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 objective of this study is 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.

Our main research questions are as follows:

1) Are year-round patterns of cyanobacterial abundance (chlorophyll a) in Falls Lake associated with toxin presence for microcystin (MCY), cylindrospermopsin (CYN), anatoxin (ANA), saxitoxin (STX) and/or beta-Methylamino-L-alanine (BMAA)?
2) What are the relationships between CyanoHAB dynamics and pertinent physicochemical parameters? Can specific environmental factors be associated with algal abundance and/or toxin concentrations in Falls Lake?

Research Methods

For the first year of this project, we conducted monthly surveys in collaboration with the Department of Environmental Quality through NC DEQ's Ambient Water Monitoring Program. On monthly field trips, whole lake water was collected at 11 stations to determine particulate (intracellular) and dissolved toxin levels and chlorophyll a (chl a) concentrations. At 4 out of 11 sampling stations **S**olid **P**hase **A**dsorption **T**oxin **T**racking (or SPATT) units were deployed (Lane et al. 2010; Miller et al. 2010) to screen for MCY and CYL (NEU013B, NEU019E, LI01 and NEU19P; Fig. 1). 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 µg L^{-1}), CYN (sensitive to CYN and deoxy-CYN; LDL = 0.04 µg L^{-1}), ANA (sensitive to anatoxin-a and homoanatoxin-a; LDL = 0.1 µg L^{-1}), STX (sensitive to STX and other paralytic shellfish poison [PSP] toxins; LDL = 0.015 µg L^{-1}), and BMAA (sensitive to BMAA and other amino acids; limit of quantitation = 4 µg 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 a YSI sonde equipped to measure DO, temperature, conductivity and pH.

Preliminary Data and Initial Findings

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 µg L^{-1}, respectively (Fig. 2). Chl a levels remained relatively high throughout late fall and early winter months and concentrations exceeded 40 µg L^{-1} in 11% of the measurements (n = 122) confirming the to be impaired based on algal biomass (Section 303(d) of the Clean Water Act).

Cyanotoxin analyses for whole water samples were completed for MCY and CYL for 7 stations for surveys from July 2019 through Feb 2020 (Fig. 3). Preliminary data showed that MCY (dissolved and particulate fractions combined) averaged 0.10 µg L^{-1} (range = below detection (bd) – 0.27 µg L^{-1}; n = 112). CYL concentrations averaged 0.04 µg L^{-1} (range = bd – 0.54 µg L^{-1}; n = 112). Both MCY and CYL were primarily detected in the dissolved phase with little or no detectable concentrations in the particulate phase (intracellular). These findings indicated that sampling may have not coincided with peak bloom conditions when toxins are expected to be mostly bound inside the cells. 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 (July 2019 through Jan 2020) with dissolved MCY values consistently exceeding those for CYL by ~two orders of magnitude (Fig. 4).

Three of the stations (NEU013B, LI01 and NEU19P) were prioritized for preliminary ANA, STX and BMAA analyses to gain insight into the prevalence of these less common toxins in Falls Lake. In contrast to MCY and CYL, both ANA and BMAA were exclusively detected in the particulate fractions with an average of 0.13 and 0.52 µg L^{-1} and ranging from bd – 0.20 and from bd – 0.95 µg L^{-1}, respectively (n = 48 each). No STX was detected at any of the 3 priority stations during any of the surveys (n = 48). Overall, multiple stations tested positive for up to 4 of the toxins and throughout the surveys (Fig. 2). We were able to continue sampling with NC DEQ throughout the remainder of the year but, due to COVID-related laboratory closures, have yet to complete all toxin analyses. As research activities have recently resumed, we are in the process of completing MCY and CYL measurements for all 11 stations, ANA and BMMA for seven of the 11 stations and to screen for STX at the original 3 priority stations to confirm that STX does not pose a risk during CyanoHABs in Falls Lake. Also completed will be SPATT extractions and analyses for MCY and CYL.

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. Especially the prevalence of MCY may warrant future examination of toxin transfer to higher trophic levels (i.e., fish) which could pose a risk for human consumption . Furthermore, it is

likely that our monthly observations did not reflect maximal toxin concentrations during peak bloom conditions. 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

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