

## Screening of phytoplankton blooms for Cyanobacteria species composition, abundances, and toxins

DR-18-09; September 2018

Brent Bellinger, Ph.D.  
City of Austin  
Watershed Protection Department  
Environmental Resource Management Division

### ABSTRACT

*Increased incidence and magnitude of cyanobacterial blooms in freshwater systems are a concern, especially if significant concentrations of toxins are produced. Recreational and drinking water supplies can be impaired by these blooms as certain toxins have been linked to afflictions such as skin rashes and liver cancer. In extreme instances, toxins have been linked to death of livestock and vulnerable human populations. Health organizations and municipalities have increased monitoring efforts to evaluate the presence of potentially toxigenic cyanobacteria and ambient concentrations of toxins in regionally important lakes and reservoirs. During a recent drought, cyanobacterial biomass was observed to increase by orders of magnitude in two reservoirs in Austin, Texas: Lake Austin and Lady Bird Lake. In two consecutive summer bloom events, City of Austin staff collected grab samples from these two reservoirs to identify species composition and screen for a suite of toxins. Cyanobacteria densities (30,000 – 96,000 org/mL) exceeded thresholds associated with a low risk to humans (>20,000 org/mL), and included potentially toxigenic species (e.g., *Pseudanabaena limnetica*, *Cylindrospermopsis raciborskii*). Fortunately, only trace amounts (<0.3 µg/L) of cylindrospermopsin, a toxin of potential concern in drinking water, were measured during the first year of screening. Results indicate that, while cyanobacteria may become prolific under certain environmental conditions in Austin's reservoirs, users do not currently appear at risk of exposure to significant concentrations of toxins. However, as the region experiences rapid development that changes land use under climate change, continued monitoring of Austin's reservoirs is a prudent measure to track cyanobacterial blooms and associated risks.*

### INTRODUCTION

Increased incidence of cyanobacterial blooms and related production of toxins is a growing concern for municipalities that manage lakes and reservoirs for recreational use and drinking water supply. Increased frequency, duration, and magnitude of cyanobacterial bloom and toxin events have been linked to anthropogenic eutrophication due to watershed development, and warmer waters and increased duration of stratification with climate change (Paerl and Huisman 2008, 2009; Brooks et al. 2016; Paerl et

al. 2016). Exposure to toxins can inhibit beneficial uses of a water body due to illnesses including skin and respiratory irritation, neurological effects, liver and kidney cancer, and in extreme instances may lead to the death of livestock, pets, waterfowl, and children (Carmichael 2001; Fleming et al. 2002; WHO 2003; Osswald et al. 2007). The World Health Organization (WHO 2003), proposed guidelines protective of water supplies for cyanobacterial abundances (e.g., >20,000 org/mL is low risk) and the toxin microcystin-LR (<1 µg/L). The United States Environmental Protection Agency has provided protective concentration criteria for two toxins, microcystin (<1.0 µg/L) and cylindrospermopsin (<3 µg/L) (USEPA 2015a, b), and some states have adopted drinking water guidelines for a suite of additional toxins (<https://www.epa.gov/nutrient-policy-data/guidelines-and-recommendations>). As urban areas grow, monitoring of drinking water supplies for cyanobacteria and associated toxins will be critical to prevent public health crises.

Lake Austin, a popular recreational reservoir and the primary drinking water supply for the City of Austin, was observed to have cyanobacterial blooms that increased in magnitude and duration during a recent drought period (Bellinger et al. 2018). In response, the department that provides drinking water, Austin Water, began screening for three toxins (microcystin, cylindrospermopsin, and anatoxin-a) when cyanobacterial blooms exceeded 15,000 org/mL. However, there are many more potential toxins that may be present in the water supply, and not all cyanobacterial species produce toxins. The popular recreational reservoir immediately down river, Lady Bird Lake, was not monitored for toxins, although the Watershed Protection Department does conduct routine monitoring of phytoplankton species composition and abundance. This pilot study was designed to clarify the need for future monitoring and management actions by evaluating species composition and a broad suite of toxins from samples collected near the peak seasonal cyanobacterial biomass.

## METHODS

Utilizing Austin Water's daily reporting of phytoplankton bloom conditions in Lake Austin, Watershed Protection staff attempted to capture cyanobacterial biomass near its annual peak. Due to irregular rainfall patterns and discharge rates, only two large phytoplankton bloom events were sampled; one on 25 September 2015 and the other on 29 August 2016. Five sites were sampled in 2015 and four sites were sampled in 2016 (Fig. 1). In 2015, Lake Austin sample sites included the Loop 360 bridge (#933), Davis water treatment plant (WTP; #4543, formerly identified as site #10758), and Ullrich WTP (#4534). Lady Bird Lake sample sites were the 1<sup>st</sup> St. bridge (#2) and the basin (#1). In 2016 the same sites were sampled except the Loop 360 site of Lake Austin. At each site, two 1 L surface water (0.3 m) grab samples were collected approximately 100–200' from shore, one for phytoplankton community composition and one for toxin screening. Species composition results were only available from the 2016 samples due to improper preservation of the 2015 samples. Samples were analyzed by GreenWater Laboratories (Palatka, FL) for species composition and toxins. The toxin suite included: microcystin/nodularin Suite (-LR, -RR, -yR, -LA, -dmLR, -LY, -LW, -LF, NOD); cylindrospermopsin, anatoxin-a, saxitoxin suite (C1/C2, GTX (1,2,3,4,5), dcGTX2/3, dcSTX, NEO, STX); lyngbyatoxin-A, and aplysiatoxin/debromoaplysiatoxin. Identification and quantification of all toxins was with liquid chromatography-mass spectrometry/mass spectrometry (LC-MS/MS). Austin Water staff concomitantly screened for microcystin and cylindrospermopsin on 10, 14, and 15 September 2015 and 30 August through 1 September 2016. Their grab samples were taken at the water intake structure at the Davis and Ullrich WTPs. Their results are included here for comparison of nearshore relative to offshore variability.

## RESULTS & DISCUSSION

Cyanobacteria cell counts from the water treatment plants in Lake Austin on September 25, 2015, were above 46,000 org/mL (Table 1), two orders of magnitude above Austin Water's non-bloom criteria of 300 org/mL, and exceeding the criteria established by the WHO as being a low risk cyanobacterial bloom (WHO 2003). Taxonomic data collected as part of routine monitoring in August and October for each reservoir identified *Oscillatoria* sp. as the dominant taxa (Winsborough unpubl. data), which may be the homotypic synonym of *Pseudanabaena limnetica*, the dominant species identified in the 2016

samples (Table 2). Genera from the Oscillatoriaceae family have been shown capable of producing toxins and compounds contributing to taste-and-odor problems (Carmichael 2001). From all sites, only cylindrospermopsin was detected. Concentrations were greater in Lake Austin (0.25–0.29 µg/L) than in Lady Bird Lake (0.07 µg/L), but both reservoirs fell well below EPA criteria for concern. Austin Water cyanobacteria counts from the week before our sampling event were between 15,000 and 26,000 org/mL, approximately half the biomass we captured (Table 1). They also detected cylindrospermopsin at each WTP, but concentrations were similarly half (12–14 µg/L) our findings.

In 2016, a larger bloom event was captured. Cyanobacteria cell counts estimated by GreenWater Laboratory were >81,000 org/mL at Ullrich and >76,000 org/mL at the Davis sites (Table 2). Based on cell counts at the WTP intakes on our sampling date, cyanobacteria concentrations were approximately 68,000 org/mL (Table 1). Density differences between labs could be due to methodology (i.e., how filaments/cells/colonies are counted) or sampling location. Austin Water collects samples at the intake structures (nearshore), whereas samples we collected for this study were collected closer to the middle of the reservoir. Sampling by AW from 30 Aug to 1 Sept captured the bloom in senescence, as counts declined from 54,000 to 17,000 org/mL. The dominant species from both sites in Lake Austin was *Pseudanabaena limnetica*, representing over 50% of the total cell biomass (Table 2). Additional prominent, and potentially toxigenic, species present were: *Geitlerinema/Jaaginema* sp., *Cylindrospermopsis raciborskii*, and *Pseudanabaena* sp. (Flaherty et al. 2007). In Lady Bird Lake cell counts were greater, between 92,000 and 96,000 org/mL, which approached the WHO threshold of 100,000 org/mL for a moderate probability of adverse health effects (Table 2; WHO 2003). The dominant species and relative abundances in Lady Bird were similar to Lake Austin. Despite the abundance of cyanobacteria, and notably of potentially toxigenic species, no toxins were detected from our or AW samples in 2016 (Table 1).

At the end of the recent drought period, we sampled some of the largest cyanobacteria bloom events recorded for Lake Austin. Although these blooms did give the water a blue-green appearance, they did not result in the thick scums that have recently afflicted lakes around the world (Paerl and Huisman 2009; Steffen et al. 2014; Paerl 2017). In addition, appreciable concentrations of toxins were not observed to be associated with the Austin reservoir's bloom events, despite the presence of cyanobacterial species capable of producing toxins. We hypothesize that the lack of toxicity associated with the sampled bloom events is related to the lower nutrient concentrations relative to the eutrophic systems generally afflicted with toxic blooms. Differences in cell counts and toxin concentrations in 2015 between our screening and AW screening suggest that near- or off- shore sampling may impact findings. Toxin concentrations at the former locations will be important for recreational users accessing the water from shore or municipal intakes, whereas blooms and toxins measured from the limnetic will impact recreational boaters.

The recent colonization of the Austin reservoirs by zebra mussel could result in changes in nutrient concentrations or stoichiometry and shifts in phytoplankton species. Increases and shifts in either could impact critical ecological thresholds or species environmental responses leading to production of toxins (Knoll et al. 2008; Yuan and Pollard 2015). Future monitoring will be especially important during drought periods when the probability of a cyanobacterial bloom significantly increases (Bellinger et al. 2018). Depending on the phytoplankton bloom magnitude and duration, supplemental sampling and screening for toxins would increase the likelihood that reservoir users will be informed about and protected from potential risks, and remediation efforts could be implemented to ensure the water supplies remain useable.

## RECOMMENDATIONS

- Continue monitoring nutrient concentrations, stoichiometry, and phytoplankton assemblages of Austin's reservoirs due to large cyanobacteria bloom development potential
- Develop a monitoring program/plan to identify statistically based shifts in nutrients and phytoplankton assemblages that could lead to undesirable conditions
- Develop corroborating methods that effectively identify cyanobacteria to the species level

- Develop methods and techniques to more rapidly screen for presence of toxigenic cyanobacteria
- Identify whether the cyanobacteria present have the genes necessary to produce toxins
- During bloom events, screen for presence of a broad suite of toxins beyond those for which criteria have been developed
- Relate annual patterns in cyanobacteria abundances with zebra mussel population dynamics
- Couple screening efforts nearshore and offshore to ensure cross-sectional variability is captured
- Continue to engage Austin Water staff as screening programs are developed to ensure the most robust data sets are being collected and evaluated for the protection Austin’s reservoir users.

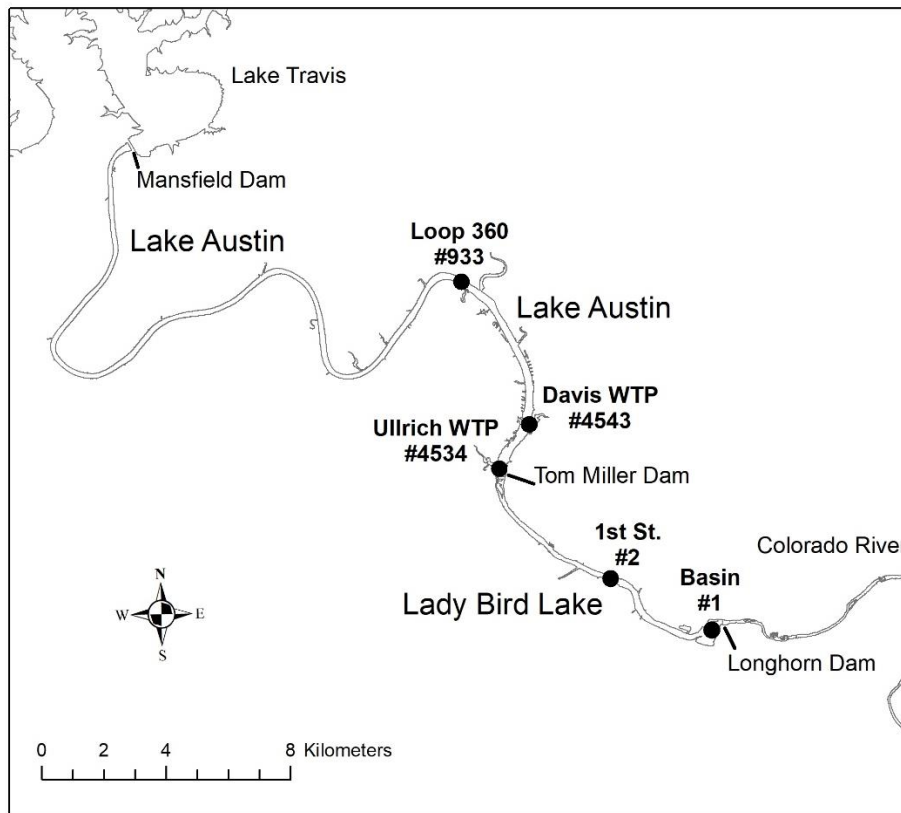


Figure 1. Locations in Lake Austin and Lady Bird Lake where surface water grab samples were collected for determination of cyanobacteria species and toxin concentrations.

Table 1. Cyanobacteria cell counts (org/mL) and toxin concentration ( $\mu\text{g/L}$ ) determined by Austin Water for samples collected at the Davis (#4543) and Ullrich (#4534) water treatment plant intakes in 2015 and 2016.

Date	Site	Cell count (org/mL)	Cylindrospermopsin ( $\mu\text{g/L}$ )
10 September 2015	Davis	15,000	0.14
	Ullrich	19,000	0.12
14 September	Davis	No Data	0.12
	Ullrich	26,000	0.14
15 September	Davis	25,000	No Data

25 September	Davis	46,000	0.25*
29 August 2016	Ullrich	68,000	Not Detected*
30 August	Davis	54,000	No Data
31 August	Ullrich	41,000	Not Detected
1 September	Davis	17,000	No Data

\*Based on samples collected offshore and analyzed by GreenWater Laboratory.

Table 2. Cyanobacteria species and cell counts (cell/mL) from samples collected from Lake Austin (Davis and Ullrich WTPs) and Lady Bird Lake (1<sup>st</sup> St. and Basin) on 29 August 2016.

Genus	Species	Cell Counts (cells/mL)			
		Davis WTP (#4543)	Ullrich WTP (#4534)	1 <sup>st</sup> Street (#2)	Basin (#1)
<i>Aphanocapsa</i>	sp.	3			
A.	cf. <i>elachista</i>			393	418
<i>Chroococcus</i>	sp.		4		
<i>Chrysochloris</i>	sp.	668	120	24	48
<i>Cyanogranis</i>	<i>ferruginea</i>	698	1,885	1,257	1,257
<i>Cyanophyte cell spp.</i>		873	1,571	1,885	2,376
<i>Cylindrospermopsis</i>	<i>raciborskii</i>	2,560	2,513	3,770	6,911
<i>Dactylococcopsis</i>	<i>irregularis</i>	4,247	3,770	5,655	5,812
<i>Geitlerinema/Jaaginema</i>	sp.	116	6,350	11,938	11,938
<i>Komvophoron</i>	<i>schmidlei</i>	7			
<i>Merismopedia</i>	<i>punctata</i>		1,629		543
M.	<i>tenuissima</i>	1,862			
M.	<i>punctata</i>	181			
<i>Microcystis</i>	<i>wesenbergii</i>	17			
Nostocalean filament	sp. 1	66	110	308	198
Nostocalean filament	sp. 2	3		10	
<i>Planktolyngbya</i>	f. <i>limnetica</i>	9,425	14,137	11,310	16,964
<i>Planktolyngbya</i>	<i>microspira</i>			79	
<i>Pseudanabaena</i>	<i>limnetica</i>	49,102	48,380	54,663	46,495
<i>Pseudanabaena</i>	sp.	2,211	42	746	2,381
Total cell count		76,343	81,263	92,037	95,342

## References

Bellinger BB, Richter A, Porras A, Davis SL. 2018. Drought and management effects of biophysicochemistry in a rapidly-flushed reservoir. *Lake Reserv. Manage.* 34: 182-198.

Brooks BW, Lazorchak JM, Howard MDA, Johnson M-VV, Morton SL, Perkins DAK, Reavie ED, Scott GI, Smith SA, Steevens JA. 2016. Are harmful algal blooms becoming the greatest inland water quality threat to public health and aquatic ecosystems? *Environ. Toxicol. Chem.* 35: 6-13.

Carmichael WW. 2001. Health effects of toxin-producing cyanobacteria: "The cyanoHABS". Hum. Ecol. Risk Assess. 7: 1393-1407.

Flaherty KW, Walker HL, Britton CH, Lembi CA. 2007. Response of *Cylindrospermopsis raciborskii* and *Pseudanabaena limnetica* to a potential biological control agent, bacterium SG-3 (*Lysobacter* cf. *brunescens*). Lake Reserv. Manage. 23: 255-263.

Fleming LE, Rivero C, Burns J, Williams C, Bean JA, Shea KA, Stinn J. 2002. Blue green algal (cyanobacterial) toxins, surface drinking water, and liver cancer in Florida. Harmful Algae 1: 157-168.

Knoll LB, Sarnelle O, Hamilton SK, Kissman CEH, Wilson AE, Rose JB, Morgan MR. 2008. Invasive zebra mussels (*Dreissena polymorpha*) increase cyanobacterial toxin concentrations in low-nutrient lakes. Can. J. Fish. Aquat. Sci. 65: 448-455.

Paerl HW, Huisman J. 2008. Blooms like it hot. Science 320: 57-58.

Paerl HW, Huisman J. 2009. Climate change: A catalyst for global expansion of harmful cyanobacterial blooms. Env. Microbiol. Rep. 1: 27-37.

Paerl HW, Scott JT, McCarthy MJ, Newell SE, Gardner WS, Havens KE, Hoffman DK, Wilhelm SW, Wurtsbaugh WA. 2016. It takes two to tango: When and where dual nutrient (N & P) reduction are needed to protect lakes and downstream ecosystems. Environ. Sci. Tech. 50: 10805-10813.

Paerl HW. 2017. Controlling cyanobacterial harmful blooms in freshwater ecosystems. Microb. Biotechnol. 10: 1106-1110.

Steffen MF, Belisle BS, Watson SB, Boyer GL, Wilhelm SW. 2014. Status, causes and controls of cyanobacterial blooms in Lake Erie. J. Great Lakes Res. 40: 215-225.

United States Environmental Protection Agency (USEPA). 2015a. Health effects support document for the cyanobacterial toxin microcystins. Office of Water. Report #EPA-820R15102.

United States Environmental Protection Agency (USEPA). 2015b. Health effects support document for the cyanobacterial toxin cylindrospermopsin. Office of Water. Report #EPA-820R15103.

WHO. 2003. Guidelines for safe recreational water environments. Vol. 1: Coastal and fresh waters. Geneva. World Health Organization.

Yuan LL, Pollard AI. 2015. Deriving nutrient targets to prevent excessive cyanobacterial densities in U.S. lakes and reservoirs. Freshwater Biol. 60: 1901-1916.

Electronic references

<https://www.epa.gov/nutrient-policy-data/guidelines-and-recommendations>