Oceanological and Hydrobiological Studies

International Journal of Oceanography and Hydrobiology

Volume 45, Issue 4, December 2016
pages (466-484)

ISSN 1730-413X
eISSN 1897-3191

Occurrence of microcystins and anatoxin-a in eutrophic lakes of Saint Petersburg, Northwestern Russia

by

Ekaterina Chernova*, Iana Russkikh, Ekaterina Voyakina, Zoya Zhakovskaya

DOI: **10.1515/ohs-2016-0040**
Category: **Original research paper**
Received: **February 25, 2016**
Accepted: **May 11, 2016**

Department of Eco-chemical Studies, Saint-Petersburg Scientific-Research Centre for Ecological Safety, Russian Academy of Sciences, Korpusnya 18, 197110 Saint Petersburg, Russian Federation

Abstract

Cyanobacteria are natural components of many freshwater bodies worldwide. In Russian lakes, the presence of potentially toxic cyanobacteria was also frequently observed. Our study was conducted in Sestroretskij Razliv Lake (Razliv) and Lower Suzdal Lake (Suzdal) in Saint Petersburg region, Northwestern Russia, which differ from one another in eutrophic status and composition of the phytoplankton community. In large, shallow, artificial and hypertrophic Razliv, *Aphanizomenon flos-aquae* and *Microcystis* spp. dominated. Fourteen microcystin variants were identified in this lake. The maximum concentration of extracellular microcystins was 41.37 µg l^{-1}. In eutrophic and shallow Suzdal, dominated by *Planktothrix agardhii*, nine microcystin variants and anatoxin-a (<0.54 µg l^{-1}) were found. The maximum total concentration of extracellular MCs in this lake was 2.89 µg l^{-1}. Regular studies into the production of cyanotoxins in these water bodies were carried out for the first time. The analyses performed with the application of high-resolution tandem mass spectrometry revealed the presence of microcystins in 59% of the samples collected during a 3-year study. Since both lakes are used for recreational purposes, the regular monitoring program should be implemented to protect water users from a potential risk that was identified in our study.

Key words: microcystins, anatoxin-a, mass spectrometry, Northwestern Russia

* Corresponding author: *s3561389@ya.ru*

DE GRUYTER

The *Oceanological and Hydrobiological Studies* is online at oandhs.ocean.ug.edu.pl

Introduction

In the last two decades, the health risk associated with the presence of cyanobacteria in lakes, rivers, reservoirs, and ponds has been widely recognized. At least 46 toxin-producing cyanobacterial species were identified (Apeldoorn et al. 2007). Moreover, both toxic and non-toxic strains were identified within a given species in the same habitat (Osswald et al. 2007). Simultaneous production of several variants of hepatotoxic microcystins, the most common group of cyanotoxins, and less common neurotoxic anatoxin-a was also reported (Rantala et al. 2006).

Microcystins are persistent compounds and they accumulate in different compartments of aquatic environments, including fish, mussels and sediment. These compounds are highly toxic and different exposure routes are possible (Chorus 2012). The World Health Organization established a guideline value for the daily intake of microcystin-LR (MC-LR), i.e. the most common and most toxic MC congener (WHO, 2011). In many countries, contact with cyanobacteria at bathing sites is the most probable route of exposure to cyanotoxins and can result in health problems, particularly in children. According to the European Bathing Water Directive 2006/7/EC, when the bathing water profile indicates a potential for cyanobacterial proliferation, an appropriate monitoring program should be implemented (Directive 2006/7/EC).

In Russia, there are over 2.7 million lakes with a total water surface area of about 409 000 km^2. The lake area covers about 4% of the territory of the Russian Federation. Most lakes (98%) are small (with the surface area below 1 km^2) and shallow (average depth 1.0-1.5 m). Many lakes (14%) are situated in Northwestern Russia. According to hydrobiological data, green algae, diatoms and cyanobacteria dominate in the lakes (Trifonova & Pavlova 2008).

Reports on the occurrence of cyanobacteria and cyanotoxins in Russia are scarce and limited to several presentations at international conferences. Occasional studies on cyanotoxins were conducted in Central Russia, in particular in Volga Reservoirs (Korneva et al. 2014; Sidelev et al. 2015) and Lake Nero (Babanazarova et al. 2011), in lakes of Northwestern Russia, including Lake Ladoga (Gromov et al. 1996) and Karelian Isthmus lakes (Voloshko et al. 2008), and Lake Baikal in East Siberia (Belykh et al. 2015). Different analytical methods (PCR, ELISA, LC-MS) were used in the above-mentioned works to detect and quantify the microcystins. The obtained results showed the presence of toxigenic species and cyanotoxins in the studied water bodies.

Cyanobacterial blooms have also been reported from water bodies located in Saint Petersburg region. Limnologic studies conducted in these lakes during the last 50 years showed a shift in the dominant species from diatoms to cyanobacteria (Trifonova & Pavlova 2008). In addition, an increase in the range and intensity of phytoplankton blooms was observed and attributed to anthropogenic impact and increasing eutrophication. It was shown that phytoplankton biomass in Sestroretskij Razliv Lake increased six to seven times for the last two decades (Trifonova & Pavlova 2008).

Our preliminary studies conducted in two eutrophic lakes located in the territory of Saint Petersburg and Leningrad Region, Razliv and Suzdal, documented the dominance of *Microcystis* spp. and *Planktothrix agardhii* in the phytoplankton community (Russkikh et al. 2012). These cyanobacteria belong to the most efficient producers of microcystins (Apeldoorn et al. 2007, Welker 2008), therefore, the use of these water bodies for recreational purposes by the population of the megalopolis can be potentially dangerous.

This paper presents the results of the regular research performed for the first time on the toxin production by cyanobacteria occurring in freshwater reservoirs located in Northwestern Russia, Razliv and Suzdal. The research covered a 3-year period (2010-2012), from May to October. Data on phytoplankton composition, as well as on intra- and extracellular concentration of cyanotoxins were collected. The aim of the study was to assess the potential threat to water users related to the occurrence of toxic cyanobacteria at the examined bathing sites.

Materials and methods

Study site and sampling

The present study was conducted in two shallow, humic, eutrophic lakes with a different nutrient status – Lake Sestroretskij Razliv (Razliv) and Lower Suzdal Lake (Suzdal) – located in Saint Petersburg region, Russia, and used for recreational purposes. Razliv is a large artificial reservoir with a surface area of 1100 ha and average depth of 1.6 m. The surface area of Suzdal is 97 ha and the mean depth is 3.0 m.

Surface water samples (1 l) were collected from these lakes with plastic bottles and phytoplankton biomass was collected with a net (mesh size of 85 µm). Sampling was performed every 2 weeks from May to October in 2010-2012. Altogether, 45 water samples and 40 phytoplankton samples were collected from

the two lakes. The water samples in 1 l bottles were fixed with a Lugol-formalin solution. Cyanobacterial material from net samples was transported to a laboratory in screw-cap tubes (Axigen, California, USA) and immediately frozen at –20°C until freeze-drying.

Phytoplankton analysis

Qualitative and quantitative analyses of phytoplankton were carried out under a light microscope (Mikromed-3, LOMO, Saint Petersburg, Russia). The volume of the counting chamber Uchinskaya was 0.1 ml. The biovolume of algae and cyanobacteria was calculated using species-specific geometric formulas (Olenina et al. 2006). The phytoplankton biomass was determined from the total volume of algae according to the counted cell density and the measured average cell density.

Chemicals

Microcystin standards (MC-LR, MC-RR, and MC-YR) were purchased from Sigma-Aldrich (St. Louis, MO, USA) and (+/-) anatoxin-a fumarate was obtained from Tocris Bioscience (Bristol, UK). Acetonitrile (HPLC-grade) and methanol (LiChrosolv hypergrade for LC-MS) were purchased from Merck (Darmstadt, Germany). Water was purified to 18.2 MΩ cm^{-1} in a Millipore Direct-Q water purification system (Bedford, MA, USA). Formic acid was obtained from Fluka Chemika (Buchs, Switzerland). Membrane filter discs (nylon 66, 47-mm diameter) were obtained from Supelco (Bellefonte, PA).

Sample preparation for LC-MS toxin analysis

Water samples (1 l) for the analysis of extracellular cyanotoxins (AN and MCs) were filtered on a Supelco mobile-phase filtration apparatus (nylon-66 membrane filter discs, 47-mm diameter, Supelco, Bellefonte, PA). Solid phase extraction (SPE) with Oasis HLB cartridges (60 or 200 mg, Waters, Milford, Massachusetts, USA) was used. The toxins were eluted from the cartridges with 10 ml methanol. The collected extracts were dried using a rotary evaporator and stored at –20°C until further analysis. The dried extracts were reconstituted in 1 ml of 80% aqueous methanol and centrifuged (CM-50 centrifuge, ELMI, Riga, Latvia) at 14 000 rpm for 10 min prior to analysis by high-performance liquid chromatography/high-resolution tandem mass spectrometry (LC-HR-MS/MS).

For the extraction of biomass-bound (intracellular) MCs and anatoxin-a, the dry lyophilized cells (10-30 mg) were treated twice with 1 ml of 75% aqueous methanol by sonication in an ultrasound bath (US) for 15 min, and then centrifuged at 14 000 rpm for 15 min. The obtained extracts were combined.

LC- MS

The LC-MS experiments were carried out in the Accela HPLC system (Thermo Finnigan, San Jose, CA, USA) coupled with a Hybrid Ion Trap-Orbitrap Mass Spectrometer – LTQ Orbitrap XL (Thermo Fisher Scientific, San Jose, USA) with the electrospray interface. Separation of the toxins was performed on a Thermo Hypersil Gold RP C18 column (100 × 3 mm, 3 µm) with a Hypersil Gold drop-in guard column (Thermo Fisher Scientific). The mobile phase consisted of water (solvent A) and acetonitrile (solvent B), both containing 0.05% formic acid. The gradient program started at 5% B (held for 10 min), then the content of phase B increased to 75% over 18 min (held for 3 min), and then increased to 95% B over 1 min (held for 5 min). The flow rate was 0.2 ml min^{-1}. The column temperature was 40°C and the injection volume was 25 µl for water sample extracts and 10 µl for biomass sample extracts.

The MS conditions using electrospray ionization were as follows: Ion Spray voltage 3.2 kV, capillary Voltage 20 V, ion-transfer capillary temperature 320°C, tube lens 130 V. The mass spectrometer operated in the positive ion mode at a resolving power of 30 000. A scan function (*m/z* 500–1200) for the detection of all MC variants was used. The identification of target compounds, whose standards were available (AN and MC-LR, -RR, -YR), was based on the accurate mass measurement of $[M+H]^+$ or $[M+2H]^{2+}$ ions (accuracy within 5 ppm), the collected fragmentation spectrum of the ions and the retention times (coincidence with the analytical standard within 0.3 min). The detection of anatoxin-a was based on the presence of a peak with *m/z* at 166.12 and retention time corresponding to the anatoxin-a standard. The structure of the neurotoxin was confirmed using fragmentation spectra (MS/MS) of the precursor ion with *m/z* 166.12 covering *m/z* 110-170. Other MC congeners, whose standards were not available, were identified based on the exact *m/z* values for described brutto formulas (Mayumi et al. 2006; Furey 2008), calculated using the NIST IsoForm program ver. 1.02 (NIST Formula and Isotopic Pattern Generator, NIST, USA) and fragmentation spectra of their pseudomolecular ions.

For quantitative analysis of cyanotoxins, the calibration curves for standards solutions of AN and MC-LR, -RR, and -YR were determined (r^2 = 0.996-0.998) within a calibration range of 5-500 ng ml^{-1}. Procedural LODs were 3-6 ng l^{-1} for water samples (SPE extraction) and 0.08-0.30 µg g^{-1} DW for freeze-dried biomass

(ultrasonic extraction), respectively. Procedural LOQs were 9-20 ng l^{-1} for natural water samples and 0.27-0.90 µg g^{-1} for freeze-dried biomass, respectively.

Measurements of the concentrations of the singly charged MC variants, whose standards were not available, were based on the standard curve for MC-LR. As there were large variations in the MS response of the singly charged MCs and the doubly charged ions of different MC-RR congeners, the concentrations of their desmethylated (dm) and methylated counterparts were determined on the basis of the curve drawn for *m/z* 519.79 of the MC-RR standard.

Results

Hydrochemical parameters

Major hydrochemical parameters for both lakes were similar. The maximum gradients for ammonia nitrogen (0.6-2.0 mg N l^{-1}) and phosphates (0.002-0.600 mg P l^{-1}) were typical of eutrophic lakes of Northwestern Russia. During the study period, the pH values in the lakes changed within the range of 6.2-8.5 in Razliv, and within the range of 7.6-9.0 in Suzdal.

Phytoplankton composition

The seasonal cyanobacterial dynamics in Razliv and Suzdal was determined based on the analysis of 41 phytoplankton samples. The total number of phytoplankton taxa identified in the collected samples was 138. The identified species belonged to nine taxonomic groups of algae, with the dominance of green algae (41% of the total number of species), cyanobacteria (18%), euglenoids (13%), and diatoms (11%). Among green algae, the family Chlorococcoaceae was represented by the largest number of species. The data on the abundance and biomass of the observed taxonomic groups are presented in supplementary tables (Supplementary tables S1A-S1F).

Both lakes were described as β-mezosaprobic. During the study period, the phytoplankton biomass varied significantly within and between the seasons and lakes (Fig. 1). Among the identified species, the following were most numerous: *Aphanizomenon flos-aquae*, *Planktothrix agardhii* (Cyanobacteria), *Aulacoseira ambigua*, *Aulacoseira islandica*, *Aulacoseira italica* (Bacillariophyta), and *Cryptomonas rostrata* (Cryptophyta).

The highest mean seasonal cyanobacterial biomass was recorded in 2012 in Suzdal (10.97 mg l^{-1}) and in 2011 in Razliv (15.05 mg l^{-1}). The contribution of cyanobacteria to the total phytoplankton biomass varied from 0.4% (0.02 mg l^{-1}) to 99% (61.94 mg l^{-1}) in Razliv and from 0.1% (0.01 mg l^{-1}) to 41% (26.03 mg l^{-1}) in Suzdal (Fig. 1, Supplementary tables S1A-S1F).

In 2010 and 2012, *Aph. flos-aquae* and *Dolichospermum flos-aquae* dominated in the plankton community in Razliv at the beginning of the sampling seasons. In summer, the dominance shifted to representatives of the genus *Microcystis* (*M. aeruginosa, M. wesenbergii* and *M. viridis*) (Fig. 1 A, C). In 2011, *Microcystis* species and *Aph. flos-aquae* co-occurred almost throughout the whole summer season (Fig. 1 B). In Suzdal, the variability in the dominant species composition was not as significant as that in Razliv. In 2010 and 2012, the dominance of *P. agardhii* and *Aph. flos-aquae* in the cyanobacterial community at the beginning of the warm season shifted in August-September to *Microcystis* spp. (Fig. 1D, F). During 2011, the quantitative dominance of *P. agardhii* over *Aph. flos-aquae* was observed (Fig. 1 E).

Toxin concentrations

The studies carried out in 2010-2012 showed the occurrence of cyanotoxins both in Razliv and in Suzdal. The concentrations of microcystins exceeding the value of ≥ 0.1 µg l^{-1} for the extracellular fraction, or 0.1 mg g^{-1} on a dry-weight (DW) basis for the intracellular fraction, were determined in 54 water and biomass samples (59% of all the collected samples). Extracellular microcystins were detected in 25 of 46 water samples (54% of the water samples) (Tables 1, 2, Supplementary tables S2-S7). AN was present only in 5 water samples from Suzdal (Table 2, Supplementary tables S5, S6).

The median seasonal and maximum concentrations of the total MCs, extracellular and intracellular MCs concentrations, and the concentrations of the three main toxins identified in the two water bodies by LC-MS/MS in each sampling period are shown in Tables 1 and 2. The sampling periods are highlighted in the tables according to the change in the dominant species. The total number of detected MCs variants during our three-year study was 14 in Razliv, and 9 in Suzdal. The number and structures of microcystin congeners identified in the lakes are presented in Table 3.

Although cyanobacterial biomass was not high in 2010 (Fig. 1A, 1D), the median values for total extracellular toxins reached their maxima in Razliv (1.61 µg l^{-1}) and Suzdal (0.44 µg l^{-1}) (Tables 1, 2). The maximum extracellular MCs concentrations in each water body for the whole study period were recorded in the same season. In Razliv, the maximum

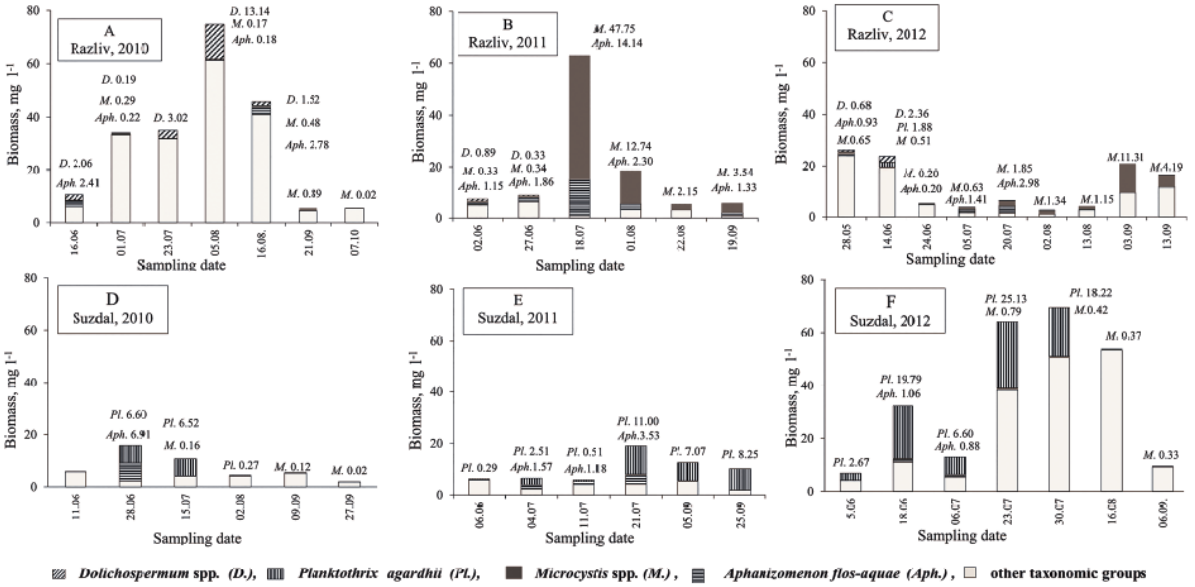

Figure 1

Seasonal changes in the total phytoplankton and cyanobacterial biomass in the studied lakes from May to October 2010-2012. A, B, C, Sestroretskij Razliv Lake in 2010, 2011 and 2012, respectively; D, E, F, Lower Suzdal Lake in 2010, 2011 and 2012, respectively

Table 1

Concentration of extracellular and intracellular cyanotoxins, three main toxin variants, and biomass of dominant species in samples from Lake Sestroretskij Razliv. MC = microcystin, dm = desmethyl, didm = double desmethyl, *denotes trace concentration (below 50 pg per injection), nd = values below LOD, DW = on a dry-weight basis

Sampling period (number of events)	Concentration of extracellular cyanotoxins in water (µg l^{-1})		Concentration of intracellular cyanotoxins in biomass (µg g^{-1} DW)		Dominant species according to the number of cells	Cyanobacterial biomass (mg l^{-1})
	Total	Three main toxin variants	Total	Three main toxin variants		
2010	seasonal median 1.61 (min. 0.11, max 41.37, N=8)		seasonal median 132 (min. 8, max 380, N=8)			
June-July (2)	0.10-0.19	dmMC-LR (0.01-0.06), MC-LR (0.03-0.15), MC-YR *	8-51	MC-RR (4-20), MC-LR (2-20)	*Dolichospermum flos-aquae*, *Aphanizomenon flos-aquae*, *Microcystis wesenbergii*, *Dolichospermum planctonica*	0.73-4.47
July-Aug. (2)	0.16-2.60	dmMC-RR (0.01-1.03), MC-LR (0.08-0.87), MC-YR (nd-0.24)	11-23	MC-RR (6), didmMC-RR (nd-6)	*Dolichospermum planctonica*	3.19-13.96
Aug.-Sept. (3)	0.61-41.37	dmMC-RR (0.46-38.54), dmMC-LR (0.05-1.65), MC-LR (0.03-1.41)	213-380	MC-RR (51-100), MC-LR (34-80), didmMC-RR (14-104)	*Dolichospermum planctonica*, *Microcystis* spp., *Aphanizomenon flos-aquae*	1.00-5.44
Oct. (1)	13.38	dmMC-RR (6.54), MC-LR (2.80), MC-RR (2.16)	255	dmMC-LR (133), MC-LR (74), dmMC-YR (15)	*Aulacoseira italica*	0.02
2011	seasonal median 0.08 (min. < 0.003, max 0.70, N=7)		seasonal median 463 (min.< 0.08, max 2095, N=7)			
June-Sept. (7)	<0.10-0.70	MC-RR (nd-0.26), MC-LR (nd-0.21), dmMC-RR (nd-0.28)	< 0.08-2095	MC-LR (nd-306), MC-YR (nd-345), dmMC-RR (nd-1245)	*Microcystis* spp., *Aphanizomenon flos-aquae*	2.19-61.94
2012	seasonal median 0.06 (min. < 0.003, max 1.24, N=9)		seasonal median 490 (min. 45, max 2294, N=8)			
May (1)	0.06	MC-LR (0.02), MC-RR (0.04)	1377	MC-RR (578), dmMC-RR (349), MC-LR (244)	*Microcystis* spp., *Aphanizomenon flos-aquae*	2.33
mid-June (1)	1.24	MC-RR (0.74), MC-LR (0.50)	1859	MC-LR (734), MC-RR (807), MC-LF (112)	*Planktothrix agardhii*	7.11
end of June (1)	0.12	MC-LR (0.07), MC-RR (0.05)	298	MC-RR (215), MC-LR (78)	*Microcystis* spp.	0.45
July (2)	<0.10-0.87	MC-LR (0.01-0.31), MC-RR (0.03-0.44), MC-YR (0.01-0.06)	260-489	MC-LR (51-116), MC-RR (186-334), MC-YR (20-36)	*Aphanizomenon flos-aquae*, *Microcystis* spp.	2.10-4.84
Aug.-Sept. (4)	<0.10-0.92	MC-LR (nd-0.40), MC-RR (nd-0.29), MC-YR (0.06-0.23)	45-2294	MC-RR (6-995), [L-Ser^{7}]MC-RR (nd-973), MC-LR (8-344)	*Microcystis* spp.	1.43-11.34

Table 2

Concentration of extracellular and intracellular cyanotoxins, three main toxin variants, and biomass of dominant species in samples from Lower Suzdal Lake. MC = microcystin, AN = anatoxin-a, dm = desmethyl, *denotes trace concentration (below 50 pg per injection), nd = values below LOD, DW = on a dry-weight basis.

Sampling period (number of events)	Concentration of extracellular cyanotoxins in water (µg l^{-1})		Concentration of intracellular cyanotoxins in biomass (µg g^{-1} DW)		Dominant species according to the number of cells	Cyanobacterial biomass (mg l^{-1})
	Total	Three main toxin variants	Total	Three main toxin variants		
2010	seasonal median 0.44 (min. 0.01, max 2.89, N=7)		seasonal median 18 (min. 1, max 234, N=7)			
mid-June (1)	0.39	[L-Ser^7]-MC-RR (0.18), MC-LR (0,14), dmMC-LR (0.05)	7	MC-LR (5), MC-RR (2)	*Aphanizomenon flos-aquae (0.5%)*	0.01
end of June-Aug. (4)	<0.10-1.68	dmMC-RR (0.01-0.36), [L-Ser^7]-MC-RR (0.01-0.32), AN (nd-0.54)	4-234	MC-RR (2-98), MC-LR (2-7), MC-WR (2-8)	*Planktothrix agardhii*, *Aphanizomenon flos-aquae*	0.30-13.57
Sept. (2)	0.56-2.89	[L-Ser^7]MC-RR (0.13-0.87), dmMC-RR (0.26-0.95), dmMC-LR (0.12-0.45)	1-18	dmMC-RR (nd-12), dmMC-YR (nd-6)	*Microcystis* spp.	0.03-0.13
2011	seasonal median 0.02 (min. 0.01, max 0.40, N=7)		seasonal median 120 (min. < 0.08, max 700, N=7)			
June-July (4)	<0.10-0.40	AN (nd-0.27), MC-RR (0.01-0.20), dmMC-RR (nd-0.14)	< 0.08-698	MC-LR (nd-225), MC-YR (nd-351), MC-LW (4-7), AN (nd-30)	*Aphanizomenon flos-aquae*, *Planktothrix agardhii*	0.43-16.72
Aug.-Sept. (3)	<0.10-0.24	AN (nd-0.24), MC-RR*, dmMC-RR*	121-142	MC-LR (28-41), MC-RR (31-32), dmMC-RR (34-35)	*Planktothrix agardhii*	7.38-8.78
2012	seasonal median 0.05 (min. < 0.003, max 0.71, N=8)		seasonal median 250 (min. 50, max 1800, N=8)			
June-July (5)	<0.10-0.70	dmMC-RR (0.05-0.60), MC-RR (nd-0.22), MC-LR (nd-0.21)	51-470	MC-RR (33-318), MC-LR (8-121), dmMC-RR (1-97)	*Planktothrix agardhii*	2.75-26.03
Aug.-Sept. (3)	<0.10	MC-RR*, MC-LR*	50-1796	MC-RR (15-1345), MC-LR (26-353), MC-YR (9-116)	*Microcystis* spp.	0.36-0.44

extracellular MCs concentration (41.37 µg l^{-1} mainly due to 38.54 µg l^{-1} of desmethyl-MC-RR) was determined in August 2010 (Table 1, Supplementary tables S2). The maximum concentration of extracellular MCs in Suzdal was noted in September 2010 and it amounted to 2.89 µg l^{-1} (Table 2, Supplementary tables S5). Concentrations of intracellular MCs were low during this sampling season in both lakes, their median values were 132 µg g^{-1} DW in Razliv and 18 µg g^{-1} DW in Suzdal (Tables 1, 2).

In 2011, in spite of higher cyanobacterial biomass and higher contribution of cyanobacteria to the phytoplankton community than in 2010 (Fig. 1 B, E), the values of median concentration of extracellular MC were low in both lakes (Tables 1 and 2). The maximum dissolved MCs concentrations recorded this season were also low in both lakes and amounted to 0.70 µg l^{-1} in Razliv (Table 1, Fig. 1B, and Supplementary Table S3) and 0.40 µg l^{-1} in Suzdal (Table 2, Fig. 1F, and Supplementary table S6). On the other hand, the intracellular MC content was high and reached 2095 µg g^{-1} in Razliv and 698 µg g^{-1} in Suzdal.

The highest median concentrations of intracellular MCs in both lakes were recorded in 2012. At the beginning of August 2012, the maximum

Table 3

The number and structure of microcystin (MC) variants identified in Sestroretskij Razliv and in Lower Suzdal Lake in 2010-2012

Year	MCs in Razliv (total variants 14)	MCs in Suzdal (total variants 9)
2010	dmMC-LR MC-LR didmMC-RR dmMC-RR MC-RR MC-YR, [L-Ser^7]MC-LR [L-MeSer^7]MC-LR MC-YA MC-HtyR MC-LF	dmMC-LR MC-LR [L-Ser^7]MC-LR dmMC-RR MC-RR [L-Ser^7]MC-RR MC-YR MC-WR
2011	dm-MC-LR MC-LR dm-MC-RR MC-RR MC-YR MC-WR MC-LW	dm-MC-LR MC-LR dm-MC-RR MC-RR dmMC-YR MC-YR MC-LW
2012	dmMC-LR MC-LR didmMC-RR dmMC-RR MC-RR [L-Ser^7]MC-RR dmMC-YR MC -YR MC-LF	MC-LR dmMC-RR MC-RR MC-YR

concentration of intracellular MCs for the entire study period (2294 µg g^{-1} DW) was recorded in Razliv. In Suzdal, peak values of intracellular MCs concentrations (1075-1796 µg g^{-1} DW) were recorded in September (Table 2, Supplementary table S7).

Neurotoxic AN was detected only in Suzdal. It was found in water samples collected from late June to early August 2010 and 2011 (Table 2, Supplementary tables S5, S6). Extracellular concentration of AN in filtered water samples was in the range of 0.16-0.54 µg l^{-1} (2 samples) in 2010 and 0.14-0.27 µg l^{-1} (3 samples: two samples in the sampling period June-July and one in August) in 2011 (Table 2, Supplementary table S5). Intracellular AN (30 µg g^{-1} DW) was recorded only in one biomass sample at the beginning of July. The presence of *P.agardhii* and *Aph.flos-aquae* was observed in all anatoxin-positive samples, *Dolichospermum* spp. was present only in a trace amount (Table 2, Supplementary table S6).

Discussion

The occurrence and distribution of cyanobacterial toxins were studied in two lakes typical for Saint Petersburg region: Sestroretskij Razliv Lake and Lower Suzdal Lake. The obtained data showed the constant presence of species able to produce cyanotoxins in both water bodies. However, the phytoplankton structure and dynamics were different between the studied lakes and seasons. In Suzdal, a higher variability of different taxonomic groups of phytoplankton was observed (Supplementary Table S1), whereas in Razliv – mainly Cyanobacteria were present.

The mass occurrence of cyanobacteria, cyanobacterial biomass values and the pattern of changes in their structure were typical for other hypertrophic or eutrophic water bodies and dam reservoirs across Europe (Teubner et al. 1999; Nixdorf et al. 2003; Pawlik-Skowrońska et al. 2004; Stefaniak et al. 2005; Dittmann & Wiegand 2006; Grabowska & Mazur-Marzec 2011; Kokocinski et al. 2011; Ostermaier et al. 2012).

Among the dominant species, the common producers of cyanotoxins were present. Therefore, we found it important to investigate the concentration of microcystins in lake water and phytoplankton samples. The dissolved (extracellular) fraction of cyanotoxins, which is released during bloom events, is sometimes overlooked in environmental studies, even though it may have a significant effect on the current state and quality of the water. Knowledge about the maximum MCs concentrations in reservoirs is of great importance, particularly in recreational areas as health outcomes may also result from exposure through swimming (contact with skin), inhalation or ingestion of cyanotoxin-containing water. Some investigations showed that even low concentrations of MCs may pose a significant health risk during the recreational use of water bodies (Ueno et al. 1996; Chen et al. 2009).

The analysis of biomass-bound (intracellular) cyanotoxins should also be conducted as this fraction of harmful biomolecules could be an additional route of their transfer and/or accumulation in aquatic animals, increasing the potential risk of human exposure through fish consumption.

MCs congeners detected in samples from Razliv and Suzdal are known to be produced by dominant cyanobacterial species present in the lakes. According to the published data, *Microcystin* spp. usually produce MC-LR, MC-RR and their desmethylated variants (Sivonen et al. 1995; Furey et al. 2008). Two to four arginine-containing MCs variants can be detected in extracts from *Dolichospermum* species, whereas mostly desmethylated variants of MC-RR or MC-LR dominate in *Planktothrix* species (Sivonen et al. 1995; Furey et al. 2008). In this study, the increased variety of microcystins congeners in hypertrophic Razliv was likely caused by higher diversity in cyanobacteria and frequent changes in dominant species. As it was observed in other studies, active growth of cyanobacteria and their diversity are promoted in hypertrophic reservoirs (Xing et al. 2007; Pawlik-Skowrońska et al. 2008; Kokocinski et al. 2011).

Some of the microcystin congeners seem to be stable components of the cyanobacterial bloom. They were detected in samples regardless of the season and the investigated water bodies. The identified MCs were mainly hydrophilic arginine-containing counterparts. The main toxin variants were MC-LR, MC-YR, MC-RR and desmethylated-MC-RR. These variants were also the most common MCs in Finland (Spoof et al. 2003), Poland (Grabowska & Mazur-Marzec 2011) and other European countries located at the same latitude (Kokocinski et al. 2011).

Concentrations of extracellular microcystins determined in natural waters are usually in the range of 0.1-10 µg l^{-1} (Sivonen & Jones 1999; Spoof et al. 2003; Welker 2008). Most of the extracellular MCs concentrations measured in our study were also within this range. Only some of the values determined in August and September 2010 in water samples from Razliv exceeded the guideline value for recreational water (WHO, 2003) and reached the maximum of 41.37 µg l^{-1} (16 Aug. 2010). The total concentrations of dissolved toxins determined in hypertrophic Razliv (Tables 1 and 2) were generally higher as compared with eutrophic Suzdal.

In our study, the concentration of cyanotoxins in bloom samples only to some extent depends on cyanobacterial biomass (Supplementary table S2). In 2010, when the contribution of cyanobacteria to the total phytoplankton biomass was low in both lakes studied (Fig. 1 A, D), the median values of the extracellular MCs concentration were higher than in the samples from the two other years (Table 1, 2). As it was proved, there are significant differences in toxin profiles and in the intensity of toxin production among cyanobacterial strains (Apeldoorn et al. 2007; Welker 2008; Kurmayer & Christiansen 2009). In spite of the fact that *P. agardhii* was reported to be a more effective producer of microcystin per dry weight than *Microcystis* spp. (Fastner et al. 1999), the concentrations of extracellular MCs were low in Suzdal in 2011 and 2012 during the observed prolonged dominance of *P. agardhii*. In September 2011, the recorded peak value of intracellular MCs concentrations in Suzdal (1075-1796 µg g^{-1} DW) could be associated with a high contribution of *M. aeruginosa* after shifting the dominance from *P. agardhii*. It was proven by chemical and genetic methods that the cyanobacterial population is complex and composed of toxic and non-toxic strains (Lyra et al. 2001; Pawlik-Skowrońska et al. 2004; Briand et al. 2008; Kurmayer&Christiansen 2009; Rohrlack et al. 2009; Bittencourt-Oliveira et al. 2010; Grabowska & Mazur-Marzec 2011). It was also noted that monitoring of different strains of *P. agardhii* is not sufficient to predict the toxicity of the *P. agardhii* bloom. Even the toxicity of a single *P. agardhii* strain is quite variable (Tonk et al. 2005). Under the influence of environmental conditions, the concentration of intracellular toxins may vary several times (Wiedner et al. 2003; van der Merwe et al. 2012; Neilan et al. 2013).

In addition to hepatotoxins, some species of freshwater cyanobacteria can produce neurotoxins. Anatoxin-a (AN) is a secondary amine with potent neurotoxicity. This cyanotoxin is produced by several cyanobacterial genera, including *Dolichospermum* (*Anabaena*), *Aphanizomenon*, *Microcystis*, *Planktothrix* (*Oscillatoria*), *Phormidium* and *Nostoc* (Lyra et al. 2001; Osswald et al. 2007; James et al. 2008; Rantala-Ylinen et al. 2011). AN occurs less frequently than MCs. In Europe, the reported values for extracellular AN concentrations ranged from 5 ng l^{-1} in Greece lakes (Dimitrakopuolos et al. 2010) to 10 µg l^{-1} in the water of an eutrophic dam reservoir in Poland during the dominance of *Dolichospermum* spp. (Pawlik-Skowrońska et al. 2004). AN was detected in Lake Veluwemeer (The Netherlands), Lake Kasumigaura (Japan) and Lake Nørre (Denmark) during the dominance of *P. agardhii* and/or *Aph. flos-aquae* (Lyra et al. 2001).

In our study, AN was detected only in Suzdal during warm periods in 2010 and 2011. Maximum concentrations of AN were recorded at the beginning of summer seasons when *P. agardhii* and *Aph. flos-aquae* dominated. *P. agardhii* and *Aph. flos-aquae* are known to be potential producers of anatoxin-a (Lyra et al. 2001; Osswald et al. 2007). It cannot be ruled out, however, that AN was produced in Suzdal by subdominant species of cyanobacteria, such as *Dolichospermum planktonica*, the trace amounts of which were always observed in AN-containing samples.

Although the maximum concentration of AN in water (0.54 µg l^{-1}) detected in mid-July in 2010 was not high, the occurrence of this "fast-death factor" in freshwater ecosystems should be monitored due to its high toxicity

Conclusion

In this work, the occurrence and distribution of cyanobacterial toxins was studied in two lakes typical for Saint Petersburg region in Russia: Lake Sestroretskij Razliv and Lower Suzdal Lake. The observed cyanobacterial assemblages: "*P. agardhii*" and "*Aph. flos-aquae/ Microcystis* spp." are typical of the hypertrophic shallow lakes in Europe.

During our study, the presence of fifteen different congeners of microcystins, mainly hydrophilic arginine-containing counterparts, and the neurotoxic anatoxin-a was determined. The increased variety of microcystins' congeners and higher MCs concentration in hypertrophic Razliv was likely caused by higher variability of phytoplankton composition and frequent changes in dominant species.

The measured concentrations of extracellular MCs in the lakes (0.1-10 µg l^{-1}) were mainly within the reported range for natural waters and only sporadically exceeded the guideline value for recreational water.

Further monitoring investigations should be conducted for several seasons to better understand the cyanobacterial response to environmental changes and to better assess the potential risk to water users in Northwestern Russia.

Acknowledgment

The authors would like to acknowledge the European Cooperation in Science and Technology, COST Action ES 1105 "CYANOCOST – Cyanobacterial blooms and toxins in water resources: Occurrence, impacts and management" for adding value to this study through networking and knowledge sharing with European experts and researchers in the field.

References

Apeldoorn, M.E.v., Egmond, H.P.v., Speijers, G.J.A. & Bakker G.J.I. (2007). Toxins of cyanobacteria. *Mol. Nutr. Food Res.* 51: 7-60.

Babanazarova, O.V., Sidelev, S.I., Aleksandrina, E.M., Sakharova, E.G. & Kurmayer, R. (2011). Phytoplankton structure and microcystine concentration in the highly eutrophic Nero Lake. *Water Resources* 38(2): 229-236. DOI: 10.1134/S0097807811020023.

Belykh, O.I., Gladkikh, A.S., Sorokovikova, E.G., Tikhonova, I.V.& Butina, T.V. (2015). Identification of toxic Cyanobacteria in Lake Baikal. *Dokl. Biochem. Biophys.* 463(1): 220-224. DOI: 10.1134/S1607672915040067.

Bittencourt-Oliveira, M.C., Santos, D.M.S. & Moura, N.A. (2010). Toxic cyanobacteria in reservoirs in northeastern Brazil: detection using a molecular method. *Braz. J. Biol*. 70(4): 1005-1010. DOI:10.1590/S1519-69842010000500012.

Briand, E., Gugger, M., Francois, J.-Ch., Bernard, C., Humbert, J.-F. et al. (2008). Temporal Variations in the Dynamics of Potentially Microcystin-Producing Strains in a Bloom-Forming *Planktothrix agardhii* (Cyanobacterium) Population. *Applied and environmental microbiology* 74(12): 3839-3848. DOI: 10.1128/AEM.02343-07.

Chen, J., Xie, P., Li, L. & Xu, J. (2009). First identification of the hepatotoxic microcystins in the serum of a chronically exposed human population together with indication of hepatocellular damage. *Toxicol. Sci*. 108(1): 81-89. DOI: 10.1093/toxsci/kfp009.

Chorus, I. (2012). *Current approaches to cyanotoxin risk assessment, risk management and regulations in different countries*. Dessau-Rosslau, Germany: Federal Environment Agency (Umweltbundesamt). Retrieved February 15, 2016, from http:// www.umweltdaten.de/publikationen/fpdf-l/4390.pdf

Directive 2006/7/EC of the European Parliament and of the Council of 15 February 2006 concerning the management of bathing water quality and repealing Directive 76/160/EEC. Official Journal of the European Union, 2006, L64:37–51. Retrieved February 15, 2016, from http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2006:064:0037:0051:EN:PDF

Dimitrakopoulos, I.K., Kaloudis, T.S., Hiskia, A.E., Thomaidis, N.S & Koupparis, M.A. (2010). Development of a fast and selective method for the sensitive determination of anatoxin-a in lake waters using liquid chromatography–tandem mass spectrometry and phenylalanine-d5 as internal standard. *Anal. Bioanal. Chem*. 397: 2245-2252. DOI: 10.1007/s00216-010-3727-3.

Dittmann, E. & Wiegand, C. (2006). Cyanobacterial toxins–occurrence, biosynthesis and impact on human affairs. *Mol. Nutr. Food Res*. 50: 7-17. DOI: 10.1002/mnfr.200500162.

Fastner, J., Neumann, U., Wirsing, B., Weckesser, J., Wiedner, C. et al. (1999). Microcystins (hepatotoxic heptapeptides) in German fresh water bodies. *Environ. Toxicol.* 14: 13-22. DOI: 10.1002/(SICI)1522-7278(199905)14:2<291::AID-TOX11>3.0.CO;2-E.

Furey, A., Allis, O., Ortea, P.M., Lehane, M. & James, K. J. (2008). Hepatotoxins: Context and Chemical Determination. In L.M. Botana (Ed.), *Seafood and Freshwater Toxins. Pharmacology, Physiology and Detection* (pp. 844-886). Second Edition. Boca Raton: CRC Press Taylor & Francis Group.

Grabowska, M. & Mazur-Marzec, H. (2011). The effect of cyanobacterial blooms in the Siemianowska Dam Reservoir on the phytoplankton structure in the Narew River. *Ocean. Hydrobiol. Stud.* 40(1): 19-26. DOI: 10.2478/s13545-011-0003-x.

Gromov, B.V., Vepritsky, A.A., Mamkaeva, K.A. & Voloshko, L.N. (1996). A survey of toxicity of cyanobacterial blooms in Lake Ladoga and adjacent water bodies. *Hydrobiologia* 322: 129-136.

James, K.J., Dauphard, J., Crowley, J. & Furey, A. (2008). Cyanobacterial Neurotoxins, Anatoxin-A and Analogues: Detection and Analysis. In L. M. Botana (Ed.), *Seafood and Freshwater Toxins. Pharmacology, Physiology and Detection* (pp. 809-822). Second Edition. Boca Raton: CRC Press Taylor & Francis Group.

Kokocinski, M., Stefaniak, K., Izydorczyk, K., Jurczak, T., Mankiewicz-Boczek, J. et al. (2011). Temporal variation in microcystin production by *Planktothrix agardhii* (Gomont) Anagnostidis and *Komárek* (Cyanobacteria, Oscillatoriales) in a temperate lake. *Ann. Limnol. - Int. J. Lim.* 47: 363-371. DOI:10.1051/limn/2011046.

Korneva, L.V., Solovieva, V.V., Zhakovskaya, Z.A., Russkikh, Ia.V. & Chernova E.N. (2014). Phytoplankton and content of cyanotoxins in Rrybinsk, Gorky and Cheboksary Reservoirs during the anomalously hot summer of 2010. *Water: Chemistry and Ecology* 8: 24-29. (In Russian with English abstract).

Kurmayer, R. & Christiansen G. (2009). The genetic basis of toxin production in Cyanobacteria. *Freshwater Reviews* 2:31-50. DOI: 10.1608/FRJ-2.1.2.

Lyra, C., Suomalainen, S., Gugger, M., Vezie, Ch., Sundman, P. et al. (2001). Molecular characterization of planktic cyanobacteria of Anabaena, Aphanizomenon, Microcystis and Planktothrix genera. *International Journal of Systematic and Evolutionary Microbiology* 51: 513-526.

Mayumi, T., Kato, H., Imanishi, S., Kawasaki, Y., Hasegawa, M. et al. (2006). Structural Characterization of Microcystins by LC/MS/MS under Ion Trap Conditions. *J. Antibiot.* 59(11): 710-719. DOI: 10.1038/ja.2006.95.

Neilan, B.A., Pearson, L.A., Muenchhoff, J., Moffitt, M.C. & Dittmann, E. (2013). Environmental conditions that influence toxin biosynthesis in cyanobacteria. *Environ. Microbiol.* 15(5): 1239-1253. DOI: 10.1111/j.1462-2920.2012.02729.x.

Nixdorf, B., Mischke, U. & Rucker, J. (2003). Phytoplankton

assemblages and steady state in deep and shallow eutrophic lakes – an approach to differentiate the habitat properties of Oscillatoriales. *Hydrobiol.* 502:111-121. DOI: 10.1023/B:HYDR.0000004274.65831.e5.

Olenina, I., Hajdu, S., Edler, L., Andersson, A., Wasmund, N. et al. (2006). *Biovolumes and size-classes of phytoplankton in the Baltic Sea.* Helsinki: HELCOM Balt.Sea Environ. (Proc. No. 106). http://www.helcom.fi/Lists/Publications/BSEP106.pdf

Osswald, J., Rellan, S., Gago, A. & Vasconcelos, V. (2007). Toxicology and detection methods of the alkaloid neurotoxin produced by cyanobacteria, anatoxin-a. Review. *Environ. Int.* 33: 1070-1089.

Ostermaier, V., Schanz, F., Köster, O. & Kurmayer, R. (2012). Stability of toxin gene proportion in red-pigmented populations of the cyanobacterium Planktothrix during 29 years of re-oligotrophication of Lake Zürich. *BMC Biology* 10: 100-116. DOI: 10.1186/1741-7007-10-100.

Pawlik-Skowrońska, B., Skowroński, T., Pirszel, J. & Adamczyk, A. (2004). Relationship between cyanobacterial bloom composition and anatoxin-a and microcystin occurrence in the eutrophic dam reservoir (SE Poland). *Pol. J. Ecol.* 52: 479-490.

Pawlik-Skowrońska, B., Pirszel, J. & Kornijów, R. (2008). Spatial and temporal variation in microcystin concentrations during perennial bloom of *Planktothrix agardhii* in a hypertrophic lake. *Ann. Limnol. Int. J. lim.* 44(2): 63-68. DOI: 10.1051/limn:2008015.

Rantala, A., Rajaniemi-Wacklin, P., Lyra, C., Lepisto, L., Rintala, J. et al. (2006). Detection of microcystins-producing cyanobacteria in Finnish lakes with genus-specific microcystins synthetase gene E (mcyE) PCR and associations with environmental factors. *Appl. Environ. Microbiol*. 72(9): 6101-6110. DOI: 10.1128/AEM.01058-06.

Rantala-Ylinen, A., Kana, S., Wang, H., Rouhiainen, L., Wahlsten, M. et al. (2011). Anatoxin-a synthetase gene cluster of the cyanobacterium *Anabaena* sp. strain 37 and molecular methods to detect potential producers. *Appl. Environ. Microbiol.* 77: 7271-7278. DOI: 10.1128/AEM.06022-11.

Rohrlack, T., Skulberg, R., Skulberg, O.M. (2009). Distribution of oligopeptides of the cyanobacterium *Planktothrix* and their persistence in selected lakes in Fennoscandia. *J. Phycol.* 45:1259-1265. DOI: 10.1111/j.1529-8817.2009.00757.x.

Russkikh, Y.V., Chernova, E.N., Voyakina, E. Ju., Nikiforov, V. A. & Zhakovskaya, Z.A. (2012). Cyanotoxin determination in natural water matrix by the method of high performance liquid chromatography- mass-spectrometry of high resolution. *Izvestiya St. Peterburgskogo gosudarstvennogo technologicheskogo instituta (tekhnicheskogo universiteta)* 17(43): 61-66. (In Russian).

Sidelev, S.I., Golokolenova, T.B., Chernova, E.N., Russkikh, I.V. (2015). Analysis of Phytoplankton in Tsimlyansk Reservoir (RUSSIA) for the Presence of Cyanobacterial Hepato and Neurotoxins. *Microbiology* 84(6): 828-837. DOI: 10.1134/S0026261715060120.

Sivonen, K., Namikoshi, M., Luukkainen, R., Fardig, M., Rouhiainen, L. et al. (1995). Variation of cyanobacterial hepatotoxins in Finland. In M. Munawar & M. Luotola (Eds.), *The Contaminats in the Nordic Ecosystem: Dynamics, Processes & Fate. Ecovision World Monograph Series* (pp. 163-169). SPB Academic Publishing, Amsterdam, The Netherlands.

Sivonen, K. & Jones, G. (1999). Cyanobacterial toxins. In I. Chorus, J. Bartram (Eds.), *Toxic cyanobacteria in water: A guide to the Public Health Consequences, Monitoring and Management* (pp. 41-111). London: E & FN Spoon.

Spoof, L., Vesterkvist, P., Lindholm, T. & Meriluoto, J. (2003). Screening for hepatotoxins, microcystins and nodularin in environmental water samples by reversed-phase liquid chromatography- electrospray ionization mass spectrometry. *J. Chromatogr. A* 1020: 105-119. DOI: 10.1016/S0021-9673(03)00428-X.

Stefaniak, K., Kokocinski, M. & Messyasz, B. (2005). Dynamics of Planktothrix agardhii (Gom.) Anagn. et Kom. blooms inpolimictic Lake Laskownickie and Grylewskie (Wielkopolska region) Poland. *Ocean. Hydrobiol. Stud.* 34 (Supl. 3): 125-136.

Teubner, K., Feyerabend, R., Henning, M., Nicklisch, A., Woitke, P. et al. (1999). Alternative blooming of Aphanizomenon flos-aquae or Planktothrix agardhii induced by the timing of the critical nitrogen: phosphorus ratio in hypertrophic riverine lakes. *Arch. Hydrobiol., Spec. Issues Advanc. Limnol.* 54: 325-344.

Tonk, L., Visser, P.M., Christiansen, G., Dittmann, E., Snelder, E.O.F.M. et al. (2005) The microcystin composition of the cyanobacterium Planktothrix agardhii changes towards a more toxic variant with increasing light intensity. *Applied and Environmental Microbiology* 71: 5177-5181. DOI:10.1128/AEM.71.9.5177–5181.2005.

Trifonova, I.S. & Pavlova, O.A. (2008). Phytoplankton succession in urban water-bodies of St. Petersburg as an indicator of their ecological conditions. *Limnol. Rev*. 8(3): 137-141.

Ueno, Y., Nagata, S., Tsutsumi, T., Hasegawa, A., Watanabe, M.F. et al. (1996). Detection of microcystin, a blue–green algal hepatotoxin, in drinking water sampled in Haimen and Fusui, endemic areas of primary liver cancer in China, by highly sensitive immunoassay. *Carcinogenesis* 17(6): 1317-1321.

van der Merwe, D., Sebbag, L., Nietfeld, J.C., Aubel, M.T., Foss, A. et al. (2012). Investigation of a *Microcystis aeruginos*a cyanobacterial freshwater harmful algal bloom associated with acute microcystin toxicosis in a dog. *J. Vet. Diagn. Invest.* 24 (4): 679-687. DOI: 10.1177/1040638712445768.

Voloshko, L., Kopecky, J., Safronova, T., Pljusch, A., Titova, N. et al. (2008). Toxins and other bioactive compounds produced by cyanobacteria in Lake Ladoga. *Estonian J. of Ecology* 57(2): 100-110. DOI: 10.3176/eco.2008.2.02.

Welker, M. (2008). Cyanobacterial Hepatotoxins: Chemistry,

Biosynthesis, and Occurrence. In L.M. Botana (Ed.), *Seafood and Freshwater Toxins. Pharmacology, Physiology and Detection* (pp. 825-844). Second Edition. Boca Raton: CRC Press Taylor & Francis Group.

Wiedner, C., Visser, P.M., Fastner, J., Metcalf, J.S., Codd, G.A. et al. (2003). Effects of light on the microcystin content of Microcystis strain PCC 7806. *Appl. Environ. Microbiol.* 69 (3): 1475-1481.

WHO. (2003). *Guidelines for safe recreational water environments. Vol. 1. Coastal and fresh waters.* Geneva: World Health Organization. Retrieved December 30, 2015, from http://www.who.int/water_sanitation_health/bathing/srwe1/en

WHO. (2011). *Guidelines for Drinking Water Quality*. Geneva: World Health Organization. Retrieved December 30, 2015, from http://www.who.int/water_sanitation_health/publications/2011/dwq_guidelines/en

Xing, W., Huang, W.M., Liu, Y.D., Li, D.H., Shen, Y.W. et al. (2007). Environmental mechanism of change in cyanobacterial species composition in the Northeastern part of Lake Dianchi (China). *Fresenius Environmental Bulletin* 16(1): 82-90.

Supplementary table S1. Abundance and biomass of the observed taxonomic groups in the studied lakes in the period of 2010-2012

S1A

Sestroretskij Razliv Lake, 2010

Date	Groups	Abundance ($\times 10^6$ cell l^{-1})	Abundance (%)	Biomass (mg l^{-1})	Biomass (%)
16-Jun-2010	Cyanoprokaryota	60.8	96	4.47	42
	Cryptophyceae	1.1	2	2.70	26
	All groups	63.3	100	10.55	100
1-Jul-2010	Cyanoprokaryota	6.4	41	0.73	2
	Cryptophyceae	1.8	12	3.73	11
	Diatomophyceae	5.0	32	9.80	29
	Chlorophyta	2.3	15	19.47	57
	All groups	15.5	100	34.13	100
23-Jul-2010	Cyanoprokaryota	8.3	43	3.19	9
	Chlorophyta	10.9	56	30.89	88
	All groups	19.4	100	35.09	100
05-Aug-2010	Cyanoprokaryota	72.2	86	13.96	19
	Chlorophyta	10.0	12	56.71	75
	All groups	83.5	100	75.26	100
16-Aug-2010	Cyanoprokaryota	62.5	67	5.44	12
	Chlorophyta	12.0	13	32.28	70
	All groups	93.7	100	46.36	100
21-Sep-2010	Cyanoprokaryota	15.0	89	1.05	19
	Cryptophyceae	1.5	3	9.05	63
	All groups	16.9	100	5.47	100
7-Oct-2010	Cyanoprokaryota	0.2	5	0.02	0.4
	Cryptophyceae	0.8	30	1.57	29
	Diatomophyceae	2.1	65	2.98	56
	All groups	3.2	100	5.34	100

S1B

Sestroretskij Razliv Lake, 2011

Date	Groups	Abundance ($\times 10^6$ cell l^{-1})	Abundance (%)	Biomass (mg l^{-1})	Biomass (%)
2-Jun-2011	Cyanoprokaryota	16.4	56	2.54	33
	Cryptophyceae	10.7	37	2.14	28
	Diatomophyceae	1.0	4	1.51	20
	All groups	29.2	100	7.61	100
27-Jun-2011	Cyanoprokaryota	15.7	84	2.55	29
	Cryptophyceae	1.6	9	2.93	33
	Chlorophyta	0.8	4	2.13	24
	All groups	18.8	100	8.91	100
18-Jul-2011	Cyanoprokaryota	793.9	99.9	61.94	99
	All groups	794.2	100	62.86	100
01-Aug-2011	Cyanoprokaryota	98.0	99	15.22	82
	All groups	99.2	100	18.54	100
22-Aug-2011	Cyanoprokaryota	33.2	89	2.19	42
	Cryptophyceae	0.2	0.6	0.66	12
	Chlorophyta	3.5	9	0.90	17
	All groups	37.5	100	5.27	100
19-Sep-2011	Cyanoprokaryota	74.4	99.6	5.83	88
	All groups	74.7	100	6.61	100

S1C

Sestroretskij Razliv Lake, 2012

Date	Groups	Abundance, *10^6 cell l^{-1}	Abundance, %	Biomass, mg l^{-1}	Biomass, %
28-May-2012	Cyanoprokaryota	19.3	66	2.33	9
	Cryptophyceae	1.9	7	5.92	23
	Diatomophyceae	6.8	23	17.14	65
	All groups	29.5	100	26.29	100
14-Jun-2012	Cyanoprokaryota	103.3	87	7.11	27
	Diatomophyceae	14.0	12	16.04	62
	All groups	117.9	100	25.99	100
24-Jun-2012	Cyanoprokaryota	4.8	65	0.45	9
	Cryptophyceae	0.2	3	0.63	12
	Diatomophyceae	2.2	30	2.86	56
	Chlorophyta	0.2	2	1.06	21
	All groups	7.5	100	5.15	100
05-Jul-2012	Cyanoprokaryota	16.5	97	2.10	56
	Cryptophyceae	0.3	2	0.92	25
	All groups	17.0	100	3.72	100
20-Jul-2012	Cyanoprokaryota	58.7	99	4.84	77
	All groups	58.9	100	6.29	100
02_Aug-2012	Cyanoprokaryota	20.8	98	1.43	56
	Diatomophyceae	0.4	2	0.71	28
	All groups	21.3	100	2.55	100
13-Aug-2012	Cyanoprokaryota	19.0	95	1.44	35
	Cryptophyceae	0.3	1	0.93	23
	Diatomophyceae	0.6	3	0.91	22
	All groups	20.0	100	4.08	100
03-Sep-2012	Cyanoprokaryota	173.3	97	11.34	54
	Cryptophyceae	1.6	1	2.75	13
	Diatomophyceae	2.5	1	5.58	27
	All groups	178.1	100	20.85	100
13-Sep-2012	Cyanoprokaryota	68.2	93	4.62	29
	Diatomophyceae	4.5	6	9.60	60
	All groups	73.0	16.09	100	100
24-Sep-2012	Cyanoprokaryota	29.6	98	1.97	61
	Diatomophyceae	0.7	2	0.88	27
	All groups	30.4	100	3.24	100

S1D

Lower Suzdal Lake, 2010

Date	Groups	Abundance ($\times 10^6$ cell l^{-1})	Abundance (%)	Biomass (mg l^{-1})	Biomass (%)
11-Jun-2010	Cyanoprokaryota	0.0	0.5	0.007	0.1
	Diatomophyceae	5.6	98	11.0	96
	All groups	5.8	100	11.43	100
28-Jun-2010	Cyanoprokaryota	171.5	99	13.57	72
	All groups	174.1	100	18.80	100
15-Jul-2010	Cyanoprokaryota	103.3	98	6.87	35
	Dinophyta	0.2	0.1	4.39	22
	Euglenophyta	1.6	2	6.90	35
	All groups	105.8	100	19.52	100
02-Aug-2010	Cyanoprokaryota	4.6	85	0.30	16
	Dinophyta	0.0	0.2	0.59	31
	Euglenophyta	0.1	2	0.50	26
	Chlorophyta	0.7	13	0.50	26
	All groups	5.4	100	1.95	100
09-Sep-2010	Cyanoprokaryota	1.9	71	0.13	2
	Dinophyta	0.0	2	3.01	51
	Euglenophyta	0.5	19	1.65	28
	Chlorophyta	0.2	7	1.05	18
	All groups	2.6	100	5.90	100
27-Sep-2010	Cyanoprokaryota	0.5	8	0.03	0.3
	Euglenophyta	1.8	31	5.48	50
	Diatomophyceae	0.5	8	1.15	10
	Chlorophyta	2.9	50	4.07	37
	All groups	5.8	100	11.01	100

S1E

Lower Suzdal Lake, 2011

Date	Groups	Abundance ($\times 10^6$ cell l^{-1})	Abundance (%)	Biomass (mg l^{-1})	Biomass (%)
06-Jun-2011	Cyanoprokaryota	6.8	81	0.43	7
	Euglenophyta	0.7	8	1.96	31
	Diatomophyceae	0.5	7	1.13	18
	Chlorophyta	0.4	4	2.06	33
	All groups	8.4	100	6.24	100
04-Jul-2011	Cyanoprokaryota	127.1	99	7.45	76
	All groups	128.9	100	9.76	100
11-Jul-2011	Cyanoprokaryota	50.5	97	1.78	30
	Dinophyta	0.01	0.1	1.15	19
	Cryptophyceae	0.5	1	0.90	15
	Chlorophyta	0.7	1.4	1.54	26
	All groups	51.9	100	5.87	100
21-Jul-2011	Cyanoprokaryota	211.7	99	16.72	80
	All groups	212.9	100	20.92	100
05-Sep-2011	Cyanoprokaryota	436.9	99	7.38	58
	Dinophyta	0.0	0.01	2.37	19
	All groups	438.6	100	12.68	100
25-Sep-2011	Cyanoprokaryota	346.6	99	8.78	82
	All groups	347.1	100	10.65	100

S1F

Lower Suzdal Lake, 2012

Date	Groups	Abundance ($\times 10^6$ cell l^{-1})	Abundance (%)	Biomass (mg l^{-1})	Biomass (%)
05-Jun-2012	Cyanoprokaryota	41.6	97	2.75	40
	Euglenophyta	0.3	1	1.20	17
	Diatomophyceae	1.0	2	1.66	24
	Chlorophyta	0.1	0.2	1.26	18
	All groups	43.1	100	6.89	100
18-Jun-2012	Cyanoprokaryota	309.7	98	21.02	65
	Diatomophyceae	6.5	2	6.92	22
	All groups	317.6	100	32.16	100
06-Jul-2012	Cyanoprokaryota	105.9	97	7.55	58
	Diatomophyceae	2.8	2.5	2.52	26
	All groups	109.5	100	13.01	100
23-Jul-2012	Cyanoprokaryota	396.6	97	26.03	41
	Dinophyta	0.1	0.03	13.14	21
	Cryptophyceae	3.4	1	9.75	15
	Diatomophyceae	5.6	1	10.93	17
	All groups	407.5	100	64.17	100
30-Jul-2012	Cyanoprokaryota	284.9	96	18.64	27
	Dinophyta	0.3	0.1	26.89	39
	Diatomophyceae	9.4	3	18.38	26
	All groups	297.4	100	69.47	100
16-Aug-2012	Cyanoprokaryota	6.8	42	0.44	1
	Dinophyta	0.3	2	27.74	51
	Cryptophyceae	7.4	45	21.99	41
	All groups	16.2	100	54.04	100
06-Sep-2012	Cyanoprokaryota	5.5	92	0.36	4
	Dinophyta	0.1	1	8.46	88
	All groups	5.9	100	9.56	100

Detected concentrations of extracellular MCs in water and intracellular MCs in biomass samples, cell number and biomass of observed cyanobacterial species in Sestroretskij Razliv Lake in 2010. * denotes to cell number or biomass <1%. Aph. = *Aphanizomenon*, M. = *Microcystis*, D. = *Dolichospermum*, P. = *Planktothrix; NA* = data not available

Sestroretskij Razliv Lake, 2010		MC-LR	dmMC-LR	didmMC-LR	[L-Ser^7]MC-LR	[L-MeSer^7]-MC-LR	MC-RR	dmMC-RR	didm-MC-RR	[L-Ser^7]-MC-RR	MC-YR	dmMC-YR	MC-LF	MC-HtyR	MC-YA	Total MCs	Cell number of cyanobacterial species, x 10^6 cells l^{-1}, (N,%)	Biomass of cyanobacterial species, mg l^{-1}, (B,%)
16 Jun	Water, µg l^{-1}	0.153	0.009		0.006		0.014						0.007			0.189	*Aph. flos-aquae* 36.8 (58 %), *D. flos-aquae* 20.8 (33%), *D. planctonica* 1.3 (2%), *D. spiroides* 1.05 (2%), *D. crassum* 0.8 (1 %), *Snowella lacustris**	*Aph. flos-aquae* 2.41 (23 %), *D. flos-aquae* 1.36 (13%), *D. planctonica* 0.29 (3%), *D. spiroides* 0.23 (2%), *D. crassum* 0.18 (2 %), *Snowella lacustris**
	Bio, µg g^{-1} dw	20			3	3	20		1	1	1					51		
01 Jul	Water, µg l^{-1}	0.028	0.055				0.003				0.025					0.111	*M. wesenbergii* 4.40 (28%), *Aph. flos-aquae* 1.00 (6%), *P. agardhii* 0.60 (4%), *D. planctonica* 0.26 (2%), *D. spiroides **, *D. crassum **	*M. wesenbergii* 0.29 (1%), *Aph. flos-aquae* 0.22 (1%), *P. agardhii**, *D. planctonicum**, *D. spiroides**
	Bio, µg g^{-1} dw	2	2				4									8		
23 Jul	Water, µg l^{-1}	0.054	0.028					0.012								0.094	*D. planctonica* 5.6 (29%), *D. spiroides* 1.05 (5%), *Aph. flos-aquae* 0.4 (2%), *Limnothrix planctonica* 0.4 (2%), *M. aeruginosa* 0.52 (3%), *M. wesenbergii* 0.36 (2%)	*D. planctonica* 2.93 (8%), *D. spiroides**, *Aph. flos-aquae**, *Limnothrix planctonica**, *M. aeruginosa**, *M. wesenbergii **
	Bio, µg g^{-1} dw	3	2				6				1					11		
05 Aug	Water, µg l^{-1}	0.874	0.238		0.053		0.158	1.032			0.241		0.003			2.599	*D. planctonica* 52.00 (62%), *Limnothrix planctonica* 9.20 (11%), *Aph. flos-aquae* 2.80 (3%), *D. crassum* 2.70 (3 %), *M. wesenbergii* 2.40 (3%), *Cuspidothrix issatschenk* 1.80 (2%), *D. spiroides* 1.10 (1%), *M. aeruginosa **	*D. planctonica* 11.49 (15%), *D. crassum* 1.49 (2%), *Limnothrix planctonica**, *Aph. flos-aquae**, *M. wesenbergii**, *Cuspidothrix issatschenk**, *D. spiroides**, *M. aeruginosa **
	Bio, µg g^{-1} dw	3	3				6		6	1	1	2		1		23		
16 Aug	Water, µg l^{-1}	0.830	0.945			0.042	0.840	38.542			0.165		0.006			41.370	*Limnothrix planctonica* 18.60 (20%), *D. planctonica* 15.70 (17%), *Aph. flos-aquae* 12.60 (13%), *D. spiroides* 7.50 (8%), *M. wesenbergii* 4.80 (5%), *M. aeruginosa* 2.50 (3%), *P. agardhii* 0.75 (1%)	*Aph. flos-aquae* 2.78 (6%), *D. planctonicum* 1.03 (2%), *Limnothrix planctonica* 0.61 (1%), *D. spiroides* 0.49 (1%), *M. wesenbergii**, *M. aeruginosa**, *P. agardhii **
	Bio, µg g^{-1} dw	80	0				100		49	8	19	3				256		
06 Sep	Water, µg l^{-1}	1.410	1.650		0.020		0.451	12.490			0.036					16.057	*NA*	*NA*
	Bio, µg g^{-1} dw	34	53	1			51		14		10	14			36	213		
21 Sep	Water, µg l^{-1}	0.030	0.050		0.055		0.017	0.459								0.611	*M. wesenbergii 7.20 (43%), M. aeruginosa 6.40 (38%), P. agardhii 0.60 (4%), Aph. flos-aquae 0.48 (3%), Planktolyngbya limnetica 0.3 (2%)*	*M. wesenbergii* 0.47 (9%), *M. aeruginosa* 0.42 (8%), *Aph. flos-aquae* 0.11 (2%), *P. agardhii**, *Planktolyngbya limnetica **
	Bio, µg g^{-1} dw	47	27				90		104		33	13		65		380		
07 Oct	Water, µg l^{-1}	2.802	1.425				2.160	6.543			0.248	0.200				13.378	*M. aeruginosa 0.08 (3%), D. spiroides 0.06 (2%), Aph. flos-aquae **	*M. aeruginosa**, *D. spiroides**, *Aph. flos-aquae**
	Bio, µg g^{-1} dw	74	133	3			14		3		14	15				255		

Supplementary Table 3

Detected concentrations of extracellular MCs in water and intracellular MCs in biomass samples, cell number and biomass of observed cyanobacterial species in Sestroretskij Razliv Lake in 2011. * denotes to cell number or biomass <1%. Aph. = *Aphanizomenon*, M. = *Microcystis*, D. = *Dolichospermum*, P. = *Planktothrix; NA* = data not available, *ND* = not detected (concentration below LOD)

Sestroretskij Razliv Lake, 2011		MC-LR	dmMC-LR	[D-Glu-OCH_3^6] MC-LR	MC-RR	dmMC-RR	MC-YR	MC-WR	MC-LW	Total MCs	Cell number of cyanobacterial species, x 10^6 cells l^{-1}, (N,%)	Biomass of observed cyanobacterial species, mg l^{-1}, (B,%)
02 Jun	Water, µg l^{-1}		0.004			0.003	0.070			0.077	*Aph. flos-aquae* 5.20 (18%), *M. aeruginosa* 4.80 (16%), *D. smithii* 1.20 (4%), *D.* sp. 1.10 (4%), *D. circinale* 0.80 (3%), *D. spiroides* 0.80 (3%), *Limnothrix planctonica* 0.80 (3%), *Aphanocapsa planctonica* 0.60 (2%), *D. crassum* 0.50 (2%), *M. wesenbergii* 0.28 (1%), *D. flos-aquae* 0.28 (1%)	*Aph. flos-aquae* 1.15 (15%), *M. aeruginosa* 0.31 (4%), *D. smithii* 0.27 (3%), *D.* spp. 0.24 (3%), *D. spiroides* 0.18 (2%), *Aphanocapsa planctonica* 0.13 (2%), *D. crassum* 0.11 (2%), *D. circinalis 0.08 (1%)*, *M. wesenbergii**, *D. flos-aquae**, *Limnothrix planctonica**
	Bio,µg g^{-1} dw									*ND*		
27 Jun	Water, µg l^{-1}									*ND*	*Aph. flos-aquae* 8.40 (45%), *M. aeruginosa* 4.20(23%), *D. spiroides* 0.98 (5%), *M. wesenbergii* 0.96 (5%), *Woronichinia naegeliana* 0.60 (3%), *D. smithii* 0.50 (3%), *D. circinale**	*Aph. flos-aquae* 1.86 (21%), *M. aeruginosa* 0.27 (3%), *D. spiroides* 0.22 (2%), *D. smithii* 0.11 (1%), *M. wesenbergii**, *Woronichinia naegeliana**, *D. circinale**
	Bio,µg g^{-1} dw									*ND*		
18 Jul	Water, µg l^{-1}	0.211			0.086	0.044				0.341	*M. aeruginosa* 576.00 (73%), *M. viridis* 96.00 (12%), *Aph. flos-aquae* 64.00 (8 %), *M. wesenbergii* 57.60 (7%), *D. planctonica**, *D. spiroides**, *Woronichinia naegeliana**	*M. aeruginosa* 37.70 (60%), *M. viridis* 6.28 (10%), *Aph. flos-aquae* 14.14 (23 %), *M. wesenbergii* 3.77 (6%), *D. planctonica**, *D. spiroides**, *Woronichinia naegeliana**
	Bio,µg g^{-1} dw	264	20	8	188	20	62	17	10	593		
01 Aug	Water, µg l^{-1}		0.008		0.022	0.018				0.048	*M. wesenbergii* 45.60 (46%), *M. aeruginosa* 40.80 (41%), *Aph. flos-aquae* 10.40 (10%), *Woronichinia naegeliana**, *D. smithii**, *D. crassum**, *D. spiroides**, *Snowella lacustris**, *D. circinalis**	*M. wesenbergii* 10.07 (54%), *M. aeruginosa* 2.67 (14%), *Aph. flos-aquae* 2.30 (12%), *Woronichinia naegeliana**, *D. smithii**, *D. crassum**, *D. spiroides**, *Snowella lacustris**, *D. circinalis**
	Bio,µg g^{-1} dw	119	7	5	84	7	29	9	2	266		
22 Aug	Water, µg l^{-1}	0.024			0.025					0.049	*M. aeruginosa* 21.60 (58%), *M. wesenbergii* 11.20 (30%), *Aph. flos-aquae* 0.20 (1%), *Woronichinia naegeliana**, *Cuspidothrix elenkinii**, *D. crassum**	*M. aeruginosa* 1.41 (29%), *M. wesenbergii* 0.73 (14%), *Aph. flos-aquae**, *Woronichinia naegeliana**, *Cuspidothrix elenkinii**, *D. crassum**
	Bio,µg g^{-1} dw	196	5	5	175	1	62	19		463		
19 Sep	Water, µg l^{-1}	0.066	0.017		0.071	0.004	0.014			0.172	*M. aeruginosa* 45.60 (61%), *Aphanocapsa planctonica* 12.00 (16%), *M. wesenbergii* 8.40 (11%), *Aph. flos-aquae* 6.00 (8%), *P. agardhii* 2.10 (3%), *D. crassum**, *M. viridis**, *D. spiroides**	*M. aeruginosa* 2.98 (45%), *Aph. flos-aquae* 1.33 (20%), *Aphanocapsa planctonica* 0.79 (12%), *M. wesenbergii* 0.55 (8%), *P. agardhii* 0.14 (2%), *D. crassum**, *M. viridis**, *D.spiroides**
	Bio,µg g^{-1} dw	159	11		335	1245	345			2095		
22 Sep	Water, µg l^{-1}	0.046	0.012		0.263	0.278	0.097			0.696	*NA*	*NA*
	Bio,µg g^{-1} dw	306	21		142	43	29			541		

Supplementary Table 4

Detected concentrations of extracellular MCs in water and intracellular MCs in biomass samples, cell number and biomass of observed cyanobacterial species in Sestroretskij Razliv Lake in 2012. * denotes to cell number or biomass <1%. Aph. = *Aphanizomenon*, M. = *Microcystis*, D. = *Dolichospermum*, P. = *Planktothrix; NA* = data not available, *ND* = not detected (concentration below LOD)

Sestroretskij Razliv Lake, 2012		MC-LR	dmMC-LR	MC-RR	didmMC-RR	dmMC-RR	[L-Ser^7]MC-RR	MC-YR	dmMC-YR	MC-LF	Total MCs	Cell number of cyanobacterial species, x 10^6 cells l^{-1}, (N,%)	Biomass of cyanobacterial species, mg l^{-1}, (B,%)
28 May	Water, µg l^{-1}	0.015		0.035							0.050	*M. aeruginosa* 8.40 (28%), *Aph. flos-aquae* 4.20 (14%), *D. circinalis* 3.80 (13%), *M. wesenbergii* 1.60 (5%), *D. spiroides* 0.41 (1%), *D. planctonica* 0.40 (1%), *Cuspidothrix elenkinii* 0.40 (1%), *Limnothrix planctonica**, *D.crassum* *	*M. aeruginosa* 0.55 (2%), *Aph. flos-aquae* 0.93(4%), *D. circinale* 0.37 (1%), *M. wesenbergii**, *D. spiroides**, *D. planctonica**, *Cuspidothrix elenkinii**, *Limnothrix planctonica**, *D. crassum**
	Bio, µg g^{-1} dw	244	71	578	16	349		81	37	1	1377		
14 Jun	Water, µg l^{-1}	0.495		0.735		0.005		0.008			1.243	*Limnothrix planctonica* 43.20 (37%), *P. agardhii* 28.80 (24%), *Planktolyngbia* spp. 8.00 (7%), *D. spiroides* 7.20 (6%), *M. aeruginosa* 4.20 (4%), *D. circinalis* 4.00 (3%), *M. wesenbergii* 3.60 (3%), *D. planctonica* 2.40 (2%), *D. crassum* 1.10 (1%), *Aph. flos-aquae* 0.80 (1%)	*Limnothrix planctonica* 1.84 (7%), *P. agardhii* 1.88 (7%), *D. planctonica* 1.26 (5%), *D. spiroides* 0.47 (2%), *D. circinalis* 0.39 (2%), *M. aeruginosa* 0.27 (1%), *Planktolyngbia* spp. 0.34 (1%), *M. wesenbergii**, *D. crassum**, *Aph. flos-aquae**
	Bio, µg g^{-1} dw	743	78	807	12	89		6	22	112	1859		
24 Jun	Water, µg l^{-1}	0.067		0.054							0.121	*M. wesenbergii* 2.80 (38%), *Aph. flos-aquae* 0.90 (12%), *D. spiroides* 0.55 (7%), *M. aeruginosa* 0.30 (4 %), *Limnothrix planctonica* 0.20 (3%), *D. viguerii* 0.06 (1%), *P. agardhii* *	*M. wesenbergii* 0.18 (4%), *Aph. flos-aquae* 0.20 (4%), *D. spiroides**, *M. aeruginosa**, *Limnothrix planctonica**, *D. viguerii**, *P. agardhii**
	Bio, µg g^{-1} dw	78		215				5			298		
5 Jul	Water, µg l^{-1}	0.014		0.030				0.009			0.053	*Aph. flos-aquae* 6.40 (38%), *M. aeruginosa* 4.80 (28%), *M. wesenbergii* 4.80 (28%), *D. spiroides* 0.43 (3%), *Cryptomonas rostrate* 0.14 (1%), *D. planctonica* *, *Synechocystis crassum**	*Aph. flos-aquae* 1.41 (38%), *M. aeruginosa* 0.31 (9%), *M. wesenbergii* 0.31 (9%), *D. spiroides**, *D. planctonica**, *Synechocystis crassum**
	Bio, µg g^{-1} dw	116		334		3		36			489		
20 Jul	Water, µg l^{-1}	0.315		0.438		0.053		0.061			0.867	*Aph. flos-aquae* 30.40 (52%), *M. wesenbergii* 13.60 (23%), *M. viridis* 10.40 (18%), *M. aeruginosa* 4.20 (7%), *P.agardhii**, *D. planctonica**, *D. spiroides**	*Aph. flos-aquae* 2.98 (47%), *M. wesenbergii* 0.89 (14%), *M. viridis* 0.68 (11%), *M. aeruginosa* 0.27 (4%), *P. agardhii**, *D. planctonica**, *D. spiroides**
	Bio, µg g^{-1} dw	51		186		3		20			260		
2 Aug	Water, µg l^{-1}	0.399		0.289				0.228			0.916	*M. viridis* 10.80 (51%), *M. wesenbergii* 6.00 (28%), *M. aeruginosa* 3.60 (17%), *Aph. flos-aquae* 0.40 (2%), *D. spiroides**, *Synechocystis crassa**	*M. viridis* 0.71 (28%), *M. wesenbergii* 0.39 (15%), *M. aeruginosa* 0.24 (9%), *Aph. flos-aquae* 0.09 (3%), *D. spiroides**
	Bio, µg g^{-1} dw	308		880		2	973	131			2294		
13 Aug	Water, µg l^{-1}										*ND*	*M. aeruginosa* 8.00 (40%), *M. wesenbergii* 8.00 (40%), *M. viridis* 1.60 (8%), *Aph. flos-aquae* 1.30 (7%), *Limnothrix planctonica**	*M. aeruginosa* 0.52 (13%), *M. wesenbergii* 0.52(13%), *Aph. flos-aquae* 0.29 (7%), *M. viridis* 0.10 (3%), *Limnothrix planctonica**
	Bio, µg g^{-1} dw	344		995		2	1	124			1466		
3 Sep	Water, µg l^{-1}							0.058			0.058	*M. viridis* 76.80 (43%), *M. wesenbergii* 57.60 (32%), *M. aeruginosa* 38.40 (22%), *Aph. flos-aquae**	*M. viridis* 5.03 (24%), *M. wesenbergii* 3.77 (18%), *M. aeruginosa* 2.51 (12%), *Aph. flos-aquae**
	Bio, µg g^{-1} dw	14	2	35		11		7	1		70		
13 Sep	Water, µg l^{-1}										*ND*	*M. wesenbergii* 43.20 (59%), *M. aeruginosa* 16.00 (22%), *M. viridis* 4.80 (7%), *Aph. flos-aquae* 4.00 (5%), *D. crassum**, *Woronichinia naegeliana**	*M. wesenbergii* 2.83 (18%), *M. aeruginosa* 1.05(7%), *M. viridis* 0.31 (2%), *Aph. flos-aquae* 0.39 (2%), *D. crassum**, *Woronichinia naegeliana**
	Bio, µg g^{-1} dw	8	7	6	1	14		3	6		45		

Supplementary Table 5

Detected concentrations of extracellular MCs in water and intracellular MCs in biomass samples, cell number and biomass of observed cyanobacterial species in Lower Suzdal Lake in 2010. * denotes to cell number or biomass <1%. Aph. = *Aphanizomenon*, M. = *Microcystis*, D. = *Dolichospermum*, P. = *Planktothrix; NA* = data not available

Lower Suzdal Lake, 2010		MC-LR	dmMC-LR	[L-Ser^7]MC-LR	MC-RR	dmMC-RR	[L-Ser^7]MC-RR	MC-YR	dmMC-YR	MC-WR	Total MCs	Anatoxin-a	Cell number of cyanobacterial species, x 10 ^{6} cells l^{-1}, (N,%)	Biomass of cyanobacterial species, mg l^{-1}, (B,%)
11 Jun	Water, µg l^{-1}	0.135	0.047	0.022		0.006	0.176				0.386		*Aph. flos-aquae* 0.03 (1%)	*Aph. flos-aquae* *
	Bio, µg g^{-1} dw	5			2						7			
28 Jun	Water, µg l^{-1}					0.007	0.007				0.014	0.160	*P. agardhii* 100.80 (58%), *Aph. flos-aquae* 70.40 (40%), *D. planctonica**, *M. aeruginosa**, *Merismopedia punctate**	*Aph. flos-aquae* 6.91 (37%), *P. agardhii* 6.60 (35%), *D. planctonica**, *M. aeruginosa**, *Merismopedia punctate**
	Bio, µg g^{-1} dw	2			2						4			
15 Jul	Water, µg l^{-1}	0.089	0.079	0.255	0.021	0.357	0.315	0.564			1.680	0.540	*P. agardhii* 99.60 (94%), *M. wesenbergii* 2.40 (3%), *D. planctonica* 0.65 (1%), *Aphanocapsa planctonica* 0.60 (1%), *Synechocystis crassa**	*P. agardhii* 6.52 (33%), *M. wesenbergii* 0.16 (1%), *D. planctonica* 0.14 (1%), *Aphanocapsa planctonica**, *Synechocystis crassa**
	Bio, µg g^{-1} dw	5	3		2	11			7	2	30			
02 Aug	Water, µg l^{-1}	0.010	0.036	0.023		0.005	0.230				0.304		*P. agardhii* 4.20 (78%), *M. wesenbergii* 0.33 (6%), *Cuspidothrix elenkini**, *D. planctonica**	*P. agardhii* 0.27 (14%), *M. wesenbergii* 0.02 (1%), *Cuspidothrix elenkini**, *D. planctonica**
	Bio, µg g^{-1} dw	3	3		6	6		1	2	8	29			
16 Aug	Water, µg l^{-1}	0.035	0.078	0.047	0.018	0.081	0.079	0.098			0.436		*NA*	*NA*
	Bio, µg g^{-1} dw	67	10		98	1		22	31	5	234			
09 Sep	Water, µg l^{-1}	0.031	0.115		0.025	0.262	0.131				0.564		*M. wesenbergii* 1.80 (68%), *Aph. flos-aquae* 0.04 (1%), *P. agardhii* 0.02 (1%)	*M. wesenbergii* 0.12 (2%), *P. agardhii**, *Aph. flos-aquae**
	Bio, µg g^{-1} dw	1									1			
27 Sep	Water, µg l^{-1}	0.049	0.454	0.123	0.199	0.954	0.874	0.089	0.143		2.885		*M. wesenbergii* 0.30 (5%), *P. agardhii* 0.15 (3%), *Aph. flos-aquae**, *D. planctonica**	*Aph. flos-aquae**, *M. wesenbergii**, *P. agardhii**, *D. planctonica**
	Bio, µg g^{-1} dw					12			6		18			

Supplementary Table 6

Detected concentrations of extracellular MCs in water and intracellular MCs in biomass samples, cell number and biomass of observed cyanobacterial species in Lower Suzdal Lake in 2011. * denotes to cell number or biomass <1%. Aph. = *Aphanizomenon*, M. = *Microcystis*, D. = *Dolichospermum*, P. = *Planktothrix; NA* = data not available

Lower Suzdal Lake, 2011		MC-LR	dmMC-LR	MC-RR	dmMC-RR	MC-YR	dmMC-YR	MC-LW	Total MCs	Anatoxin-a	Cell number of cyanobacterial species, x 10^6 cells l^{-1}, (N,%)	Biomass of cyanobacterial species, mg l^{-1}, (B,%)
6 Jun	Water, µg l^{-1}		0.006	0.008	0.002				0.016		*P. agardhii* 4.50 (54%), *M. aeruginosa* 1.38 (17%), *Aph. flos-aquae* 0.60 (7%), *M. wesenbergii* 0.15 (2%), *Aphanocapsa planctonica* 0.12 (1%)	*P. agardhii* 0.29 (5%), *M. aeruginosa* 0.09 (2%), *Aph. flos-aquae* 0.04(1%), *M. wesenbergii**, *Aphanocapsa planctonica**
	Bio, µg g^{-1} dw											
4 Jul	Water, µg l^{-1}			0.007					0.007	0.139	*Limnothrix planctonica* 67.20 (52%), *P. agardhii* 38.40 (30%), *Aph. flos-aquae* 16.00 (12 %), *Aphanocapsa incerta* 2.40 (2%), *M. wesenbergii* 1.80 (1 %), *Cuspidothrix elenkinii* 1.20 (1%), *D. planctonica**	*Limnothrix planctonica* 2.86 (29%), *P. agardhii* 2.51 (30%), *Aph. flos-aquae* 1.57 (16 %), *Aphanocapsa incerta* 0.16 (2%), *M. wesenbergii* 0.12 (1 %), *Cuspidothrix elenkinii* 0.20 (2%), *D. planctonica**
	Bio, µg g^{-1} dw			1	3				4	30		
11 Jul	Water, µg l^{-1}			0.008					0.008	0.266	*P. agardhii* 31.20 (60%), *Aph. flos-aquae* 18.00 (35%), *M. wesenbergii* 0.60 (1%), *Aphanocapsa planctonica* 0.42 (1%), *Limnothrix planctonica**, *D. planctonica**, *D. spiroides**	*P. agardhii* 0.51 (9%), *Aph. flos-aquae* 1.18 (20%), *M. wesenbergii**, *Aphanocapsa planctonica**, *Limnothrix planctonica**, *D. planctonica**, *D. spiroides**
	Bio, µg g^{-1} dw							4	4			
21 Jul	Water, µg l^{-1}	0.054	0.009	0.203	0.138				0.404		*P. agardhii* 168.00 (79%), *Aph. flos-aquae* 16.00 (8%), *Limnothrix planctonica* 16.00 (8%), *Cuspidothrix elenkinii* 6.40 (3%), *M. wesenbergii* 4.80 (2%), *D. planctonica**, *D. spiroides**	*P. agardhii* 11.00 (53%), *Aph. flos-aquae* 3.53 (17%), *Limnothrix planctonica* 0.68 (3%), *Cuspidothrix elenkinii* 1.09 (5%), *M. wesenbergii 0.31 (2%)*, *D. planctonica**, *D. spiroides**
	Bio, µg g^{-1} dw	225	17	71	24	351	3	7	698			
5 Aug	Water, µg l^{-1}	0.006		0.006					0.012	0.240	*NA*	*NA*
	Bio, µg g^{-1} dw	33	15	28	23	16	5		121			
5 Sep	Water, µg l^{-1}	0.0011	0.007	0.018	0.029				0.065		*P. agardhii* 432.00 (98.5%), *M. aeruginosa* 2.40 (1%), *M. wesenbergii**, *Aphanocapsa planctonica**, *Woronichinia naegeliana**, *Snowella lacustris**, *D. circinalis**	*P. agardhii* 7.07 (56%), *M. aeruginosa* 0.16 (1%), *M. wesenbergii* 0.10 (1%), *Aphanocapsa planctonica**, *Woronichinia naegeliana**, *Snowella lacustris**, *D. circinalis**
	Bio, µg g^{-1} dw	41	13	32	35	19			142			
29 Sep	Water, µg l^{-1}				0.070				0.007		*P. agardhii* 336.00 (97%), *Aphanocapsa planctonica* 4.80 (1%), *Woronichinia karelica* 3.90 (1%), *Aphanocapsa holsatica**, *M. aeruginosa**, *M. wesenbergii**	*P. agardhii* 8.25 (77%), *Aphanocapsa planctonica* 0.31 (3%), *Woronichinia karelica* 0.13 (1%), *Aphanocapsa holsatica**, *M. aeruginosa**, *M. wesenbergi**
	Bio, µg g^{-1} dw	28	26	31	34		2		121			

DE GRUYTER

Supplementary Table 7

Detected concentrations of extracellular MCs in water and intracellular MCs in biomass samples, cell number and biomass of observed cyanobacterial species in Lower Suzdal Lake in 2012. * denotes to cell number or biomass <1%. Aph. = *Aphanizomenon*, M. = *Microcystis*, D. = *Dolichospermum*, P. = *Planktothrix; NA* = data not available

Lower Suzdal Lake, 2012		MC-LR	MC-RR	dmMC-RR	MC-YR	Total MCs	Cell number of cyanobacterial species, x10 ^{6} cells l^{-1}, (N,%)	Biomass of cyanobacterial species, mg l^{-1}, (B,%)
05 Jun	Water, µg l^{-1}		0.002	0.045		0.047	*P. agardhii* 40.80 (95%), *M. aeruginosa* 0.60 (1%), *Aph. flos-aquae* *	*P.agardhii 2.67 (39%), M.aeruginosa*, Aph.flos-aquae**
	Bio, µg g^{-1} dw	21	33	97	10	161		
21 Jun	Water, µg l^{-1}		0.010	0.618		0.628	*P. agardhii* 302.40 (95%), *Aph. flos-aquae* 4.80 (2%), *M.aeruginosa*, M. wesenbergii**	*P.agardhii* 19.79 (62%), *Aph.flos-aquae 1.06 (3%), M.aeruginosa*, M.wesenbergii**
	Bio, µg g^{-1} dw	8	33	5	5	51		
06 Jul	Water, µg l^{-1}	0.031	0.124	0.528	0.022	0.705	*P. agardhii* 100.80 (92 %), *Aph. flos-aquae* 4.00 (4%), *M.wesenbergii* 0.60 (1%), *M. aeruginosa*, Snowella lacustris**	*P. agardhii* 6.60 (51 %), *Aph. flos-aquae* 0.88 (7%), *M.wesenbergii*, M.aeruginosa*, Snowella lacustris**
	Bio, µg g^{-1} dw	69	221	11	13	315		
23 Jul	Water, µg l^{-1}		0.010	0.048		0.058	*P. agardhii* 384.00 (94%), *M. aeruginosa* 12.00 (3%), *Cuspidothrix elenkinii*, Aph. flos-aquae*, Limnothrix planctonica*, Synechocystis crassa**	*P. agardhii* 25.13 (39%), *M. aeruginosa* 0.79 (1%), *Cuspidothrix elenkinii*, Aph. flos-aquae*, Limnothrix planctonica**
	Bio, µg g^{-1} dw	121	318	1	30	470		
30 Jul	Water, µg l^{-1}	0.208	0.218	0.099	0.020	0.545	*P. agardhii* 278.40 (94%), *M. aeruginosa* 6.40 (2%)	*P. agardhii* 18.22 (26%), *M. aeruginosa* 0.42 (1%)
	Bio, µg g^{-1} dw	46	131	2	12	191		
16 Aug	Water, µg l^{-1}	0.002				0.002	*M. aeruginosa 5.60 (35%), Snowella lacustris* 0.60 (4%), P.agardhii* 0.27 (2%), Limnothrix planctonica* 0.20 (1%), Cuspidothrix elenkinii *, Aph. flos-aquae*, Oscillatoria tenuis *, Synechocystis crassa**	*M. aeruginosa* 0.37 (1%), *Snowella lacustris*, P.agardhii*, Limnothrix planctonica*, Cuspidothrix elenkinii*, Aph. flos-aquae**
	Bio, µg g^{-1} dw	26	15		9	50		
06 Sep	Water, µg l^{-1}	0.003	0.003			0.006	*M. aeruginosa* 3.60 (61%), *M. wesenbergii* 1.40 (24%), *P. agardhii* 0.30 (5%), *Snowella lacustris* 0.12 (2%), *Aph. flos-aquae* 0.04 (1%)	*M. aeruginosa 0.24 (3%), M. wesenbergii 0.09 (1%), P.agardhii *, Snowella lacustris*, Aph. flos-aquae**
	Bio, µg g^{-1} dw	353	1345	2	96	1796		
12 Sep	Water, µg l^{-1}		0.004			0.004	*NA*	*NA*
	Bio, µg g^{-1} dw	262	695	2	116	1075		