

Project Title: Cyanotoxin presence and year-round dynamics in Falls Lake, North Carolina

Overview: Cyanobacterial Harmful Algal Blooms (CyanoHABs) in North Carolina freshwater systems can adversely impact drinking water, fisheries, tourism and food web resilience. The main goals of this study are to examine the spatiotemporal dynamics of CyanHABs in relation to cyanotoxins in Falls Lake. To acquire this information we determine algal growth together with cyanotoxin presence at multiple sampling sites throughout the lake and aim to identify environmental conditions that seem to favor algal growth and/or toxin production (“hot spots”). Furthermore, we aim to identify the cyanobacterial taxa that dominate the system and are associated with toxin presence and determine if our data indicate the accumulation of cyanobacterial biomass and/or toxins in certain regions of the lake.

Research Methods

On monthly surveys, in collaboration with the Department of Environmental Quality through NC DEQ’s Ambient Water Monitoring Program, we collected whole lake water at 11 stations to determine particulate (intracellular) and dissolved toxin levels and chlorophyll a (chl a) concentrations. At 4 out of 11 sampling stations Solid Phase Adsorption Toxin Tracking (or SPATT) units were deployed (Lane et al. 2010; Miller et al. 2010) to screen for MCY, ANA and CYL (NEU013B, NEU019E, LI01 and NEU19P). The SPATT method is an integrative, cost-effective, *in situ* screening tool that allows for the detection of specific dissolved toxins when traditional monitoring approaches (e.g., analyses of monthly “grab” whole water samples) cannot resolve toxin presence (Wood et al. 2011). All toxin samples were analyzed using commercially available enzyme-linked immunosorbent assays or ELISAs (Abraxis, Eurofin) following previously established protocols (Wiltsie et al. 2018). The toxin kits used were MCY-ADDA (sensitive to MCY-LR, -YR, -LF, -RR, LW, and nodularin; LDL = 0.10 $\mu\text{g L}^{-1}$), CYN (sensitive to CYN and deoxy-CYN; LDL = 0.04 $\mu\text{g L}^{-1}$), ANA (sensitive to anatoxin-a and homoanatoxin-a; LDL = 0.1 $\mu\text{g L}^{-1}$), STX (sensitive to STX and other paralytic shellfish poison [PSP] toxins; LDL = 0.015 $\mu\text{g L}^{-1}$), and BMAA (sensitive to BMAA and other amino acids; limit of quantitation = 4 $\mu\text{g L}^{-1}$). Chl a was measured following previously established non-acidification protocols (Welschmeyer 1994). Pertinent physicochemical parameters were collected with support of NC DEQ using an YSI sonde equipped to measure DO, temperature, conductivity and pH.

Preliminary Results

Overall, algal biomass (chl a) averaged slightly higher within the upper, wider portion of the lake (stations NEU013 through L01) compared to the more restricted, narrower, lower part of the lake (stations NEU019E through NEU020D) with 25.65 and 19.45 $\mu\text{g L}^{-1}$, respectively (Fig. 1A and Fig. 2). Chl a levels remained relatively high throughout late fall and early winter months and concentrations exceeded 40 $\mu\text{g L}^{-1}$ in 11% of the measurements (n = 122) confirming the lake to be impaired based on algal biomass (Section 303(d) of the Clean Water Act). Based on WHO-guidelines, these observed chl a ranges (i.e., from 10 to 50 $\mu\text{g L}^{-1}$) would align Falls Lake with water bodies that are characterized to hold moderate exposure risks for MCY (Fig 1A). Refining this categorization based on the actual MCY measurements from this study (i.e., 0.1 to 10 $\mu\text{g MCY}$

L⁻¹) exposure risks can be re-categorized as low for MCY (Fig. 1B and 3A). Overall, MCY concentrations tended to be slightly elevated for the lower part of Falls Lake further supporting the notion that chl a often do not match spatiotemporal trends for toxins. Over the 2 years of observations, we found that MCY and ANA (Fig. 3C)) are the most prevalent toxins found across the lake with CYL being present at times (Fig. 3B) and BMAA showing up sporadically (Table 1). For MCY and CYL, this was reflected using both a grab sampling approach and the *in-situ* toxin tracking tool (SPATTs, Table 1). The only toxin that could not be detected based on screening a subset of the locations and time points was STX (Table 1).

Cyanotoxin analyses for whole water samples showed that total MCY (dissolved and particulate fractions combined) averaged 0.11 µg L⁻¹ (range = below detection (bd) – 0.34 µg L⁻¹; n = 175). CYL concentrations averaged 0.04 µg L⁻¹ (range = bd – 0.54 µg L⁻¹; n = 112). MCY could be detected both intracellularly and dissolved, ANA was primarily detected in the particulate phase and CYL was only measured in the dissolved fraction (Table 1). No STX was detected at any of the 3 priority stations during any of the surveys. There is currently limited data available on how quickly varying toxins are released from cyanobacterial cells throughout the early phases of a bloom but several studies suggest release of toxins might be quick as a bloom reaches its peak and starts to decline (Caldwell Eldridge et al. 2012; Lahti et al. 1997). Our MCY and CYL results for whole lake water were in good agreement with toxin patterns based on the SPATT measurements with dissolved MCY values consistently exceeding those for CYL by ~two orders of magnitude. Comparing MCY grab data with those from SPATT deployments indicated that certain areas within the lake showed relatively high dissolved concentrations where toxin residence times may be prolonged (Figure 4). Overall, multiple stations tested positive for up to 4 of the toxins throughout the surveys. For the continuation of our study we intend to continue toxin measurements for MCY at all 11 stations, for CYL at 7 and for ANA, BMMA and STX at 3 priority stations. SPATTs extraction are pursued for all 4 deployment sites to analyze for MCY, CYL and ANA. A major focus of this coming project period will be to characterize the cyanobacterial communities throughout the lake, establish how species composition is related to toxin patterns and establish whether populations are connected throughout the lake indicating areas that are primary bloom and/or toxin production sites.

Management Implications

Maximal toxin concentrations – based on monthly coverage – did not exceed regulatory thresholds through the World Health Organization. However, the presence of multiple toxins year-round indicated potential risks from chronic low-level exposure to multiple toxins. Overall average MCY toxin concentrations were highest in fall of 2020 when the lower part of the lake seemed to be more affected (Fig. 3A). The prevalence of MCY and the simultaneous presence of multiple cyanotoxins may warrant future examination of toxin transfer to higher trophic levels (i.e., fish) which could pose a risk for human consumption. It should be noted, that our monthly observations may not capture maximal toxin concentrations. Since this is the first comprehensive study that focuses on cyanotoxin dynamics in Falls Lake we believe the information to be of value for local residents, monitoring agencies and recreational users. Based on our preliminary data, the inclusion of regular toxin monitoring seems warranted for several of the substances to more adequately

assess water quality and ecosystem health for Falls Lake. This study will be the first to create a baseline for cyanotoxin dynamics in Falls Lake and inform future investigations evaluating potential food web impacts and public health exposure risks for recreational users of the lake.

References

- Caldwell Eldridge, S. L., T. M. Wood, and K. R. Echols. 2012. Spatial and Temporal Dynamics of Cyanotoxins and Their Relation to Other Water Quality Variables in Upper Klamath Lake, Oregon, 2007–09. Scientific Investigations Report 2012-5069. USGS.
- Lahti, K., J. Rapala, M. Färdig, M. Niemelä, and K. Sivonen. 1997. Persistence of cyanobacterial hepatotoxin, microcystin-LR in particulate material and dissolved in lake water. *Water Research* **31**: 1005-1012.
- Lane, J. Q., C. M. Roddam, G. W. Langlois, and R. M. Kudela. 2010. Application of Solid Phase Adsorption Toxin Tracking (SPATT) for field detection of the hydrophilic phycotoxins domoic acid and saxitoxin in coastal California. *Limnology and Oceanography Methods* **8**: 645–660.
- Miller, M. A. and others 2010. Evidence for a Novel Marine Harmful Algal Bloom: Cyanotoxin (Microcystin) Transfer from Land to Sea Otters. *PLoS ONE* **5**: e12576.
- Welschmeyer, N. A. 1994. Fluorometric analysis of chlorophyll a in the presence of chlorophyll b and pheopigments. *Limnol. Oceanogr.* **39**: 1985-1992.
- Wiltsie, D., A. Schnetzler, J. Green, M. Vander Borgh, and E. Fensin. 2018. Algal Blooms and Cyanotoxins in Jordan Lake, North Carolina. *Toxins (Basel)* **10**: 92.
- Wood, S. A., P. T. Holland, and L. Mackenzie. 2011. Development of solid phase adsorption toxin tracking (SPATT) for monitoring anatoxin-a and homoanatoxin-a in river water. *Chemosphere* **82**: 888-894.