118 km^2 drained by five perennial inflows. The only outflow of the reservoir is the water supply tower (Fig. 1). With an operational volume of 34 million cubic metres, the reservoir was the main source for supplying drinking water to Attica region until 1959 (Athens Water Supply and Sewerage Company (EYDAP SA), unpublished data). Nowadays, it operates as a backup source for the water supply system of the

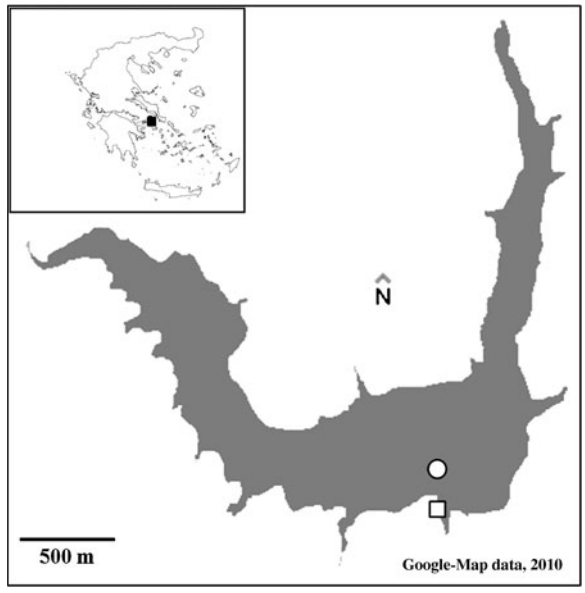

Fig. 1 Map of Marathonas Reservoir. The *circle* indicates the sampling site and the *square* the reservoir's water supply tower. *Insert*, map of Greece (*solid square* indicates the location of Marathonas Reservoir)

greater Attica region and as a primary regulating reservoir.

During the study period (July–September 2007), two major decreases in the reservoir's water volume were recorded. The first one, in July, was more abrupt and resulted in loss of 8% of the reservoir's water volume. It was characterized by periodic flushing pulses (0.1–0.7% of the reservoir's water volume per day) that lasted for 25 days. The second one, in late August–early September, was shorter (15 days) with weaker flushing pulses (0.06–0.6% of the reservoir's water volume per day) and resulted in loss of 3% of the reservoir's water volume. The fluctuations of the reservoir's water volume and the flushing rate per day for the period July–September 2007 are shown in Fig. 2. Data on the reservoir's water volume were provided by the EYDAP SA. Flushing rate (given as % of the reservoir's water volume per day) was calculated from the total reservoir's water volume and its daily changes.

Field work

Sampling covered the warm period of 2007 and was conducted from July to September biweekly. The sampling station (15 m deep at highest water level) was located at the area of the reservoir's outflow (water supply tower) that was strongly affected by changing flushing rate (Fig. 1). Depth discrete water samples were collected with a 2-l Niskin sampler from the surface to the end of the euphotic zone (maximum depth 12.5 m) at 2-m intervals. In addition, integrated samples were taken from the euphotic zone. Sub-samples were preserved in Lugol solution.

Water temperature, pH and dissolved O_2 were measured in situ using a portable multi-parameter analyzer WQC-24 (DKK TOA) and transparency was measured with a Secchi disc. The euphotic zone (z_{eu}) was calculated as 2.5 times the Secchi depth and the mixing zone (z_{mix}) was indentified using temperature profiles. Stratification was considered to occur at the depth where water temperature changed by $>1°C\ m^{-1}$.

Laboratory and data analysis

Fresh and preserved samples were examined under a light inverted microscope (Nikon SE 2000), and species were identified using taxonomic keys. Phytoplankton counts were performed using the sedimentation method of Utermöhl (1958). At

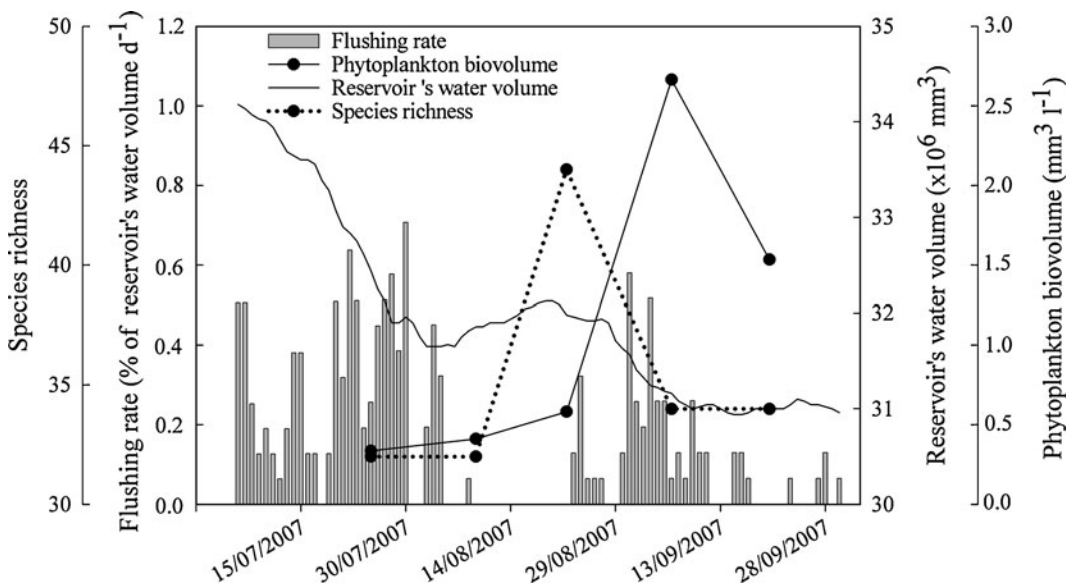

Fig. 2 Phytoplankton biovolume and species richness during the study period (July–September 2007) in Marathonas Reservoir. Biovolume values are means and refer to the euphotic zone. Flushing rate and daily fluctuations of the reservoir's water volume from early July to late September are shown

least 400 phytoplankton individuals were counted in each sample. The dimensions of 30 individuals (cells, filaments or colonies) of each species were measured using tools of a digital microscope camera (Nikon DS-L1). For the colonial cyanobacterium *Microcystis aeruginosa*, the number of cells per colony (y) was determined using the equation: $y = 1.475 \times (\text{colony diameter})^{1.55}$ according to Moustaka (1988). Since colonies were not always spherical, the colony diameter used in the final computation was the diameter of a sphere with an equal colony volume according to Reynolds and Jaworski (1978). Mean cell or filament volume estimates were calculated using appropriate geometric formulae (Hillebrand et al. 1999). Species and taxonomical groups comprising more than 10% (v/v) of the total phytoplankton biovolume were considered to be dominant. Reynolds et al. (2002) functional groups classification was applied to the phytoplankton species supplemented by the review of Padisák et al. (2009). In this paper, 'good quality' species refer to those phytoplankton species which are typical for oligo-mesotrophic environments such as species of the genus *Cyclotella* and desmids (e.g. species of *Cosmarium*). The functional group of these species gets the highest score when assessing ecological water quality (Padisák et al. 2009).

To assess the ecological water quality, the boundaries of Good/Moderate (G/M) ecological potential were used, related to total phytoplankton biovolume and the percentage (%) contribution of cyanobacteria for calcareous Mediterranean reservoirs. These boundaries were proposed by the European Commission in the Water Framework Directive Intercalibration technical report Part 2: Lakes (JRC European Commission 2009). To assess the potential health risks associated with the presence of cyanobacteria in the reservoir, the WHO Alert Levels Framework model (Bartram et al. 1999) for the monitoring and management of cyanobacteria in drinking waters in relation to cyanobacteria biovolume was used.

In order to identify interrelationships among samplings, cluster analysis using the Bray–Curtis similarity index (Clarke and Warwick 1994) was applied to the (biovolume of each species) × (samples) matrix data. Prior to the analysis, data were log (x+1) transformed. For the determination of the species responsible for within group similarities and between groups' dissimilarities (Clarke and Warwick 1994), similarity percentage analysis (SIMPER) was used.

Results

Environmental parameters

The reservoir's environmental parameters examined in this study are given in Table 1. Mean water column temperature varied from 25.1°C (in July) to 22.6°C (in September). Maximum temperature reached 25.7°C (surface layer, in late August), whereas minimum temperature was 22.2°C (end of euphotic zone, in late September). Thermal stratification of the water column was observed during July–August, with an epilimnion of 8 m in depth. Secchi disc transparency was relatively high (varied between 3.5 and 5 m) with a decreasing tendency in September. The euphotic zone ranged from 8.8 to 12.5 m. It was deeper than the mixing zone ($z_{eu}/z_{mix} > 1$) until the end of August whereas in September after the break down of the thermal stratification, the ratio z_{eu}/z_{mix} decreased to <1. Mean, maximum and minimum values of dissolved oxygen and pH did not vary considerably during the study period. The dissolved O_2 ranged between 6.2 and 7.3 mg l^{-1} and pH values ranged between 7.8 and 8.7.

Phytoplankton compositional diversity and biovolume

A total of 62 phytoplankton species belonging to 16 functional groups were indentified (Table 2) in the reservoir from July to September 2007. The main taxonomic groups were: chlorophytes exhibiting the highest number of species (27), cyanobacteria (nine), diatoms (eight) and desmids (seven) followed by the less diverse dinophytes (five), cryptophytes (two), chrysophytes (two), xanthophytes (one) and prymnesiophytes (one). The more diverse functional groups were J and Lo with 15 and eight species, respectively. Species richness was lower (32 species) in late July–early August, a period of increased periodic flushing

Table 1 Physical and chemical parameters of Marathonas Reservoir during the study period of July–September 2007

		Transparency (m)	Temperature (°C)*	Dissolved O_2 (mg l^{-1})*	pH*	Z_{mix}(m)	Z_{eu}(m)	Z_{mix}/Z_{eu}
25/07/07	A	4	25.1	7.0	8.1	8	10	0.8
	B		25.5	7.3	8.7			
	C		22.7	6.8	7.8			
09/08/07	A	5	24.8	6.6	8.2	8	12.5	0.6
	B		25.4	6.7	8.4			
	C		23.2	6.2	7.8			
22/08/07	A	4.5	25.0	6.9	8.2	8	11.25	0.7
	B		25.7	7.0	8.4			
	C		22.9	6.4	8.0			
06/09/07	A	4.5	24.3	7.0	8.2	14	11.25	1.2
	B		24.8	7.1	8.3			
	C		23.7	6.8	8.1			
20/09/07	A	3.5	22.6	6.9	8.3	14	8.75	1.6
	B		22.7	7.0	8.5			
	C		22.2	6.8	8.2			

*A mean, *B* maximum, *C* minimum values

(Fig. 2). It exhibited a maximum of 44 species in late August after a 14-day period of no flushing and shifted back to similar species number (34 species) in September during the second period of increased flushing.

Phytoplankton biovolume was low (0.45 ± 0.1 mm^3 l^{-1}) during the first period of increased flushing (Fig. 2). A peak value (2.7 mm^3 l^{-1}) was observed in early September following a 20-day period of very low flushing. After the second period of increased flushing, the biovolume decreased to almost half (1.5 mm^3 l^{-1}) of its peak value. The main contributors to phytoplankton (comprising>80% of total biovolume) during the study period were cyanobacteria and diatoms and to a lesser extent desmids and dinophytes (Fig. 3). The more diverse in species number chlorophytes contributed less than 5% to the total phytoplankton biovolume. Cyanobacteria were observed to dominate from early August until the end of September, forming a peak in the first week of September when water temperature was still high (25°C). The increase in cyanobacterial biovolume (Figs. 2 and 3) started only when no water withdrawal from the reservoir occurred for 14 days. Accumulation of cyanobacteria biovolume continued until a flushing rate greater than 0.5% of the reservoir's water volume per day occurred.

M. aeruginosa was the dominant species constituting 50–88% of the cyanobacterial biovolume. This species was present in low numbers (<300 cells per millilitre) in July, started increasing in August during the 14-day period with no flushing and dominated exclusively the peak of phytoplankton biovolume early in September at the breakdown of the thermal stratification. During the peak, 63% of *Microcystis* colonies were larger than 200 μm (range 117–466 μm). *Microcystis* decreased when mixing prevailed after the second flushing period. At that time, most of its colonies were smaller than 100 μm in maximum dimension. During this period the zooplankton community was dominated by rotifers, small-sized cladocera and copepods (Katsiapi et al., unpublished data). The other cyanobacteria species recorded in the reservoir did not form substantial amounts of biovolume and did not dominate during the study period. The filamentous *Anabaena bergii* and *Pseudoanabaena* sp. although present through the whole study period, remained in low numbers (1 filament per millilitre). The chroococcal *Chroococcus dispersus* with a maximum density of 730 cells per millilitre was not dominant, while *Limnothrix redekei* and *Merismopedia* sp. were rare in the water samples.

Diatoms, the second most important group in terms of biovolume, were the main dominants

Table 2 List of phytoplankton species and functional groups (according to Reynolds et al. 2002 and Pádisak et al. 2009) identified in water samples collected during the study period (July–September 2007) from Marathonas Reservoir

Taxa	Functional group
Cyanobacteria	
Anabaena bergii OSTENF.	H1
Aphanizomenon sp.	H1
Aphanocapsa delicatissima W. et G. S. WEST	L_O
Chroococcus dispersus (KEISSL.) LEMM.	L_O
Chroococcus limneticus LEMM.	L_O
Limnothrix redekei (VAN GOOR) MEFFERT	S1
Merismopedia sp.	L_O
Microcystis aeruginosa (KÜTZ.) KÜTZ.	M
Pseudoanabaena sp.	S1
Chlorophytes	
Chlamydomonas sp.	X2
Choricystis coccoides (RODHE and SKUJA) FOTT	X1
Coelastrum astroeideum DE-NOT.	J
Dictyosphaerium pulchellum WOOD	F
Didymocystis bicellularis (CHOD.) KOM.	X1
Elakatothrix genevensis (REVERD.) HIND.	F
Haematococcus sp.	X2
Kirchneriella contorta (SCHMIDLE) BOHL.	F
Lagerheimia quadriseta (LEMM.) G. M. SMITH	J
Lagerheimia sp.	J
Monoraphidium komarkovae NYG.	X1
Monoraphidium minutum (NÄG.) KOM.-LEGN.	X1
Monoraphidium sp.	X1
Oocystis sp.	F
Pediastrum duplex MEYEN	J
Pediastrum simplex var. *echinulatum* WITTR	J
Pediastrum simplex var. *simplex* MEYEN	J
Scenedesmus acuminatus (LAGERH.) CHOD.	J
Scenedesmus acutus MEYEN	J
Scenedesmus ecornis (EHRENB.) CHOD.	J
Scenedesmus linearis KOM.	J
Scenedesmus sp.	J
Sphaerocystis schroeteri CHOD.	F
Tetraedron caudatum (CORDA) HANSG.	J
Tetraedron minimum (A. BR.) HANSG. sensu SKUJA	J
Tetrastrum triangulare (CHOD.) KOM.	J
Treubaria triappendiculata BERN.	F
Diatoms	
Asterionella formosa HASSALL	C
Aulacoseira granulata (EHRENB.) RALFS	P
Cyclotella sp.	B
Fragilaria crotonensis KITT.	P
Nitzschia sp.	D
Rhizosolenia eriensis H. L. SMITH	A
Rhizosolenia longiseta ZACHARIAS	A
Synedra acus KG.	D

along with desmids and dinophytes from late July to early August, the period of the highest flushing rate and thermal stratification. Nevertheless, their biovolume was low (0.2 mg l^{-1}). From late August to early September, the period of the exclusive dominance of cyanobacteria, diatoms exhibited even lower biovolume values. When flushing rate increased again and turbulent mixing prevailed,

Table 2 (continued)

Taxa	Functional group
Desmids	
Closterium acutum BRÉB.	P
Cosmarium blyttii WILLE	N_A
Cosmarium depressum var. *planctonicum* REVERD.	N_A
Cosmarium phaseolus BRÉB.	N_A
Cosmarium reniforme (RALFS) ARCHER	N_A
Mougeotia sp.	N_A
Staurastrum sp.	P
Cryptophytes	
Cryptomonas sp.	Y
Rhodomonas minuta SKUJA	X2
Dinophytes	
Ceratium furcoides (LEVANDER) LANGHANS	L_O
Glenodinium sp.	Y
Peridinium gatunense NYG.	L_O
Peridinium goslaviense WOLOSZ.	L_O
Peridinium sp.	L_O
Xanthophytes	
Goniochloris smithii (BOURR.) FOTT	J
Chrysophytes	
Dinobryon divergens IMHOF	E
Ochromonas ludibunda PASCHER	X2
Prymnesiophytes	
Chrysochromulina parva LACKEY	X2

they dominated in the reservoir's phytoplankton comprising ca. 50% of the total phytoplankton biovolume. During the whole study period, their biovolume consisted almost exclusively of small-sized cells of *Cyclotella* sp. (6×10^3 cells per millilitre). Desmids co-dominated (comprising less than 15% of total biovolume) along with the other groups until the end of August. The nanoplanktic *Cosmarium phaseolus* was the main contributor to their biovolume. Dinophytes were found to

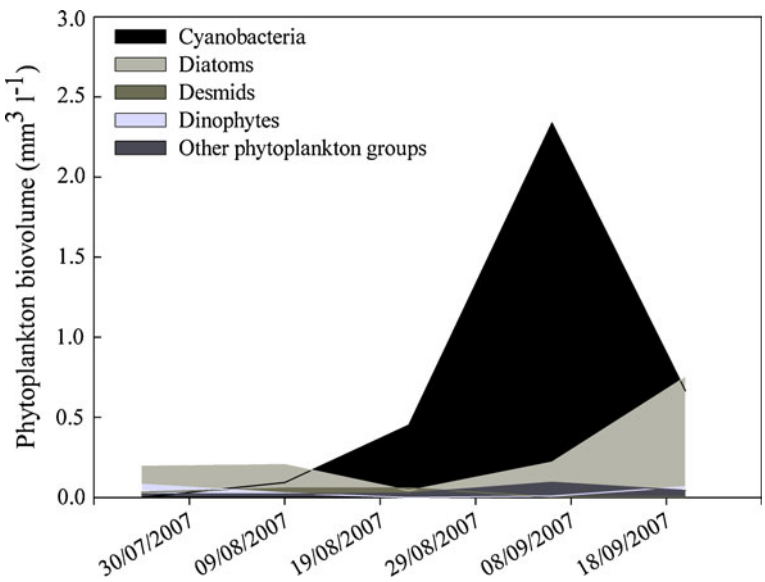

Fig. 3 Temporal changes in biovolume of the main phytoplankton taxonomic groups during the study period (July–September 2007) in Marathonas Reservoir. Values are means and refer to the euphotic zone

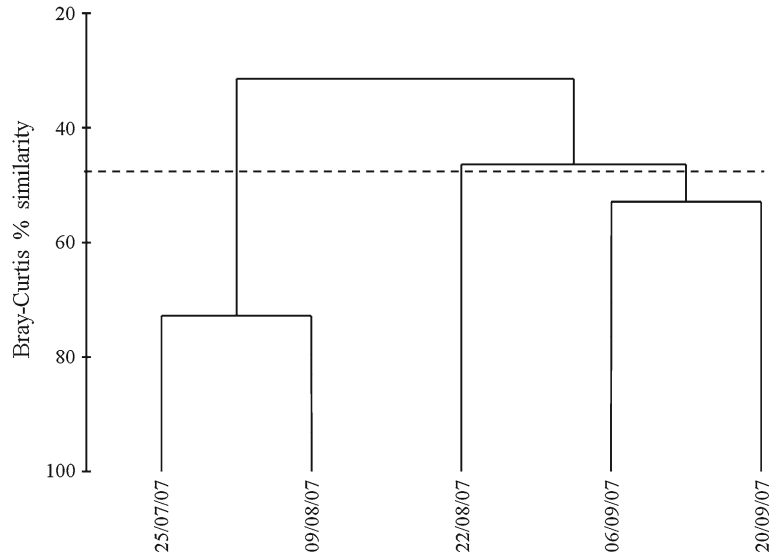

Fig. 4 Dendrogram for clustering of the water samplings during the study period (July–September 2010) in Marathonas Reservoir based on Bray–Curtis similarities. Cluster analysis was applied to the (biovolume of each species) × (samples) matrix data

Table 3 Results of SIMPER analysis of the water samples from Marathonas Reservoir during the study period (July–September 2010)

	Contribution (%)	Cumulative (%)
Group I		
Average similarity 72.77		
Cyclotella sp.	70.01	70.01
Cosmarium phaseolus	12.96	82.97
Ceratium furcoides	10.92	93.88
Group II		
Less than 2 samples in group		
Group III		
Average similarity 52.95		
Microcystis aeruginosa	66.82	66.82
Cyclotella sp.	27.59	94.41
Groups I and II		
Average dissimilarity 68.09		
Microcystis aeruginosa	56.77	56.77
Cyclotella sp.	24.43	81.21
Ceraium furcoides	7.82	89.03
Cryptomonas sp.	2.44	91.47
Groups I and III		
Average dissimilarity 68.83		
Microcystis aeruginosa	65.67	65.67
Cyclotella sp.	18.23	83.90
Cosmarium phaseolus	3.52	87.42
Ceratium furcoides	2.71	90.13
Groups II and III		
Average dissimilarity 53.62		
Microcystis aeruginosa	44.53	44.53
Cyclotella sp.	35.67	80.20
Cosmarium phaseolus	5.43	85.63
Dinobryon sp.	2.93	88.56
Peridinium goslaviense	2.69	91.25

co-dominate with diatoms and desmids only in late July due to the presence of few (one individual per millilitre) but large-sized *Ceratium furcoides* individuals.

Cluster analysis separated the samplings in three groups (Fig. 4). Group I consisted of samples collected in late July and early August, during the first period of water withdrawal from the reservoir, Group II of the sample collected in late August when no considerable water withdrawal was observed, and Group III of samples collected in September, during the second period of water withdrawal from the reservoir. Results of SIMPER analysis (Table 3) indicated that *M. aeruginosa* was the species differentiating the three groups.

Water quality

Total phytoplankton biovolume exhibited values lower than the boundary (2.1 $mm^3\ l^{-1}$) of G/M ecological potential indicating good ecological water quality, with the exception of a short shift to moderate levels in early September when the highest value (2.7 $mm^3\ l^{-1}$) was recorded. On the contrary, the 88% contribution of cyanobacteria to the phytoplankton biovolume (the composition metric of phytoplankton) was much higher than the recommended boundary of 28% for G/M ecological potential.

According to WHO, the cyanobacteria biovolume (and particularly of *M. aeruginosa*) although very low in July, exceeded the Vigilance Level (one colony per millilitre) early in August and for the rest of the study period cyanobacteria exceeded the recommended threshold (0.2 $mm^3\ l^{-1}$) of Alert Level 1 for drinking water (Fig. 5).

Discussion

This is the first paper on phytoplankton and water quality of Marathonas Drinking-Water Reservoir. During the study period, the reservoir's water column was characterized by high transparency and relatively homogeneous depth distribution of pH and dissolved O_2 values. Thermal stratification was observed in July and August while mixing of the water column occurred in September.

The relatively high number of species identified in the reservoir's phytoplankton for the given period implies a rather diverse phytoplankton community, compared to the poor species richness of oligotrophic drinking-water reservoirs (e.g. Moustaka-Gouni and Nikolaidis 1992). The majority of the species belonged to functional groups that are typical of eutrophic water systems (Padisák et al. 2009). Many of them have a cosmopolitan distribution (e.g. *M. aeruginosa*,

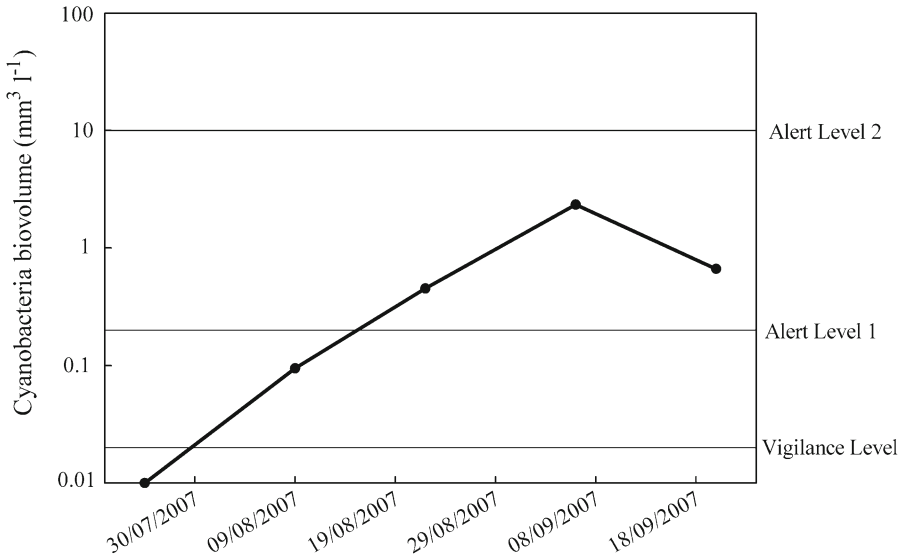

Fig. 5 Cyanobacteria biovolume during the study period (July–September 2007) in Marathonas Reservoir in relation to the Vigilance and Alert Levels for drinking water supplies proposed by the World Health Organization (Bartram et al. 1999). Values are means and refer to the euphotic zone

Dinobryon divergens, *Synedra acus*, *C. phaseolus*, *Tetraedron minimum*) while others are of special interest regarding their taxonomy and biogeography such as *A. bergii* (Gkelis et al. 2005b). Among the species identified, *M. aeruginosa* and *A. bergii* are well-known toxin-producing species (Schembri et al. 2001; Sivonen and Jones 1999). The occurrence of *M. aeruginosa* is well-documented in many eutrophic Mediterranean freshwaters (Cook et al. 2004) whereas *A. bergii* is reported in this paper for the first time in Greek freshwaters. Its close taxonomic relative *Aphanizomenon ovalisporum* has been reported from other Greek freshwaters used for drinking-water supply (Chrisostomou et al. 2009; Gkelis et al. 2005b).

Phytoplankton diversity in the reservoir, as expressed by species richness, was found to be maximum at the period of no considerable water withdrawal that followed pulses of increased flushing rate. Such flushing pulses with a peak every 4–7 days might act as intermediate disturbances triggering increased diversity as has been documented for intermediate disturbances in other freshwater systems and phytoplankton cultures (e.g. Gaedeke and Sommer 1986; Padisák et al. 1999). Phytoplankton biovolume did not reach high values during the study period. Among the 62 species identified, only four were dominant, as there was a great number of rare species (relative biovolume <1%). The dominance progressed from mid-summer diatoms (with desmids and dinoflagellates as subdominants) to late summer cyanobacteria, with a sudden shift to an equally shared assemblage of these two groups at the end of September. The summer dominance of cyanobacteria has been commonly observed in eutrophic lakes in Greece (e.g. Moustaka-Gouni 1993) and worldwide, regardless of their stratification pattern and geographical location (Padisák and Reynolds 1998). However, the change or share in dominance between diatoms and cyanobacteria implies less stable physical conditions (Reynolds et al. 1993). The short dominance of desmids on the other hand during the stratified period that is common in tropical lakes which exhibit a special mixing pattern referred as atelomixis or partial atelomixis (Barbosa and Padisák 2002; Souza et al. 2008) might be associated with a partial atelomixis in this Mediterranean reservoir.

In reservoirs, phytoplankton can be profoundly affected by the rather special hydrological regime and particularly by changing flushing rate (Naselli-Flores 1999; Padisák et al. 1999). In Marathonas Reservoir, phytoplankton composition and biovolume appear to be related to changes in water withdrawal and flushing rate. In July, a flushing rate with a maximum of 0.7% of reservoir's water volume per day occurring in pulses for a period of 25 days coincided with low phytoplankton biovolume. This phytoplankton biovolume consisted mainly by small, fast-growing species (*Cyclotella* sp., *C. phaseolus*). Species with similar life strategies dominated phytoplankton in another Greek reservoir under a similar hydrological regime (e.g. Moustaka-Gouni et al. 2000). Phytoplankton biovolume decrease was observed in late September coinciding with the lowest observed reservoir's water volume (Fig. 2). The question was whether inflows simply replaced the water volume by direct displacement or by a flushing action, in which the inflow volume mixed extensively with the standing volume (see Reynolds 2006). In September, phytoplankton individuals in the reservoir were in a mixed water column and were less affected by dilution. Furthermore, the preceding phytoplankton increase and cyanobacterial dominance were observed when the water volume decreased to the lowest level. This occurred after a period of no flushing in the reservoir indicating the flushing impact on phytoplankton dynamics.

In the peak of phytoplankton biovolume with the exclusive dominance of cyanobacteria, *M. aeruginosa* had the primary role. This species exhibited a maximum biovolume of 2.3 $mm^3\ l^{-1}$ and was not observed to increase further when subsequent flushing pulses persisted for 15–20 days and mixing prevailed. The impact of physical mixing on *Microcystis* and its replacement by diatoms is well known (Huisman et al. 2004). The possible impact of different flushing rates on *Microcystis* was shown by the separation of the samplings in three groups according to this species: (1) absence, (2) increase and peak and (3) decrease. During the study period, the reservoir's zooplankton (low abundance, rotifers as the dominant

group; Katsiapi et al. unpublished data) could not affect *Microcystis* negatively. Padisák et al. (1999) showed that *Microcystis* may tolerate flushing even at a range of 0.5–1% of reservoir's water volume per day (persisting for more than a month) when a considerable population biovolume has already developed. In the case of the hypertrophic Greek Lake Kastoria, a flushing rate of 0.53% of lake's volume per day in spring time proved very effective in breaking the increase of filamentous (both oscillatorialean and nostocalean)—sensitive to flushing—cyanobacteria that usually appeared and dominated early in summer (Moustaka-Gouni et al. 2006). Nevertheless, this did not affect *M. aeruginosa* which appeared late in the succession (Moustaka-Gouni et al. 2007).

The studied phytoplankton parameters are critical for the reservoir's ecological water quality (European Parliament Council 2000). In terms of phytoplankton biovolume, water quality was good with a slight deviation to moderate. However, the high contribution of cyanobacteria in the phytoplankton biovolume was not indicative of a good quality. In addition to the above phytoplankton metrics, both associated with the same pressure (i.e. eutrophication), species and functional group composition reflected a rather moderate ecological water quality. This is a first attempt to assess ecological water quality since neither the rules for combining different metrics for a phytoplankton biological element index have been established at a national and Mediterranean level nor all the types of Mediterranean reservoirs have been included in the Intercallibration Exercise (JRC European Commission 2009). Though the sampling period did not cover a whole year, it was concentrated in the period that is considered to be critical for water quality issues and monitoring (Padisák et al. 2006; Hajnal and Padisák 2008).

The implementation of the Alert Levels Framework proposed by WHO revealed that for a proper management of cyanobacteria in Marathonas Reservoir: (1) a suitable monitoring program is required and (2) analysis of cyanotoxin concentration in raw water is required once the presence of known toxic species is detected. In 2007, the biovolume of *M. aeruginosa*, known to produce toxins, exceeded the Alert Level 1 threshold in the reservoir. The molecular analysis of cyanobacterial diversity in the reservoir at the same period of this study revealed a phylotype related to *Microcystis*, which was practically identical to a *M. aeruginosa* strain whose genome bears the whole *mcy* operon of microcystin synthesis (Lymperopoulou et al. 2010). In addition, according to Kurmayer et al. (2003) larger colonies of *M. aeruginosa*, as was our case at the peak, are more likely to be toxic. Furthermore, *Microcystis* blooms identified in Greek freshwater systems have shown to be 100% toxin-containing (Gkelis et al. 2005a). Under such conditions, *Microcystis* blooms provide a serious threat for the water quality of Marathonas drinking-water Reservoir and immediate action must take place to minimize the risk of adverse human health effects.

In conclusion, phytoplankton dynamics during the warm period of the year in Marathonas Reservoir was characterized by shifts between 'good quality' species (small centric diatoms) and 'nuisance' ones (cyanobacteria). These shifts appear to be related to the reservoir's flushing and mixing regime indicating possible powerful tools for improving the reservoir's water quality; both by means of the implementation of the WFD for surface waters and for minimizing any potential human health risk.

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