Published in final edited form as:

Environ Toxicol. 2007 August ; 22(4): 399–404. doi:10.1002/tox.20268.

Toxic cyanobacterial blooms in reservoirs under a semiarid Mediterranean climate: the magnification of a problem

Luigi Naselli-Flores¹, Rossella Barone¹, Ingrid Chorus², and Rainer Kurmayer³

¹Dipartimento di Scienze Botaniche, Università di Palermo, Via Archirafi, 38, I-90123 Palermo, Italy. luigi.naselli@unipa.it; rossella.barone@unipa.it

²Federal Environment Agency, P.O. Box 330022, D-14191 Berlin, Germany, ingrid.chorus@uba.de

³Austrian Academy of Sciences, Institute for Limnology, Mondseestrasse 9, A-5310 Mondsee, Austria. rainer.kurmayer@oeaw.ac.at

Abstract

Sicilian reservoirs constitute the most important water resources available on the island. During summer 2001, the intense water utilization of Lake Arancio reservoir reduced the water level significantly. This coincided with the formation of intense blooms formed by the microcystin-producing cyanobacterium *Microcystis aeruginosa*. During summer 2003, Lake Arancio was continuously filled and the vertical stratification of the water column was maintained, resulting in 5–6 fold lower cell numbers of *Microcystis aeruginosa*. For both years a significant linear relationship between microcystin net production and *Microcystis* cell division was observed, implying that *Microcystis* cell numbers can be used to infer microcystin concentrations in water. Unexpectedly, dense blooms of the microcystin-producing cyanobacterium *Planktothrix rubescens* occurred during winter 2005/2006 in the reservoirs Lake Pozzillo, Prizzi, Nicoletti and Garcia, but have not been reported earlier. In this season, microcystin concentrations were higher than those recorded in summer. This implies that monitoring of Mediterranean drinking water reservoirs needs to be intensified during winter, a season usually considered to be less prone to the formation of cyanobacterial blooms.

Keywords

microcystins; *Microcystis aeruginosa*; *Planktothrix rubescens*; Sicily; risk assessment; environmental management

Introduction

Mediterranean reservoirs, especially those located in the semiarid part of the Mediterranean, represent a most important resource for human development. As compared to their temperate counterparts, semiarid environments are characterized by longer periods of relatively high temperatures favouring algal growth. Eutrophication of water resources by humans often deteriorates water quality and this may be enhanced due to the artificial nature of the reservoirs and the semiarid climate conditions. Because of its insularity, Sicily lacks both a permanent river network and aquifers large enough to ensure a minimum water supply to the reservoirs during the dry summer season (Naselli-Flores, 2003). Reservoirs fill up during the rainy season in autumn and winter and, in general, water residence times and nutrient loading increase during this time period (Kennedy et al., 2002). In consequence a higher algal production and algal populations adapted to an increased water depth have been observed (Naselli-Flores and Barone, 2005).

As a consequence of eutrophication mass developments of cyanobacteria have recently increased in intensity and duration in Sicilian reservoirs (Naselli-Flores and Barone, 2007). Since 1979, the relative importance of *Microcystis* and other planktonic cyanobacteria gradually increased at the expense of green algae typically dominating those reservoirs (Naselli-Flores and Barone, 2003, 2006). From 2000 onwards *Microcystis aeruginosa* and *M. panniformis* occurred with a frequency of 50-100% of total phytoplankton biovolume in 70% of the water stored in Sicilian reservoirs. Due to the Mediterranean climate characteristics, *Microcystis* spp. starts growing at the end of February and grows until October/November. Surprisingly, during winter 2005/2006, intense blooms of the filamentous cyanobacterium *Planktothrix rubescens* occurred in four reservoirs (Lakes Garcia, Nicoletti, Pozzillo, and Prizzi) which altogether contributed 30% of the total water volume stored in Sicily. Conversely to *P. agardhii*, which commonly develops in Sicilian eutrophic reservoirs at the end of summer, the occurrence of *P. rubescens* had never been recorded before on the island.

Planktonic cyanobacteria of the genera *Microcystis* and *Planktothrix* often produce small peptides, most prominently microcystins, that are toxic to a variety of organisms including humans (Chorus and Bartram, 1999). Beside the direct ingestion of microcystins via drinking, water contaminated with microcystins that is used for irrigation may inhibit germination and root growth, potentially causing crop failures (Pflugmacher et al., 2006). In particular, inhibition of germination and root growth of alfalfa seedlings was observed at relatively low microcystin concentrations of $5.0 \mu\text{g L}^{-1}$. The total quantity of microcystins produced by a cyanobacterial bloom varies in response to the proportion of microcystin-producing genotypes within a specific population (Kurmayer et al., 2002). Genotypes differing in morphological characteristics (so called morphospecies) in *Microcystis* spp. have been shown to vary in microcystin production (ViaOdorika et al., 2004). Because of this potential hazard to human health, cyanobacterial blooms need particular attention, and effective tools for surveillance need to be developed.

In this paper we present data on microcystin concentrations regularly gathered during summer 2001 and 2003 from the hypertrophic Lake Arancio, dominated by *Microcystis aeruginosa* blooms, as well as data from other Sicilian reservoirs dominated by blooms of *Planktothrix rubescens* collected during winter 2005/2006.

Material and Methods

Study sites

Table 1 shows morphometric characteristics of the reservoirs studied. Detailed morphological and limnological descriptions of the reservoirs are given in Naselli-Flores (1999). Due to the strong decrease in water volume during summer 2001 the thermal stratification of Lake Arancio was atelomictic (*sensu* Barbosa and Padisák, 2002) with a pronounced warming of the surface water during the day and diurnal, even irregular, mixing events resulting in large fluctuations in the redox conditions of the water column (Naselli-Flores, 2003). In 2003, changes in water level management resulted in a more stable water level and in the maintenance of thermal stratification during the summer period, thus reducing the internal nutrient transport from the sediments to the water column (Naselli-Flores and Barone, 2005). Autumn of 2005 was characterized by intense precipitation, especially in the central part of the island, and four reservoirs (Garcia, Nicoletti, Prizzi and Pozzillo), which had maintained summer stratification, were subjected to floods. These caused a sudden decrease in transparency and the abrupt breaking of their thermoclines.

Sampling

Water samples were collected from July – October weekly during the summer of 2001 and every two weeks during the summer of 2003. The Sicilian Regional Environmental Protection Agency (ARPA Sicilia) sampled the other four reservoirs during winter of 2005/2006. Water was collected 250 m from the dam at depths corresponding to 100%, 50% and 1% of the subsurface irradiance, as measured with a LI-COR quantum sensor (LI-COR Biosciences, Lincoln, Nebraska USA). The samples from different depths were mixed, and aliquots were used both for phytoplankton counting and biovolume estimation as well as for the analysis of microcystins. Water samples were kept cool and dark during the transport to the laboratory and were filtered through glass fibre GF/F filters (Whatman, Kent Great Britain) within two hours. The filters were folded and dried at 55-60 °C for two hours. The dry filters were put in a 100 cc glass bottle containing silica gel and stored at -20 °C.

Microscopical methods of plankton enumeration and the analysis of population dynamics of *Microcystis* spp. in Lake Arancio are given in Naselli-Flores and Barone (2003, 2005).

Briefly, Lugol fixed samples were diluted and counted in the inverted microscope after sedimentation over night. The different *Microcystis* morphospecies were identified in live samples according to Komárek et al. (2002). *Microcystis* colonies disintegrated in Lugol's solution and single cells were assigned to *M. aeruginosa* (large diameter,) and *M. panniformis* (small diameter, ...).

Microcystins were analysed as described previously (Fastner et al., 1999, Kurmayer et al., 2003). Briefly, filters were cut into pieces and extracted in 70% methanol (w/v) for three times. The clear supernatant was injected into high performance liquid chromatography coupled to diode array detection (HPLC-DAD). Microcystins were identified by their characteristic absorption maximum at 240 nm and quantified using microcystin-LR as external standard. Given a filtered volume of 100 ml and assuming an average cellular microcystin content of 100 fg cell⁻¹, a reliable quantification of microcystin was possible above a concentration of 60,000 cells ml⁻¹. Consequently, for cell concentrations below this threshold no cellular microcystin contents were calculated.

From biovolume and microcystin concentrations, cell division rates (r day⁻¹) and microcystin (net) production rates (day⁻¹) were calculated using the formula $r = (\ln N_2 - \ln N_1)/\Delta t$, where $N_{1,2}$ are the cell concentrations/peptide concentrations at consecutive sampling days and Δt is the time interval in days.

Results

Microcystin production by *Microcystis aeruginosa*

During summer 2001, *Microcystis aeruginosa* dominated the phytoplankton in Lake Arancio with a proportion of 93-100% of the total biovolume. The *Microcystis* morphospecies most frequently found included *M. aeruginosa* (91-100% with an average percentage of 97%). During most of the study period *M. panniformis* occurred with <2%, however, higher proportions were observed during the first two weeks of July and during the second week of October (min: 0, max: 6%, mean: 1.5%). Besides *Microcystis* spp., the assemblage included a few chlorophytes such as *Botryococcus braunii*, *B. terribilis* and *Pediastrum* spp. During the year 2003, *Microcystis* spp. reached a lower biovolume and occurred at subdominant proportions only (min: 1%, max: 37%, mean: 10%). The seasonal maxima of *Microcystis* spp. were observed during October of 2001 and September of 2003 (Fig. 1).

Total microcystin concentrations in the samples, expressed as microcystin-LR equivalents, ranged between 0-2753 µg L⁻¹. During both years *Microcystis aeruginosa* biovolume (BV) and the total microcystin (MC) concentration (expressed as microcystin-LR equivalents)

were closely correlated: $(MC[\mu\text{g L}^{-1}]=2.24 \times BV[\text{mm}^3 \text{L}^{-1}]-50.2; r^2=0.98, n=16, p<0.001)$. The maxima of microcystin concentrations, $2359 \mu\text{g L}^{-1}$ during 2001 and $40 \mu\text{g L}^{-1}$ during 2003, coincided with the maxima of *Microcystis aeruginosa* biovolume.

The dominant microcystin variants were microcystin-YR and -LR, while microcystin-RR occurred in lowest proportions only (<2%), and further variants were present in yet lower shares of the total content. In the two years the proportion of microcystin-YR/microcystin-LR was almost constant (min: 60%, mean: $73\% \pm 1.6(\text{SE})$, max: 79%). The microcystin content of the *Microcystis aeruginosa* biovolume differed significantly between the two years ($p<0.01$), with 0.9 to $3.5 \mu\text{g}$ (mean: 1.8 ± 0.2) of microcystin-LR equivalents per mm^3 of biovolume during 2001, and 0.3 to $1.2 \mu\text{g}$ (mean: 0.72 ± 0.20) during 2003 (Fig. 2). During 2001, the microcystin content was constant until the mid of August, and then increased until the mid of September. This increase in microcystin content coincided with a period of low physical stability of the water column due to the yearly minimum content of water in the reservoir. This period was also characterized by an improvement in the underwater light availability and by a re-oxygenation of the water column (Naselli-Flores, 2003; Naselli-Flores and Barone, 2003) favouring a strong increase in *Microcystis* cells. Although microcystin contents were lower during the summer of 2003, for both years the microcystin net production rates were related very closely, i.e. almost one to one, to the *Microcystis aeruginosa* cell division rate calculated on the basis of the biovolume (Fig. 3). Consequently, the seasonal variation in microcystin content was considered of minor importance relative to the dependence of microcystin net production on the increase or decrease of *Microcystis aeruginosa* biovolume.

Microcystin production by *Planktothrix rubescens*

During winter 2005/2006, *P. rubescens* formed surface blooms in Lake Pozzillo, the largest Sicilian reservoir. A population density peak ($>50 \times 10^9 \text{ cells L}^{-1} \approx 5600 \text{ mm}^3 \text{L}^{-1}$) was reached at the end of December 2005. *P. rubescens* coloured the water surfaces dull purple by a 60 cm thick layer floating on the surface. At the beginning of March the density of filaments decreased down to $<20 \text{ filaments mL}^{-1}$ ($10 \times 10^6 \text{ cells L}^{-1} \approx 1.1 \text{ mm}^3 \text{L}^{-1}$). *P. rubescens* was also observed in the other three reservoirs (Garcia, Nicoletti and Prizzi) and showed lower cell numbers but a similar seasonal development in population density increase. Cell numbers of *Planktothrix* increased in mid-November at the end of the stratification, and *Planktothrix* persisted until mid-February, when the water temperature was at the annual minimum (9-10 °C).

Analysis of microcystins in a surface sample collected at highest cell density (December 2005) in Lake Pozzillo revealed the presence of demethylated microcystin variants of microcystin-RR (95%), microcystin-LR (3%) and small amounts of unknown microcystin variants (2%). The total concentration of microcystin (calculated as equivalents of MC-LR) was 34 mg L^{-1} . This concentration corresponds to a microcystin content of $6.0 \mu\text{g mm}^{-3}$ of biovolume. After this peak, lower microcystin concentrations were detected, i.e. the microcystin concentration in a surface sample from Lake Prizzi at 15 February 2006 was $7 \mu\text{g L}^{-1}$ (microcystin-RR determined as microcystin-LR equivalents).

Discussion

In this study the microcystin content of the *Microcystis* population varied seasonally by a factor of four. This range corresponds to the results of laboratory studies, which in most cases also report a factor of three to four difference between cultures grown under different environmental conditions (i.e. micro- and macronutrients, light, temperature, pH, Sivonen and Jones, 1999).

In their unifying theory, Orr and Jones (1998) suggested microcystin net production to be coupled linearly to the cell division of the organism and concluded that – although microcystin is a secondary metabolite – it rather displays the attributes of essential intracellular nitrogenous compounds in cyanobacteria. According to this theory, the strongest influence of all possible environmental factors is indirect – through their effect on cell division and growth, whereas the direct effects of environmental conditions on microcystin biosynthesis are of minor importance.

The situation becomes more complex in the field since in contrast to the laboratory numerous different genotypes co-exist and therefore may influence the microcystin concentration in the biovolume and water. The wax and wane of microcystin-producing vs. non-microcystin-producing genotypes has been suggested as a most important factor regulating microcystin net production in the field (Sivonen and Jones, 1999). In our study, a significant linear relationship between microcystin net production and *Microcystis* cell division was observed, implying that the number of co-existing microcystin-producing and non-microcystin-producing genotypes was seasonally stable, although it may have differed between the years 2001 and 2003. We cannot exclude the occurrence of shifts within the group of microcystin-producing genotypes. For example Amé and Wunderlin (2005) identified iron and ammonium concentrations and high temperature as factors that might influence the proportion of microcystin-producing genotypes. Indeed, during summer of 2001, Lake Arancio showed an atelomictic stratification with diurnal and irregular mixing events causing large fluctuations in the redox conditions of the water column. However, nonetheless the proportion of microcystin variants, microcystin-LR, YR and RR was relatively constant during both years, implying that shifts among genotypes producing different microcystin variants scarcely occurred. Notably, a number of field studies also reported significant linear relationships between microcystin net production and *Microcystis* biovolume (Kotak et al., 2000; Chorus et al., 2001; Kurmayer et al., 2003). Even the re-analysis of data of a study postulating that seasonal and spatial shifts in microcystin genotype composition determine microcystin net production in Lake Biwa (Ozawa et al., 2005) revealed a significant linear relationship between microcystin concentrations and *Microcystis* cell numbers ($y=3.15+0.08x$, where y was the microcystin concentration in $\mu\text{g ml}^{-1}$ and x was the cell concentration in cells ml^{-1} , $R^2=0.59$, $n=85$, cell contents of $>1000 \text{ fg cell}^{-1}$ were considered as outliers and have not been included). This evidence is of importance in order to use *Microcystis* cell numbers or biovolumes to predict microcystin concentrations in water. Thus, cell numbers and/or biovolume can be important surrogate parameters for authorities and water managers performing environmental monitoring and risk assessment. They become most meaningful through occasional microcystin analyses and determination of the ratio of microcystin per cell or per unit biovolume. Once this ratio is known to remain stable in a given water-body, conducting microcystin analyses once or 2-3 times per growing season would be sufficient to validate the prediction of microcystin concentrations in the water from cell density or biovolume determined more frequently, e.g. weekly or in 14-day intervals. This approach is particularly useful for water-bodies with unialgal cyanobacterial populations that persist for extended periods of time.

As shown by Naselli-Flores and Barone (2005), the water level reduction during summer in Sicilian reservoirs enhanced the cell division of *Microcystis* spp. *Vice versa* a different water management, i.e. continuously refilling the reservoir and maintaining the vertical stratification reduced internal nutrient loading and by this the cell division of *Microcystis* spp. On the other hand, these stratifying conditions may favour the occurrence of shade-adapted species such as *Planktothrix rubescens*. *P. rubescens* typically occurs in deep, physically stratified and oligo- to mesotrophic waters in which it can form metalimnetic layers because light penetrates to this depth (Walsby et al., 2005). It has been shown both empirically and by modelling that during periods of low insolation such as in December the

filaments receive low light doses only and react by floating up from deeper regions of the lake, thus forming visible water blooms (Walsby et al., 2005).

It is unclear why *P. rubescens* was observed only recently and not during the regular phytoplankton monitoring in the years before. *P. rubescens* has been known for its sudden mass appearances from other studies (Padisák et al., 2003). During the last years the autumnal precipitation increased, as predicted by current climate models (Tin, 2006). High precipitation events occurred during early November 2005 (Sicilian Hydrological Service, unpublished data), causing a mixing of the water column but also decreasing the water transparency of the reservoirs. These conditions may have favoured low light adapted phytoplankton species such as *Planktothrix* spp. In general, *P. agardhii* has been shown to outcompete green algae in shallow, eutrophic lakes (Scheffer et al., 1997). *P. rubescens* has a highly efficient light harvesting complex as well, and frequently dominates deep reservoirs of the temperate climatic zone. *P. rubescens* has been suggested to outgrow *P. agardhii* under low light conditions and lower temperatures in the stratified lake Blelham Tarn (<21°C) while *P. agardhii* may be more successful in shallow and warmer waters (Davis and Walsby, 2002).

The pinkish colour of these blooms frequently alerts the population. Due to concern of toxicity as well as public perception, local authorities stopped the water use in this reservoir until the beginning of March when the density of filaments decreased below 20 ml⁻¹ (about 10 × 10⁶ cells L⁻¹ ≈ 1.1 mg L⁻¹). The same measure was taken for the drinking water reservoirs Garcia and Prizzi. Red pigmented *P. rubescens* (*sensu* Suda et al., 2002) always produces microcystins (Kurmayer et al., 2004). Similarly as for *Microcystis aeruginosa* for *Planktothrix rubescens* significant correlations between the cell numbers and the microcystin concentrations have been reported (Briand et al., 2005). Consequently the counting of filaments/cell numbers can be useful to infer potential microcystin concentrations in water dominated by *P. rubescens*. However, compared with *Microcystis aeruginosa* in Lake Arancio the *Planktothrix* showed higher population densities leading to higher microcystin net production rates, and it contained 2-6 fold more microcystin (6.2 µg mm⁻³ biovolume). This study demonstrates that health risks caused by microcystin-producing cyanobacteria in reservoirs are present during winter, as well as during summer, and if overlooked may have public health consequences. Consequently, water monitoring efforts need to be intensified during winter, a season that is usually considered to be less prone to the formation of surface sums in the temperate region of the Northern hemisphere.

Acknowledgments

Financial support for microcystin analyses was provided by the EU R&D PEPCY project, contract no. QLK4-CT-2002-0263 and the Austrian Science Funds FWF P18185. We thank Inge Flieger for conducting the microcystin analyses of the samples from 2001. We are grateful to Judith Padisák for detailed comments on the earlier version of the manuscript.

References

- Amé MV, Wunderlin DA. Effects of iron, ammonium and temperature on microcystin content by a concentrated *Microcystis aeruginosa* natural population. *Water Air Soil Poll.* 2005; 168:235–248.
- Barbosa FAR, Padisák J. The forgotten lake stratification pattern: atelomixis, and its ecological importance. *Verh int Verein Limnol.* 2002; 28:2381–2392.
- Briand JF, Jacquet S, Flinois C, Avois-Jacquet C, Maisonneuve C, Leberre B, Humbert JF. Variations in the microcystin production of *Planktothrix rubescens* (cyanobacteria) assessed from a four-year survey of Lac du Bourget (France) and from laboratory experiments. *Microb Ecol.* 2005; 50:418–428. [PubMed: 16328657]
- Chorus, I.; Bartram, J. Toxic Cyanobacteria in water. E & FN Spon; London: 1999. p. 416

- Chorus, I.; Niesel, V.; Fastner, J.; Wiedner, C.; Nixdorf, B.; Lindenschmidt, K-E. Environmental factors and microcystin levels in water bodies. In: Chorus, I., editor. *Cyanotoxins. Occurrence, Causes, Consequences*. Springer; Berlin: 2001. p. 159-177.
- Davis PA, Walsby AE. Comparison of measured growth rates with those calculated from rates of photosynthesis in *Planktothrix* spp. isolated from Blelham Tarn, English Lake District. *New Phytol.* 2002; 156:225–239.
- Fastner J, Erhard M, Carmichael WW, Sun F, Rinehart KL, Röncke H, Chorus I. Characterization and diversity of microcystins in natural blooms and strains of the genera *Microcystis* and *Planktothrix* from German freshwaters. *Arch Hydrobiol.* 1999; 145:147–163.
- Kennedy, RH.; Cole, T.; Boyd, W.; Barko, K. Operational influences on the limnological characteristics of reservoirs: a model study; Extended Abstracts of the 4th International Conference on Reservoir Limnology and Water Quality; eské Bud jovice, Czech Republic. August 12-16, 2002; 2002. p. 168-170.
- Komárek J, Komárková-Legnerová J, Sant' Anna C, de Paiva Azevedo MT, Senna PAC. Two common *Microcystis* species from tropical America. *Cryptogamie Algol.* 2002; 23:159–177.
- Kotak BG, Lam AKY, Prepas EE, Hrudey SE. Role of chemical and physical variables in regulating microcystin-LR concentration in phytoplankton of eutrophic lakes. *Can J Fish Aquat Sci.* 2000; 57:1584–1593.
- Kurmayer R, Dittmann E, Fastner J, Chorus I. Diversity of microcystin genes within a population of the toxic cyanobacterium *Microcystis* spp. in Lake Wannsee (Berlin, Germany). *Microb Ecol.* 2002; 43:107–118. [PubMed: 11984633]
- Kurmayer R, Christiansen G, Chorus I. The abundance of microcystin-producing genotypes correlates positively with colony size in *Microcystis* and determines its microcystin net production in Lake Wannsee. *Appl Environ Microbiol.* 2003; 69:787–795. [PubMed: 12570996]
- Kurmayer R, Christiansen G, Fastner J, Börner T. Abundance of active and inactive microcystin genotypes in populations of the toxic cyanobacterium *Planktothrix* spp. *Environ Microbiol.* 2004; 6:831–841. [PubMed: 15250885]
- Naselli-Flores, L. Limnological aspects of Sicilian reservoirs: a comparative ecosystemic approach. In: Tundisi, JC.; Straškraba, M., editors. *Theoretical reservoir ecology and its applications*. Backhuys Publishers; Leiden: 1999. p. 283-311.
- Naselli-Flores L. Man-made lakes in Mediterranean semi-arid climate: The strange case of Dr Deep Lake and Mr Shallow Lake. *Hydrobiologia.* 2003; 506/509:13–21.
- Naselli-Flores L, Barone R. Steady-state assemblages in a Mediterranean hypertrophic reservoir. The role of *Microcystis* ecomorphological variability in maintaining an apparent equilibrium. *Hydrobiologia.* 2003; 502:133–143.
- Naselli-Flores L, Barone R. Water-level fluctuations in Mediterranean reservoirs: setting a dewatering threshold as a management tool to improve water quality. *Hydrobiologia.* 2005; 548:85–99.
- Naselli-Flores L, Barone R. Pluriannual morphological variability of phytoplankton in a highly productive Mediterranean reservoir (Lake Arancio, Southwestern Sicily). *Hydrobiologia.* 2007 in press.
- Orr PT, Jones GJ. Relationship between microcystin production and cell division rates in nitrogen-limited *Microcystis aeruginosa* cultures. *Limnol Oceanogr.* 1998; 43:1604–1614.
- Ozawa K, Fujioka H, Muranaka M, Yokoyama A, Karagami Y, Homma T, Ishikawa K, Tsujimura S, Kumagai M, Watanabe MF, Park H-D. Spatial distribution and temporal variation of *Microcystis* species composition and microcystin concentration in Lake Biwa. *Environ Toxicol.* 2005; 20:270–276. [PubMed: 15892068]
- Padisák J, Scheffler W, Kasprzak P, Koschel R, Krienitz L. Interannual variability in the phytoplankton composition of Lake Stechlin (1994-2000). *Arch. Hydrobiol. Spec. Issues Advanc. Limnol.* 2003; 58:101–133.
- Pflugmacher S, Jung K, Lundvall L, Neumann S, Peuthert A. Effects of cyanobacterial toxins and cyanobacterial cell-free crude extract on germination on alfalfa (*Medicago sativa*) and induction of oxidative stress. *Environ Toxicol Chem.* 2006; 25:2381–2387. [PubMed: 16986793]
- Scheffer M, Rinaldi S, Gragnani A, Mur LR, van Nes EH. On the dominance of filamentous cyanobacteria in shallow, turbid lakes. *Ecology.* 1997; 78:272–282.

- Sivonen, K.; Jones, G. Cyanobacterial toxins. In: Chorus, I.; Bartram, J., editors. Toxic Cyanobacteria in Water. A Guide to Their Public Health Consequences, Monitoring and Management. E & FN Spon, WHO; London: 1999. p. 41-112.
- Suda S, Watanabe MM, Otsuka S, Mahakahant A, Yongmanitchai W, Nopartnaraporn N, Liu Y, Day JG. Taxonomic revision of water-bloom-forming species of oscillatoroid cyanobacteria. *Int J Syst Evol Microbiol.* 2002; 52:1577–1595. [PubMed: 12361260]
- Tin, T. Mediterranean and climate change; Proceedings of the Workshop “Mediterranean: Water and Drought”, Zaragoza (Spain); Sevilla: CENTA. 23-24 February 2006; 2006.
- Via-Ordorika L, Fastner J, Kurmayer R, Hisbergues M, Dittmann E, Komárek J, Erhard M, Chorus I. Distribution of microcystin-producing and non-microcystin-producing *Microcystis* sp. in European freshwater bodies: detection of microcystins and microcystin genes in individual colonies. *Syst Appl Microbiol.* 2004; 27:592–602. [PubMed: 15490561]
- Walsby AE, Schanz F, Schmid M. The Burgundy-blood phenomenon: a model of buoyancy change explains autumnal waterblooms by *Planktothrix rubescens* in Lake Zürich. *New Phytol.* 2005; 169:109–122. [PubMed: 16390423]

Fig. 1. Biovolume of *Microcystis aeruginosa* from July – October 2001 (upper graph) and 2003 (lower graph) in Lake Arancio and corresponding concentrations of microcystin (MC-LR and MC-YR estimated as MC-LR equivalents).

Fig. 2. Microcystin content in *M. aeruginosa* from July–October of 2001 (straight line) and 2003 (dashed line) in Lake Arancio.

Fig. 3. Linear relationship between the cell division rate of *Microcystis aeruginosa* calculated from biovolume estimates in the microscope and the net production rate of microcystin-LR equivalents during 2001 (black diamonds) and 2003 (open triangles).

Table 1

Main characteristics of the reservoirs studied in Sicily

Reservoir	Volume [10 ⁶ m ³]	Surface [km ²]	Maximum depth [m]	Average depth [m]	Use	Trophic State
Arancio	30	3.2	29	9	Irrigation	Eu-Hyper
Garcia	60	5.9	43	10	Drinking	Meso-Eu
Nicoletti	17	1.8	36	9	Irrigation	Meso
Pozzillo	154	7.7	50	18	Hydropower/irrigation	Meso-Eu
Prizzi	9	1.3	44	6	Drinking	Meso-Eu