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MICROCYSTINS AFFECT ZOOPLANKTON BIODIVERSITY IN OXBOW LAKES

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Abstract: The authors tested the hypothesis that zooplankton diversity and density are affected by the presence of cyanotoxins in the water. The authors focused on 4 oxbow lakes of the Vistula River in southern Poland, which are subjected to mass cyanobacterial development. In 2 of the oxbows (Piekary and Tyniec), microcystins released into the water were found. The highest concentration of microcystins (0.246 μg/L) was observed for microcystins LR. Zooplankton diversity showed a weak response to the presence of microcystins released into the water. The Shannon index (H') of total zooplankton diversity decreased in the Piekary and Tyniec oxbows during periods when the microcystin concentrations were highest. The same trend was noted for diversity of rotifers in both oxbows and for diversity of copepods in Piekary, but not for copepods in Tyniec. No such trends were found for the diversity of cladocerans in any of the oxbows, nor was a relationship found between density of zooplankton and microcystins. Statistical analyses showed that the number of species in individual samples was negatively correlated with the levels of sulfates, phosphates, and ammonia, but the microcystin concentration was positively related to those levels. This points to the complexity of the interactions and synergies among toxins, abiotic factors, and zooplankton biodiversity. In focusing on the problem of cyanotoxins, conservation studies should pay attention to this complexity. *Environ Toxicol Chem* 2017;36:165–174. © 2016 SETAC

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INTRODUCTION

Biodiversity is often defined as species richness, the total number of species in a specified place and time [1]. Because biodiversity is highly valued [2], it is important to understand the threats that might lower it, and because biodiversity determines the stable functioning of ecosystems [3], it needs to be maintained. Each ecosystem depends on particular trophic (interspecific) connections, with each species playing a role, so it is obvious that ecosystems begin to be weakened by a decrease in species diversity. Recent years have witnessed a growing awareness of the need to protect and conserve biodiversity, and international initiatives have been launched. One example is the Water for Life Decade (2005–2015), a project of the United Nations [4].

The main cause of decreasing water biodiversity is anthropopression (e.g., intensification of agriculture) and its effects (e.g., degradation of water ecosystems, habitat loss because of modification, and fragmentation of the landscape) [5]. One effect of anthropopression on water ecosystems is eutrophication, which may result in the growth of cyanobacterial blooms [6]. A number of factors have been cited to explain the mass development of cyanobacteria, such as nutrient concentration, water temperature, light availability, and food web structure [7]. Cyanobacterial blooms usually occur in the summer, but in areas of warm climate such as the Mediterranean they are present throughout the year [8], and in regions of temperate climate from spring to late autumn [9]. Laboratory and field studies and observations, as well as various models, have shown that cyanobacterial dominance will increase in water ecosystems with changes in nutrient load, rising temperature, enhanced vertical stratification, and increased atmospheric CO_2 [10].

Blooms modify water properties such as oxygen saturation, nutrient cycling (e.g., fixing of atmospheric nitrogen), and the presence of cyanobacterial toxins [11]. The different types of cyanotoxins are grouped according to the systems or cells affected, as follows: neurotoxins (anatoxin-a, homoanatoxin-a), hepatotoxins, cytotoxins (cylindrospermopsin), irritants, and gastrointestinal toxins [12]. The most frequent cyanobacterial toxins are those belonging to the microcystin and nodularin families [13]. The occurrence of microcystins has been shown in cyanobacteria such as *Microcystis* (*Microcystis aeruginosa*, *Microcystis wesenbergii*, *Microcystis viridis*), *Anabaena (Dolichospermum) flos-aqaue*, *Nostoc*, *Planktothrix agardhii*, *Planktothrix rubescens*, *Oscillatoria tenuis*, *Anabaenopsis*, *Haphalosiphon hibernicus*, and *Aphanocapsa cumulus* [13]. These are harmful to water organisms [14,15] and especially planktonic ones, affecting their growth and reproduction [16]. In freshwater ecosystems, microcystins are quite often present during bloom episodes. These toxins can affect all zooplankton groups, including rotifers, copepods, and cladocerans (e.g., *Daphnia hyalina*, *Daphnia longispina*, *Daphnia pulicania*, *Daphnia pulex*) [17]. The groups of planktonic animals differ in their response to cyanobacterial toxins. For example, copepods and rotifers are more resistant than cladocerans, and smaller cladocerans (*Bosmina*) are more resistant than larger ones (*Daphnia*) [18].

In the present study we sought to determine whether and how zooplankton diversity and density are modified when exposed to toxins released from cyanobacterial blooms in water bodies. We focused on oxbow lakes, which are eutrophic water bodies potentially subject to the growth of cyanobacterial blooms. Oxbows are important reservoirs that support riverine systems, creating heterogeneous habitats [19,20], and they provide valuable services to human societies [21], but they are also

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among the most threatened ecosystems on Earth [22]. They are important habitats for many organisms—some of them are Natura 2000 habitats—and are used for recreation. The oxbows we studied are parts of old beds of the Vistula River, the largest river in Poland.

Knowledge of microorganism biodiversity in oxbow lakes, and an understanding of the threats to it, are fundamental to future management and conservation of these valuable ecosystems.

MATERIALS AND METHODS

We collected water samples from 4 oxbow lakes of the Vistula River system (Figure 1) in southern Poland, situated close to each other near the city of Krakow. The samples were collected during the 2014 vegetation season (May–October: spring–autumn, when cyanobacterial blooms are expected) from the deepest part of each lake. All sampling sites were in open water (without macrophytes), and are comparable. Samples were collected every month before cyanobacteria bloom growth started, and every week during bloom growth. Physicochemical and biological parameters were assessed. Water temperature, dissolved oxygen, oxygen saturation, pH, conductivity, and chlorophyll *a* concentration were measured in situ with a YSI 6600 V2 Multiparameter Sonde at 1 m depth, and in water near the bottom. Depth and transparency (with a Secchi disc) were also measured. The concentrations of ions (NO_2^-, NO_3^-, PO_4^{3-}, SO_4^{2-}, HCO_3^-, NH_4^+) and microcystins were measured in samples that were collected from the same points and depths and immediately transported to the laboratory. Ion concentrations were determined with a Dionex ion chromatograph at the laboratory of the Institute of Nature Conservation, Polish Academy of Sciences. Microcystin concentrations were analyzed using a high-performance liquid chromatography (HPLC) Agilent 1100 with a diode matrix (DAD) at the Central Laboratory of the Municipal Water and Sewage Company (Krakow, Poland) [23]. Microcystins were analyzed as microcystin-LR (MC-LR), microcystin-RR (MC-RR), and microcystin-YR (MC-YR). In total, 72 water samples were taken for analyses of microcystin concentrations and physicochemical parameters.

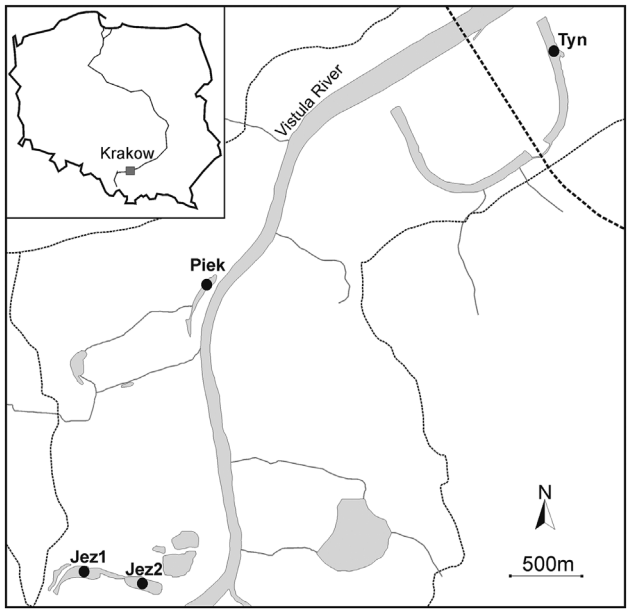

Figure 1. Locations in Poland of the oxbow lakes studied. Piek = Piekary oxbow lake, 50°00′50.1″N, 19°47′35.7″E, maximum depth 4.0 m, area 1.56 ha; Tyn = Tyniec oxbow lake, 50°01′47.0″N, 19°49′39.8″E, maximum depth 3.0 m, area 5.75 ha; Jez1 = Jeziorzany 1, 49°59′46.0″N, 19°46′52.5″E, maximum depth 2.4 m, area 2.21ha; Jez2 = Jeziorzany 2, 49°59'43.7″N, 19°47′10.6″E, maximum depth 5.5 m, area 2.19 ha.

Samples for assessment of biological parameters (36 for cyanobacteria composition and 36 for zooplankton) were taken from 1 m depth in the lakes. For zooplankton analysis, the samples were concentrated from 10 L water with a plankton net (mesh size 50 μm) and fixed in 4% formaldehyde. The zooplankton samples were analyzed under a Nikon H550L light microscope (40–400×), and was recounted per L, in a 0.5-mL chamber, with mean density based on 5 counts. The zooplankton species were identified using keys [24–29].

Samples for cyanobacterial structure analysis were concentrated from 10 L water with a plankton net (mesh size 10 μm) and fixed with Lugol's solution. Cyanobacteria were identified under a Nikon H550L light microscope (40–400×) using keys [30–32]. The biomass of cyanobacteria was calculated as biovolume by comparing specimens with their geometrical shapes [33]. Additional cyanobacteria samples were taken and not fixed; they were transported immediately to the laboratory for identification of fresh material.

Statistical analyses

The Shannon (H') index was calculated on the basis of density as a measure of zooplankton diversity. Spearman rank correlation tests in Statistica 12 (Statsoft) were used to identify correlations between microcystin concentration and zooplankton density and species diversity. Canonical correlation analyses in Canoco 5 (licence number c509112) were performed to examine relationships between abiotic (physicochemical) parameters and biotic variables (microcystin concentration, number of zooplankton species in sample). All data were log-transformed ($x + 1$).

RESULTS

Physicochemical parameters

Water pH ranged from slightly acidic to slightly alkaline in the Piekary and Tyniec oxbow lakes, and from neutral to slightly alkaline in Jeziorzany 1 and Jeziorzany 2. These lakes differed in some environmental parameters (Table 1). The water of the Tyniec oxbow showed considerably higher conductivity and Cl^- and SO_4^{2-}concentrations than the other oxbows, and the highest mean concentrations of HCO_3^-, NO_3^-, and PO_4^{3-}. The water of Jeziorzany 1 showed the lowest $PO_4^{3-}$and NH_4^+ concentrations, and the water of Jeziorzany 2 the highest variation of $PO_4^{3-}$and NH_4^+ (Table 2). The Piekary oxbow lake showed the lowest mean conductivity and lowest $SO_4^{2-}$and HCO_3^- concentrations but high NH_4^+ and the highest variation of conductivity and NO_3^-.

Blooms and microcystins

We found cyanobacteria in all sampled oxbow lakes (Table 2). In 2 oxbow lakes (Tyniec and Piekary) there were persistent blooms formed by species potentially able to produce microcystins. Blooms were present in the Tyniec oxbow lake from August to October, formed by *Microcystis ichthyoblabe* (G. Kunze) Kützing, *M. wesenbergii* (Komárek) Komárek ex Komárek in Joosen, and *Woronichinia naegeliana* (Unger) Elenkin (Table 2). In the Piekary oxbow, blooms were present in August, at the beginning of September, and in October, created by *Dolichospermum planctonicum*

Table 1. Values of physicochemical parameters in the water of Piekary ($n = 22$), Tyniec ($n = 26$), Jeziorzany 1 ($n = 12$), and Jeziorzany 2 ($n = 12$) oxbows

Parameter		Oxbow			
		Piekary	Tyniec	Jeziorzany 1	Jeziorzany 2
Temperature [°C]	Range (mean)	8.6–24.3 (15.6)	9.3–24.7 (16.9)	12.7–23.3 (19.0)	10.1–25.0 (17.1)
	CV	26	25	20	31
pH	Range (mean)	5.5–8.3 (6.7)	6.4–8.3 (7.2)	7.1–7.7 (7.4)	7.0–8.1 (7.5)
	CV	8	7	3	5
Dissolved oxygen [mg/L]	Range (mean)	0.3–9.2(5.0)	0.2–14.3 (5.4)	0.3–9.7 (4.2)	1.5–11.2 (6.1)
	CV	55	68	67	51
Oxygen saturation [%]	Range (mean)	2.6–100.8 (52.5)	2.2–169.6 (57.8)	2.4–94.6 (45.1)	14.2–115.2 (65.1)
	CV	53	74	62	55
Conductivity [μS/cm]	Range (mean)	481–1011 (668)	1259–1361 (1299)	748–1023 (802)	682–910 (734)
	CV	18	2	11	10
Cl^- [mg/L]	Range (mean)	52.9–155.4 (93.5)	250.8–284.8 (274.4)	90.1–140.5 (99.7)	90.0–97.1 (93.4)
	CV	26	3	13	3
SO_4^{2-} [mg/L]	Range (mean)	17.7–78.1 (35.0)	75.9–100.1 (84.7)	43.3–71.6 (53.5)	39.9–64.2 (48.9)
	CV	36	6	15	16
HCO_3^- [mg/L]	Range (mean)	196–270 (244)	225–327 (284)	229–316 (280)	203–327 (263)
	CV	7	11	10	14
NO_3^- [mg/L]	Range (mean)	0.1–3.5 (0.5)	nd–4.1 (0.6)	0.1–1.3 (0.5)	nd–1.2 (0.4)
	CV	148	117	67	107
NO_2^- [mg/L]	Range (mean)	nd–0.220 (0.022)	nd–0.019 (0.001)	nd–0.017 (0.003)	nd–0.015 (0.002)
	CV	250	357	233	237
NH_4^+ [mg/L]	Range (mean)	0.025–1.96 (0.38)	0.029–1.21 (0.25)	0.005–0.30 (0.13)	0.004–1.98 (0.35)
	CV	114	108	102	169
PO_4^{3-} [mg/L]	Range (mean)	nd–0.19 (0.060)	nd–0.52 (0.130)	nd–0.03 (0.007)	nd–0.43 (0.060)
	CV	85	114	161	201
Chlorophyll *a* [μg/L]	Range (mean)	nd–94.4 (23.3)	nd–140.0 (29.7)	nd–87.9 (19.7)	6.2–103.3 (28.6)
	CV	102	103	137	118

Abbreviations: nd = undetectable level; CV = coefficient of variation.

(Brunnthaler) Wacklin et al., *Dolichospermum spiroides* (Klebahn) Wacklin et al., *Microcystis wesenbergii*, and *Oscillatoria tenuis* C. Agardh ex Gomont. In Jeziorzany 1, *M. wesenbergii* developed for a short period during September. In Jeziorzany 2, cyanobacteria (*W. naegeliana*, *Cuspidothrix issatschenkoi* (Usachev) P. Rajaniemi, J. Komárek, R. Willame, P. Hrouzek, K. Kastovská, L. Hoffmann, and K. Sivonen, *Anabaena* spp.) occurred at higher concentrations in September and October, but those species do not produce microcystins. Microcystins were present in the Tyniec and Piekary oxbows but were not found in the water of Jeziorzany 1 and 2 (Table 3). The highest microcystin concentration was found for MC-LR at 1 m depth in the Tyniec and Piekary oxbows. We noted the presence of MC-LR at the beginning of September (Tyniec) and the beginning of October (Piekary). Both MC-RR and MC-YR were found at 1 m depth and in water close to the bottom in both oxbows, mostly at the end of the bloom period.

Diversity and density of zooplankton

In the sampled oxbow lakes we found 42 zooplankton species in total: 19 rotifer species, 9 copepod species, and 14 cladoceran species (Table 4). The number of zooplankton species was highest in the Tyniec oxbow (35 species noted during the whole sampling period) and lowest in Jeziorzany 1 (26 species for the whole period). Tyniec had the highest number of cladoceran and copepod species but the lowest number of rotifer species. The water bodies differed mostly in the number of cladoceran and copepod species; their numbers of rotifer species were similar.

The mean total density of zooplankton, rotifers, and cladocerans was highest in the Piekary oxbow and lowest in Jeziorzany 2 (Table 5). Jeziorzany 1 had the highest mean density of copepods, and Tyniec the lowest. Jeziorzany 1 showed the highest variation of total zooplankton, rotifer, and copepod density, and the lowest variation of copepod density. Piekary had the highest variation of cladoceran density and the lowest variation of total zooplankton, rotifer, and copepod density.

In terms of particular groups, rotifers were most diverse in the 2 Jeziorzany oxbow lakes (Figure 2), but the densest (7565 Ind/L; Table 5) in the Piekary oxbow. Cladocerans were densest (1327 Ind/L) but least diverse in the Piekary oxbow (Figure 3). Copepod density was highest (839 Ind/L) in Jeziorzany 1, but diversity was highest in the Tyniec oxbow lake (Figure 4).

The Shannon index (H') of total zooplankton diversity decreased in the Piekary and Tyniec oxbows during periods when the microcystin concentrations were highest (Table 6). The same trend was noted for diversity of rotifers in both oxbows and for diversity of copepods in Piekary, but not for copepods in Tyniec. We found no such trends for the diversity of cladocerans in any of the oxbows.

Statistical analyses

In analyses of the relationships between toxin concentrations in oxbow lakes water and zooplankton density and diversity, Spearman rank correlations revealed only 1 statistically significant correlation (negative): between the total number of zooplankton species at 1-m depth and the microcystin concentration at the same depth ($p < 0.05$; $r = -0.343$). Canonical correlation analyses confirmed that correlation (Figure 5).

The number of zooplankton species was highest (19–20 species) in individual samples from Jeziorzany 1 and 2, where cyanobacteria blooms developed during only a short period. A single sample from the Piekary oxbow also contained 19 species; samples from the Tyniec oxbow, the lake with the longest period of cyanobacterial blooming, had fewer species (maximum 17–18 species per sample) than the other water

Table 2. Chlorophyll *a* concentration (Chl *a*; μg/L), cyanobacteria species, and biomass (mg/L) present in oxbow lakes during 2014

		Oxbow			
Month		Piekary	Tyniec	Jeziorzany 1	Jeziorzany 2
May	Species	cno	cno	cno	cno
	Biomass	0.0	0.0	0.0	0.0
	Chl *a*	16.8	40.3	35.7	24.2
June	Species	*Oscillatoria tenuis*	cno	cno	cno
	Biomass	0.8	0.0	0.0	0.0
	Chl *a*	27.4	17.4	3.1	78.2
July	Species	*Dolichospermum spiroides*	cno	cno	cno
	Biomass	0.4	0.0	0.0	0.00
	Chl *a*	Data not available	27.4	5.2	Data not available
August	Species	*D. planctonicum*, *D. spiroides*, *Microcystis wesenbergii*, *O. tenuis*	*Aphanocapsa* sp. *Microcystis ichthyoblabe*, Woronichinia naegeliana	cno	*Dolichospermum planctonicum*, *Aphanizomenon* sp.
	Biomass	3.5	66.9	0.0	0.2
	Chl *a*	94.4	29.2	39.7	10.7
August	Species	Samples not taken	*M. ichthyoblabe*	Samples not taken	Samples not taken
	Biomass		16.4		
	Chl *a*		28.6		
August	Species	Samples not taken	*Aphanizomenon* sp. *M. ichthyoblabe*, *W. naegeliana*	Samples not taken	Samples not taken
	Biomass		128.3		
	Chl *a*		54.5		
September	Species	*O. tenuis*	*M. ichthyoblabe*, *M. wesenbergii*, *W. naegeliana*	*M. wesenbergii*	*D. flos-aquae*, *D. planctonicum*, *D. spiroides*, *Cuspidothrix issatschenkoi*, *Woronichinia naegeliana*
	Biomass	2.7	53.3	0.5	7.4
	Chl *a*	75.6	140.0	24.8	15.3
September	Species	*D. planctonicum*, *O. tenuis*	*M. ichthyoblabe*, *M. wesenbergii*	Samples not taken	Samples not taken
	Biomass	0.6	42.6		
	Chl *a*	21.3	38.4		
September	Species	*O. tenuis*	*Aphanizomenon* sp., *M. ichthyoblabe*, *M. wesenbergii*	Samples not taken	Samples not taken
	Biomass	0.5	60.4		
	Chl *a*	17.8	26.1		
October	Species	*O. tenuis*	*M. ichthyoblabe*, *M. wesenbergii*	cno	*Anabaena flos-aquae*, *A. planctonica*, *Aphanizomenon* cf. *issatschenkoi*
	Biomass	0.4	48.3	0.0	2.1
	Chl *a*	19.3	41.8	18.7	9.6
October	Species	*O. tenuis*	*M. ichthyoblabe*, *M. wesenbergii* *W. naegeliana*	Samples not taken	Samples not taken
	Biomass	2.0	39.3		
	Chl *a*	23.4	17.2		
October	Species	*O. tenuis*	*M. ichthyoblabe*, *M. wesenbergii*, *W. naegeliana*	Samples not taken	Samples not taken
	Biomass	0.8	93.1		
	Chl *a*	14.4	12.4		
October	Species	cno	*W. naegeliana*	Samples not taken	Samples not taken
	Biomass	0.0	55.5		
	Chl *a*	3.7	11.0		

cno = cyanobacteria not observed.

Table 3. Microcystins concentration (μg/L) in the oxbow lakes studied[a]

	8 September				6 October				14 October				28 October				
Microcystins	RR	YR	LR	MCtot	RR	YR	LR	MCtot	RR	YR	LR	MCtot	RR	YR	LR	MCtot	MC tot
Piekary 1 m	nd	nd	nd	nd	nd	nd	0.205	0.205	0.057	nd	0.081	0.138	0.035	0.018	0.027	0.080	0.423
Piekary bottom	nd	nd	nd	nd	nd	nd	nd	nd	0.082	0.029	0.018	0.129	0.041	0.014	nd	0.055	0.184
Tyniec 1 m	nd	nd	0.246	0.246	nd	nd	nd	nd	nd	nd	nd	nd	0.041	0.019	nd	0.060	0.306
Tyniec bottom	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	0.045	0.032	nd	0.077	0.077

[a]0 values not presented.
nd = not detected.

Table 4. Structure of zooplankton species in oxbows

Species	Piekary	Tyniec	Jeziorzany 1	Jeziorzany 2
Rotifera				
Asplanchna priodonta (Gosse, 1850)	+	+	+	+
Brachionus angularis (Gosse, 1858)	+	+	+	+
Brachionus calyciflorus (Pallas, 1766)	+	+	+	+
Brachionus diversicornis (Daday, 1883)	+	+	+	+
Brachionus urceolaris (Mueller, 1773)	+		+	
Filinia longiseta (Ehrenberg, 1834)	+	+	+	+
Gastropus minor (Rousselet, 1892)	+		+	+
Kellicotia longispina (Kellicott, 1879)	+	+	+	+
Keratella cochlearis (Gosse, 1851)	+	+	+	+
Keratella tecta (Gosse, 1851)	+	+	+	+
Keratella quadrata (Mueller, 1786)	+	+	+	+
Lecane stenroosi (Meissner, 1908)				+
Polyarthra major (Burckhardt, 1900)	+	+	+	+
Polyarthra minor (Voigt, 1904)	+		+	+
Polyarthra remata (Skorikov, 1896)	+	+		+
Polyarthra vulgaris (Carlin, 1943)	+	+	+	+
Pompholyx sulcata (Hudson, 1885)	+	+	+	+
Trichocerca capucina (Wierzejski and Zacharias, 1893)	+	+	+	+
Trichocerca similis (Wierzejski, 1893)		+		
Total *Rotatoria* number	17	15	16	17
Cladocera				
Alona rectangula (Sars, 1862)			+	
Bosmina longirostris (Mueller, 1785)	+	+	+	+
Ceriodaphnia pulchella (Sars, 1862)				+
Chydorus sphaericus (Mueller, 1776)	+	+		
Daphnia ambigua (Scourfield, 1947)		+	+	+
Daphnia cucullata (Sars, 1862)	+	+	+	+
Daphnia galeata (Sars, 1864)		+		
Daphnia longispina (Mueller, 1776)		+	+	+
Diaphanosoma brachyurum (Lievin, 1848)	+	+	+	+
Eubosmina coregoni (Baird, 1857)		+		+
Eubosmina gibbera (Schoedler, 1863)	+	+		
Eubosmina longispina (Leydig, 1860)	+	+	+	+
Leptodora kindtii (Focke, 1844)	+	+		
Moina micrura (Kurz, 1875)	+	+	+	+
Total *Cladocera* number	8	12	8	9
Copepoda				
Acanthocyclops venustus (Norman and Scott, 1906)	+	+		
Cyclops abyssorum (Sars, 1863)		+		
Cyclops strenuus (Fischer, 1851)		+		
Cyclops vicinus (Uljanin, 1875)	+	+		
Eudiaptomus gracilis (Sars, 1863)	+	+	+	+
Eurytemora affinis (Poppe, 1880)	+			
Mesocyclops leuckarti (Claus, 1857)		+		
Metacyclops gracilis (Lilljeborg, 1853)		+		
Thermocyclops crassus (Fischer, 1853)	+	+	+	+
Total *Copepoda* number	5	8	2	2
Total species number	30	35	26	28

bodies. For samples with fewer species, the relation to microcystin concentration was not significant.

The number of species in samples was negatively correlated with the concentrations of sulfates, phosphates, and ammonia, and the microcystin concentration was related to the concentrations of those ions.

DISCUSSION

Biodiversity is directly linked to the health of an ecosystem. Healthy water ecosystems provide valuable ecosystem services to humans [21]. The diversity of microorganisms in freshwater ecosystems is underestimated, because the emphasis generally

Table 5. Density (Ind./L) of total zooplankton and particular groups

	Piekary			Tyniec			Jeziorzany 1			Jeziorzany 2		
	Range	Mean	CV	Range	Mean	CV	Range	Mean	CV	Range	Mean	CV
Zooplankton total	766–8230	3944.1	53.05	371–3326	1774.8	59.20	1373–7843	3304.5	72.66	239–2618	1446.5	66.40
Rotifera	504–7565	3304.7	64.80	61–2983	1408.1	70.60	392–7192	2682.0	90.50	213–2307	1086.3	81.50
Cladocera	31–1327	237.7	162.60	24–233	122.7	61.10	6–633	194.8	131.74	12–151	80.6	59.30
Copepoda	161–670	401.7	43.20	32–533	244.2	70.10	26–839	427.7	73.40	14–405	279.2	52.90

CV = coefficient of variation.

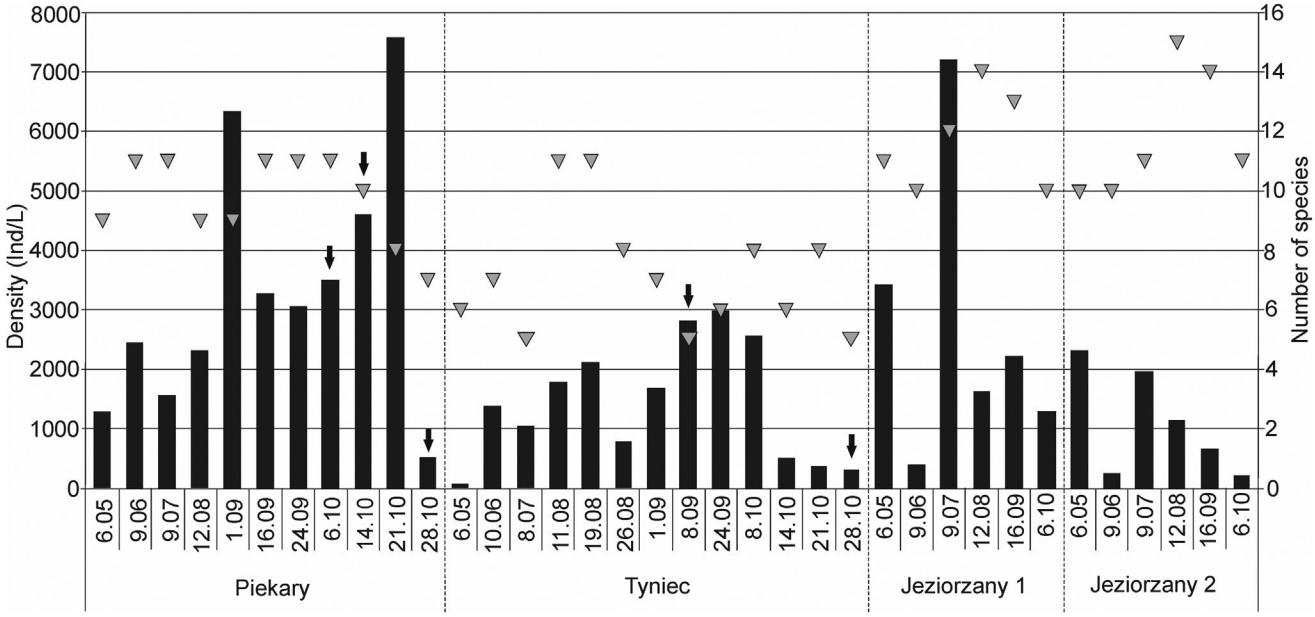

Figure 2. Diversity and density of rotifers in the oxbows studied. Bars represent density, triangles represent number of species, and arrows indicate the presence of microcystins in the water.

is on macroorganisms [21]. For holistic management, and to maintain water ecosystems in good health, we need studies on microorganism diversity and on the mechanisms by which that diversity is altered in freshwater systems. The diversity of microorganisms is driven by abiotic and biotic factors. Among the latter are toxins produced by cyanobacteria, and our understanding of their effects on microorganisms is poor.

The most frequent cyanotoxins are microcystins, which are found in most populations of *Microcystis* spp., *Anabaena*, *Nostoc*, *Planktothrix*, *Oscillatoria*, and *Anabaenopsis* [13]. Microcystins are secondary metabolites that are toxic to aquatic organisms, including protozoa, because they bind to and inhibit protein phosphatases 1 and 2A [34 and literature cited therein]. They are intracellular toxins, released into the water when the cells lyse naturally or are broken [12,17]. Once released into the water, microcystins can remain for a relatively long time before being eliminated through biodegradation or photolysis [35]. In support of that observation, we found the highest microcystin concentrations close to the end of the cyanobacterial bloom, when cyanotoxins were present in the upper layer of water and in the layer close to bottom sediment.

The presence of cyanotoxins was not significantly linked to the biomass of cyanobacteria, which was higher in the Tyniec oxbow than in Piekary. In Tyniec, the microcystin concentration was lower and its presence was of shorter duration.

In the Tyniec oxbow, only non-nitrogen–fixing cyanobacteria created blooms. In Piekary, there were nitrogen-fixing (but in smaller biomass) as well as non-nitrogen–fixing cyanobacteria.

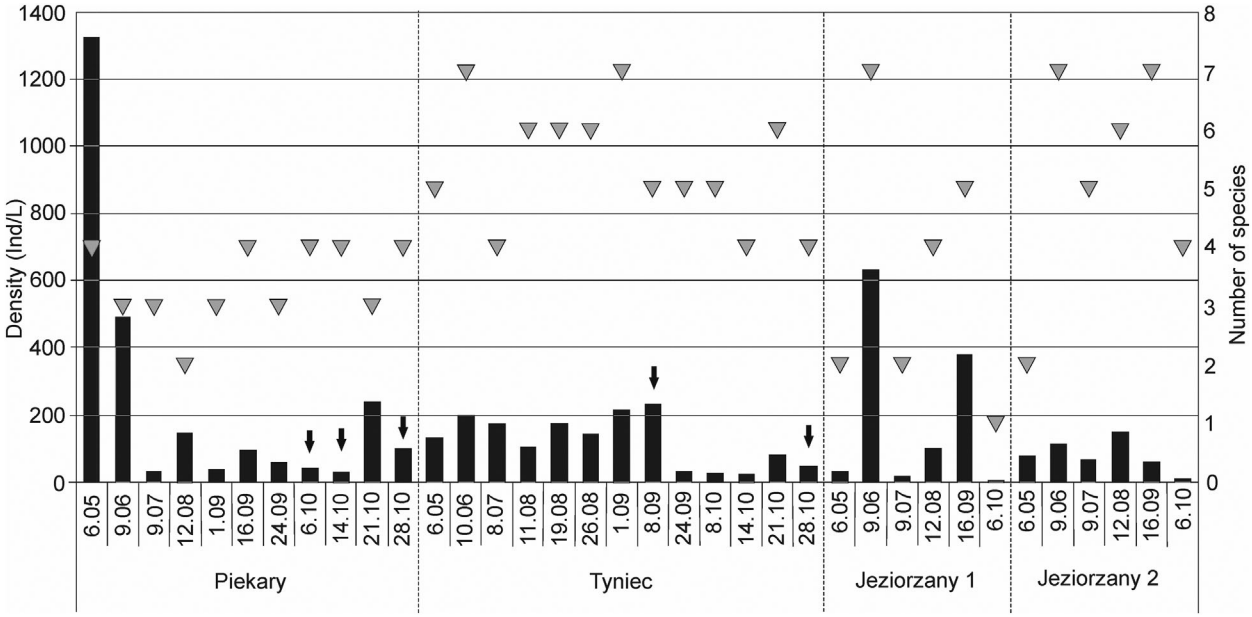

Figure 3. Diversity and density of cladocerans in the oxbows studied. Bars represent density, triangles represent number of species, and arrows indicate the presence of microcystins in the water.

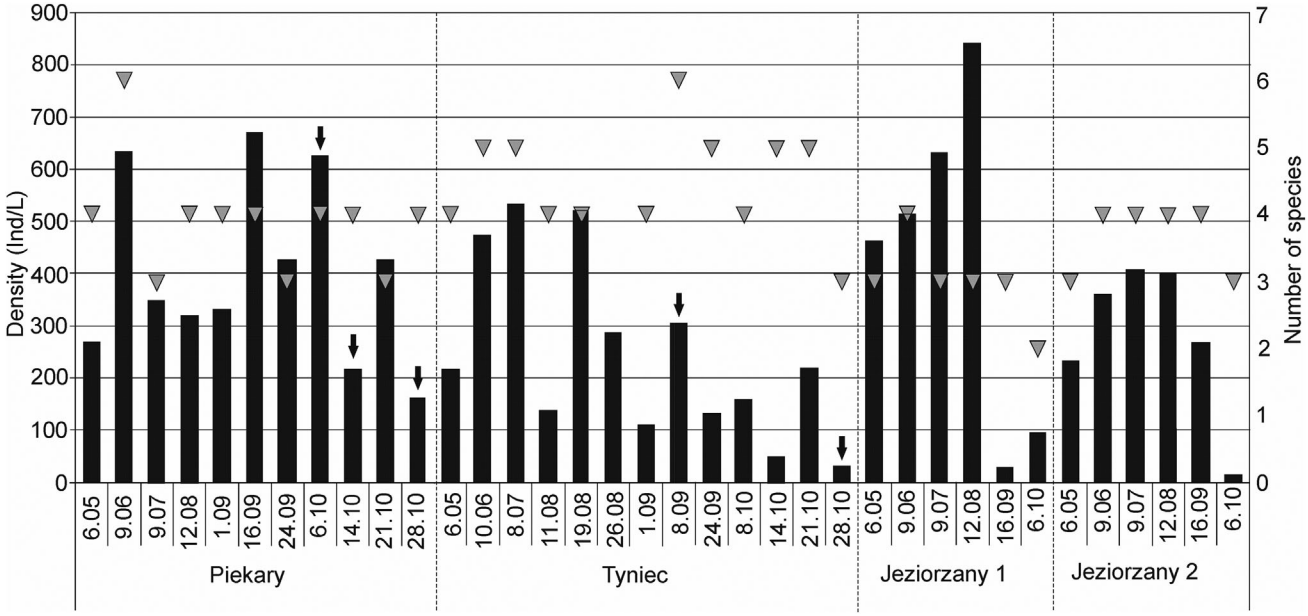

Figure 4. Diversity and density of copepods in the oxbows studied. Bars represent density, triangles represent number of species, and arrows indicate the presence of microcystins in the water.

Non-nitrogen–fixing species are favored by the presence of ammonium nitrogen, while the development of species fixing molecular nitrogen is promoted under conditions of nitrogen scarcity [7,36]. The Tyniec oxbow showed the highest mean concentrations of NO_3^- and $PO_4^{3-}$and a high concentration of NH_4^+. The higher concentration and longer duration of microcystins in Piekary, together with the presence of nitrogen-fixing cyanobacteria, support the suggestion that these toxins play an ecological role in improving nutrient availability [37]. In that oxbow we found the highest variation of conductivity and NO_3^- concentrations, and low mean PO_4^{3-}. It has been suggested that the production of toxins by cyanobacteria is not for defense against predators but for important biological functions such as cell signaling, nutrient uptake, and iron scavenging [37].

Table 6. Shannon Index (H') of total zooplankton and its particular groups in oxbows with microcystin presence

	Rotifera	Cladocera	Copepoda	Total zooplankton
Piekary oxbow				
May	1.17	0.14	0.90	1.61
June	1.69	0.26	0.98	2.10
July	1.42	0.88	0.81	1.86
August	1.85	0.40	1.00	2.23
September	0.90	0.74	0.98	1.14
September	1.30	1.04	1.00	1.80
September	1.04	0.71	0.44	1.42
October[a]	0.81[a]	0.85[a]	0.77[a]	1.28[a]
October[a]	0.61[a]	1.29[a]	0.83[a]	0.85[a]
October	0.63	0.75	0.46	0.96
October[a]	0.82[a]	1.06[a]	0.38[a]	1.63[a]
Tyniec oxbow				
May	1.24	1.09	1.07	2.09
June	0.75	0.77	0.84	1.66
July	1.18	1.02	1.07	2.03
August	1.39	1.52	1.14	1.83
August	1.29	0.74	0.73	1.86
August	1.37	0.94	1.34	2.18
September	1.14	1.09	1.25	1.69
September[a]	1.24[a]	1.30[a]	1.12[a]	1.79[a]
September	0.67	1.23	1.29	0.94
October	0.74	1.27	0.80	1.02
October	0.73	1.24	1.40	1.26
October	1.32	1.24	1.20	2.22
October[a]	1.06[a]	1.26[a]	0.88[a]	1.72[a]

[a]Data with microcystin detection in the water.

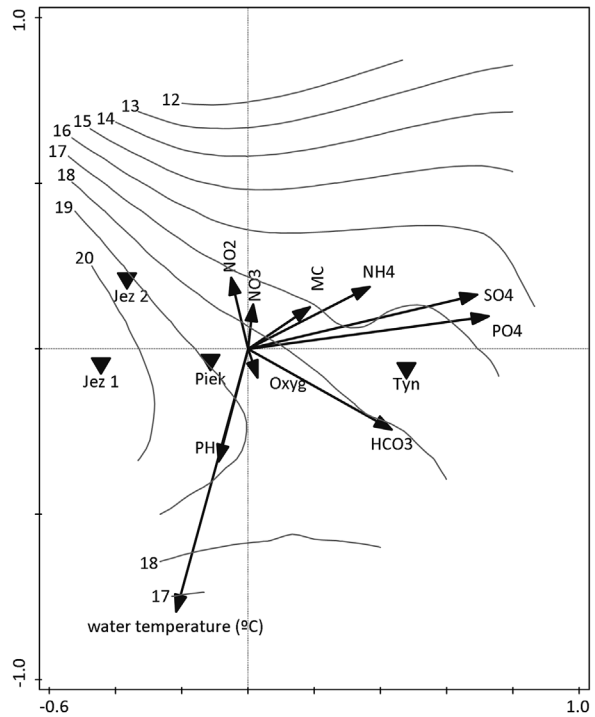

Figure 5. Canonical correspondence analysis for number of zooplankton species in individual samples, physicochemical parameters, and total concentration of microcystins in the water. Total variation: 1.02848, with explanatory variables accounting for 60.7%. Abbreviations: Tyn = Tyniec oxbow lake; Piek = Piekary oxbow lake; Jez1= Jeziorzany 1 oxbow lake; Jez2 = Jeziorzany 2 oxbow lake; water temp = water temperature; MC = microcystins in 1-m layer; oxyg = oxygen saturation; NO2, NO3, NH4 = nitrogen ions (NO_2^-, NO_3^-, NH_4^+); PO4 = phosphate ions (PO_4^{3-}); HCO_3 = hydrocarbonates (HCO_3^-); SO4 = sulfates (SO_4^{2-}). Numbered lines represent the number of species in individual samples.

Canonical correlation analyses showed a positive relationship between microcystin concentration and levels of NH_4^+, PO_4^{3-}, and SO_4^{2-}in the Tyniec oxbow. In other work it was reported that hepatotoxic cyanobacterial strains produced more toxins under conditions of high phosphorus (P), the difference between high and low P being 2-fold to 4-fold [38]. That does not negate the suggestion that microcystins improve P uptake, which is given above in discussing the findings from Piekary. The production and storage of toxins inside cells might follow a scenario different from that of their release into the water. In systems with low availability of P, the cells might degrade more rapidly and the toxins might remain in the water longer. Non-nitrogen–fixing genera such as *Microcystis* and *Oscillatoria* produce more toxins under high-nitrogen conditions [13,38,39], and in both Tyniec and Piekary the nitrogen ion concentration was high.

In the literature we have found only 1 report on the effect of sulfates on cyanotoxin production; in *A. ovalisporum* cells, sulfate and phosphate limitation were suggested to have pleiotropic effects on cyanobacterial toxin metabolism [40].

The most common and most toxic form of microcystin is believed to be MC-LR [17]; data on the other forms are insufficient [41]. According to the World Health Organization, its concentration in surface water should not exceed 0.001 mg/L [42]. We found that form of microcystin mostly in the upper layers of the Tyniec (only at the beginning of September) and Piekary oxbows (in October). Other forms such as MC-RR and MC-YR were found in the upper layers and close to the bottom at the end of the bloom period. Both of those forms are known to reduce the growth rate of adult *Daphnia* [43], and MC-RR and unknown metabolites of cyanobacterial extracts have negative effects on *D. magna* reproduction processes, similar to those observed in response to endocrine-disruptive molecules [43].

Opinions differ on the effect of cyanotoxins on zooplankton in water ecosystems. Laboratory studies strongly indicate adverse effects of cyanotoxins [44], but there are conflicting results from studies in natural ecosystems [45,46]. For example, the effect of toxins on cladocerans in field studies, including our present study, was not as clear as in laboratory experiments. *Daphnia* is the cladoceran genus most frequently tested against microcystins, because its species are extremely vulnerable to them. Field observations and large-scale lake experiments have shown that daphnid populations can be affected by toxicity if the toxins are released into the water at high concentrations following lysis of cyanobacterial cells [47]. Increasing numbers of reports suggest that *Daphnia*–cyanobacteria relationships are more complicated than previously thought, and that a decrease in the daphnid population during cyanobacterial blooms is not necessarily because of the toxins [48]. Moreover, short-term exposure to toxic cyanobacteria has been shown to improve the fitness of *D. magna* for further exposure to toxic prey during development. This trait might be transferred to offspring via maternal effects, or such an adaptation might also be clone-specific [49,50]. In the present study we did not find decreased cladoceran diversity in the presence of toxins; we observed decreased cladoceran density in Piekary but not in the Tyniec oxbow. The Tyniec oxbow yielded more species of daphnia than Piekary. Maternal effects might explain the difference in the response of the cladoceran populations of those oxbows.

Assessments of the sensitivity of rotifers to toxic cyanobacteria also differ. Some studies indicate that rotifers are more sensitive than copepods to microcystin exposure [51], and others suggest that rotifers are tolerant to cyanotoxins [52]. The present study showed decreased rotifer diversity in the presence of cyanotoxins in both of those oxbows, but the response of density under exposure to microsystins differed: it decreased in the Tyniec oxbow but not in Piekary.

Results on copepod sensitivity to cyanotoxins vary as well. One study showed reduced survival of *Eurytemora affinis* under exposure to elevated microcystin levels [53]. In the present study, *E. affinis* was present in an oxbow where microcystins were present at a high concentration (Piekary oxbow). This species might not be a good example, however, because it has been reported that *E. affinis* can detoxify nodularin, another cyanobacterial hepatotoxin, suggesting that some copepods have adapted to algal toxins and possess detoxification mechanisms [54]. In general, copepods showed decreased diversity and density under toxin exposure in Piekary but not in the Tyniec oxbow.

It is difficult to pick apart the responses of zooplankton to cyanotoxins in field data, because several factors simultaneously affect their diversity and density. The most important of these factors are water flow [55], water temperature and water chemistry [56], the food base [57], and the impact of macrophytes [58], and predation by fish [59]. The oxbows studied did not differ in water inflow, water temperature, macrophyte impacts, or fish predation. The highest differences were in the chemical properties responsible for cyanobacterial blooms, the presence of cyanobacterial blooms, and the presence, concentration, and duration of microcystins.

CONCLUSIONS

We found the highest total number of zooplankton species during the vegetation period in the Tyniec oxbow, the most polluted lake, which had the longest duration cyanobacterial bloom but a lower microcystin concentration. Surprisingly, that oxbow had the highest number of cladoceran and copepod species but the fewest rotifer species for the study period as a whole.

We conclude that zooplankton diversity showed a weak response to the presence of microcystins released into the water. Rotifers were the most sensitive group in terms of diversity but not in terms of density. Cladoceran density, but not diversity, decreased under exposure to the toxins in both oxbows with blooms. In general, cladoceran diversity was lower in the Piekary oxbow, where the concentration of cyanotoxins was highest. Copepod diversity decreased slightly in the Piekary oxbow but not in Tyniec. It is difficult to maintain that the decreased copepod density was a response to cyanotoxins, because it was lower in all oxbows studied (with and without toxins) in September and October.

Statistical analyses showed that the number of species in individual samples was negatively correlated with the levels of sulfates, phosphates, and ammonia, but the microcystin concentration was positively related to those levels. This points to the complexity of the interactions and synergies among toxins, abiotic factors, and zooplankton biodiversity. In focusing on the problem of cyanotoxins in these fragile ecosystems, conservation studies, and planning should pay attention to that complexity.

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Conflict of Interest—The authors declare that they have no conflicts of interest.

Data Availability—Data are available on request from Wojciech Krzton (krzton@iop.krakow.pl).

REFERENCES

1. Hubbell SP. 2001. *The Unified Neutral Theory of Biodiversity and Biogeography* (MPB-32, Vol 32). Princeton University Press, Princeton, NJ, USA.
2. Humphries CJ, Williams PH, Vane-Wright RI. 1995. Measuring biodiversity value for conservation. *Annu Rev Ecol Syst* 26:93–111.
3. Hooper DU, Chapin III FS, Ewel JJ, Hector A, Inchausti P, Lavorel S, Lawton JH, Lodge DM, Loreau M, Naeem S, Schmid B, Setälä H, Symstad J, Vandermeer J, Wardle DA. 2005. Effects of biodiversity on ecosystem functioning: A consensus of current knowledge. *Ecol Monogr* 75:3–35.
4. Dudgeon D, Arthington AH, Gessner MO, Kawabata Z-I, Knowler DJ, Lévêque C, Naiman RJ, Prieur-Richard A-H, Soto D, Stiassny MLJ, Sullivan CA. 2006. Freshwater biodiversity: Importance, threats, status and conservation challenges. *Biol Rev* 81:163–182.
5. Gopal B. 2013. Future of wetlands in tropical and subtropical Asia, especially in the face of climate change. *Aquat Sci* 75:39–61.
6. O'Neil JM, Davis TW, Burford MA, Gobler CJ. 2012. The rise of harmful cyanobacteria blooms: The potential roles of eutrophication and climate change. *Harmful Algae* 14:313–334.
7. Dokulil MT, Teubner K. 2000. Cyanobacterial dominance in lakes. *Hydrobiologia* 438:1–12.
8. Moustaka-Gouni M, Vardaka E, Tryfon E. 2007. Phytoplankton species succession in a shallow Mediterranean lake (L. Kastoria, Greece): Steady-state dominance of *Limnothrix redekei*, *Microcystis aeruginosa* and *Cylindrospermopsis raciborskii*. *Hydrobiologia* 575: 129–140.
9. Bucka H, Wilk-Woźniak E. 1999. Cyanobacteria responsible for planktic water blooms in reservoirs in southern Poland. *Algol Stud Arch Hydrobiol Suppl* 94:105–113.
10. Paerl HW, Paul VJ. 2012. Climate change: Links to global expansion of harmful cyanobacteria. *Water Res* 46:1349–1363.
11. Paerl HW, Fulton RS, Moisander PH, Dyble J. 2001. Harmful freshwater algal blooms, with an emphasis on cyanobacteria. *Sci World J* 1:76–113.
12. Codd GA, Morrison LF, Metcalf JS. 2005. Cyanobacterial toxins: Risk management for health protection. *Toxicol Appl Pharmacol* 203: 264–272.
13. Van Apeldoorn ME, Van Egmond HP, Speijers GJ, Bakker GJ. 2007. Toxins of cyanobacteria. *Mol Nutr Food Res* 51:7–60.
14. Paerl HW, Otten TG. 2013. Harmful cyanobacterial blooms: Causes, consequences, and controls. *Microbial Ecol* 65:995–1010.
15. Loftin KA, Clark JM, Journey CA, Kolpin DW, Van Metre PC, Bradley PM. 2016. Spatial and temporal variation in microcystins occurrence in wadeable streams in the southeastern USA. *Environ Toxicol Chem* 35: in press. DOI: 10.1002/etc.3391.
16. Ferrão-Filho ADS, Kozlowsky-Suzuki B. 2011. Cyanotoxins: Bioaccumulation and effects on aquatic animals. *Mar Drugs* 9:2729–2772.
17. Chorus I, Bartram J, eds. 1999. *Toxic Cyanobacteria in Water, A Guide to Their Public Health Consequences, Monitoring and Management*. WHO, Spon Press, London, UK.
18. Ger KA, Hansson LA, Lürling M. 2014. Understanding cyanobacteria-zooplankton interactions in a more eutrophic world. *Freshwater Biol* 59:1783–1798.
19. Obolewski K, Glińska-Lewczuk K. 2011. Effects of oxbow reconnection based on the distribution and structure of benthic macroinvertebrates. *Clean Soil Air Water* 39:853–862.
20. Wilk-Woźniak E, Ligęza S, Shubert E. 2014. Effect of water quality on phytoplankton structure in oxbow lakes under anthropogenic and non-anthropogenic impacts. *Clean Soil Air Water* 42:421–427.
21. Geist J. 2011. Integrative freshwater ecology and biodiversity conservation. *Ecol Indic* 11:1507–1516.
22. Dittrich J, Dias JD, Bonecker CC, Lansac-Tôha F, Padial A. 2016. Importance of temporal variability at different spatial scales for diversity of floodplain aquatic communities. *Freshwater Biol* 61: 316–327.
23. Meriluoto J, Codd GA. 2005. Cyanobacterial monitoring and cyanotoxin analysis. *Acta Acad Abo* 65:1–145.
24. Benzie JA. 2005. The genus *Daphnia* (including Daphniopsis) (Anomopoda: Daphniidae). T. 21. Guide to the Identification of the Microinvertebrates of the Continental Waters of the World. Leiden Kenobi Productions, Ghent and Backhuys, Leiden, The Netherlands.
25. Dussart B. 1967. Les Copépodes des eaux continentals d'Europe occidentale: Calanoides et harpacticopides. Boubée, Paris, France.
26. Dussart B. 1969. Les Copépodes des eaux continentals d'Europe occidentale: Cyclopoides et biologie. Boubée, Paris, France.
27. Ejsmont-Karabin J, Radwan S, Bielańska-Grajner I. 2004. *Polish Freshwater Fauna. Rotifers (Rotifera)*. University of Łódź, Łódź, Poland.
28. Flößner D. 2000. Die Haplopoda und Cladocera (Ohne Bosminidae) Mitteleuropas. Backhuys, Leiden, The Netherlands.
29. Rybak JI, Błędzki LA. 2010. Słodkowodne skorupiaki planktonowe: Klucz do oznaczania gatunków. Wydawnictwo Uniwersytetu Warszawskiego, Warszawa, Poland.
30. Komàrek J. 2013. *Cyanoprokaryota*, 3. Teil / 3rd part: *Heterocytous Genera*. Süsswasserflora von Mitteleuropa 19/3, Springer-Verlag, Berlin, Germany.
31. Komárek J, Anagnostidis K. 1998. *Cyanoprokaryota*, 1. Teil: *Chroococcales*. Süsswasserflora von Mitteleuropa 19/1, Gustav Fischer Verlag, Stuttgart, Germany.
32. Komárek J, Anagnostidis K. 2005. *Cyanoprokaryota*, 2. Teil/ 2nd Part: *Oscillatoriales*. Süsswasserflora von Mitteleuropa 19/2, Elsevier, Spektrum Akademischer Verlag, Heidelberg, Germany.
33. Rott E. 1981. Some results from phytoplankton counting intercalibrations. *Schweiz Z Hydrol* 43:34–62.
34. Ger KA, Teh SJ, Goldman CR. 2009. Microcystin-LR toxicity on dominant copepods *Eurytemora affinis* and *Pseudodiaptomus forbesi* of the upper San Francisco Estuary. *Sci Total Environ* 407:4852–4857.
35. Duy TN, Lam PKS, Shaw G, Connell DW. 2000 Toxicology and risk assessment of freshwater cyanobacterial (bluegreen algal) toxins in water. *Rev Environ Contam Toxicol* 163:113–186.
36. Hyenstrand P, Blomqvist P, Pettersson A. 1998. Factors determining cyanobacterial success in aquatic systems: A literature review. *Arch Hydrobiol Spec Issues* 51:41–62.
37. Holland A, Kinnear S. 2013. Interpreting the possible ecological role (s) of cyanotoxins: Compounds for competitive advantage and/or physiological aide? *Mar Drugs* 11:2239–2258.
38. Kaebernick M, Neilan BA. 2001. Ecological and molecular investigations of cyanotoxin production. *FEMS Microbiol Ecol* 35:1–9.
39. Yoshida M, Yoshida T, Takashima Y, Hosoda N, Hiroishi S. 2007. Dynamics of microcystin-producing and non-microcystin-producing *Microcystis* populations is correlated with nitrate concentration in a Japanese lake. *FEMS Microbiol Lett* 266:49–53.
40. Bácsi I, Vasas G, Surányi G, M-Hamvas M, Máthé C, Tóth E, Grigorszky I, Gáspár A, Tóth S, Borbely G. 2006. Alteration of cylindrospermopsin production in sulfate- or phosphate-starved cyanobacterium *Aphanizomenon ovalisporum*. *FEMS Microbiol Lett* 259:303–310.
41. Roegner AF, Brena B, González-Sapienza G, Puschner B. 2014. Microcystins in potable surface waters: Toxic effects and removal strategies. *J Appl Toxicol* 34:441–457.
42. Corbel S, Mougin C, Bouaïcha N. 2014. Cyanobacterial toxins: Modes of actions, fate in aquatic and soil ecosystems, phytotoxicity and bioaccumulation in agricultural crops. *Chemosphere* 96:1–15.
43. Hulot FD, Carmignac D, Legendre S, Yepremian C, Bernard C. 2012. Effects of microcystin-producing and microcystin-free strains of *Planktothrix agardhii* on long-term population dynamics of *Daphnia magna*. *Ann Limnol Int J Limnol* 48:337–347.
44. Lampert W. 1987. Laboratory studies on zooplankton–cyanobacterial interactions. *New Zeal J Mar Fresh* 21:483–490.
45. Hansson LA, Gustafsson S, Rengefors K, Bomark L. 2007. Cyanobacterial chemical warfare affects zooplankton community composition. *Freshwater Biol* 52:1290–1301.
46. Chen F, Dai X, Shu T, Gulati RD, Liu Z. 2013. Microcystins derived from lysing *Microcystis* cells do not cause negative effects on crustacean zooplankton in Lake Taihu, China. *Aquat Ecol* 47:379–387.
47. Ghadouani A, Pinel-Alloul B, Plath K, Codd GA, Lampert W. 2004. Effects of *Microcystis aeruginosa* and purified microcystin-LR on the feeding behavior of *Daphnia pulicaria*. *Limnol Oceanogr* 49:666–679.
48. Wojtal-Frankiewicz A, Bernasińska J, Frankiewicz P, Gwoździński K, Jurczak T. 2014. Response of Daphnia's antioxidant systems to spatial heterogeneity in cyanobacteria concentrations in a lowland reservoir. *PLoS One* 9:e112597.
49. Lyu K, Guan H, Wu C, Wang X, Wilson AE, Yang Z. 2015. Maternal consumption of non-toxic *Microcystis* by *Daphnia magna* induces tolerance to toxic *Microcystis* in offspring. *Freshwater Biol* 61:219–228.
50. Jiang X, Yang W, Zhao S, Liang H, Zhao Y, Chen L, Li R. 2013. Maternal effects of inducible tolerance against the toxic

cyanobacterium *Microcystis aeruginosa* in the grazer *Daphnia carinata*. *Environ Pollut* 178:142–146.
51. Gilbert JJ. 1996. Effect of food availability on the response of planktonic rotifers to a toxic strain of the cyanobacterium *Anabaena flos-aquae*. *Limnol Oceanogr* 41:1565–1572.
52. Fulton RS, Paerl H. 1987. Effects of colonial morphology on zooplankton utilization of algal resources during blue-green algal (*Microcystis aeruginosa*) blooms. *Limnol Oceanogr* 32:634–644.
53. Reinikainen M, Lindvall F, Meriluoto JAO, Pepka S, Sivonen K, Spoof L, Wahlsten FA. 2002. Effects of dissolved cyanobacterial toxins on the survival and egg hatching of estuarine calanoid copepods. *Mar Biol* 140:577–583.
54. Karjalainen M, Kozlowsky-Suzuki B, Lehtiniemi M, Engstrom-Ost J, Kankaanpaa H, Viitasalo M. 2006. Nodularin accumulation during cyanobacterial blooms and experimental depuration in zooplankton. *Mar Biol* 148:683–691.
55. Godlewska M, Mazurkiewicz-Boroń G, Pociecha A, Wilk-Woźniak E, Jelonek M. 2003. Effects of flood on the functioning of the Dobczyce reservoir ecosystem. *Hydrobiologia* 504:305–313.
56. Shurin JB, Winder M, Adrian R, Keller WB, Matthews B, Paterson AM, Pinel Alloul B, Rusak JA, Yan ND. 2010. Environmental stability and lake zooplankton diversity–contrasting effects of chemical and thermal variability. *Ecol Lett* 13:453–463.
57. Gulati R, Demott W. 1997. The role of food quality for zooplankton: Remarks on the state-of-the-art, perspectives and priorities. *Freshwater Biol* 38:753–768.
58. van Donk E, van de Bund WJ. 2002. Impact of submerged macrophytes including charophytes on phyto-and zooplankton communities: Allelopathy versus other mechanisms. *Aquat Bot* 72:261–274.
59. Pociecha A, Amirowicz A. 2003. An attempt to determine the share of zooplankton in food consumed by fish in the limnetic zone of a eutrophic dam reservoir. *Oceanol Hydrobiol Stud* 32:15–31.