#### MICHIGAN DEPARTMENT OF ENVIRONMENTAL QUALITY WATER RESOURCES DIVISION MARCH 2016

# STAFF REPORT

# ALGAL TOXIN MONITORING IN MICHIGAN INLAND LAKES: 2015 RESULTS

#### **Introduction**

The term "harmful algal bloom" (HAB) generally describes accumulations of cyanobacteria that are aesthetically unappealing and produce algal toxins. In 2015 the Michigan Department of Environmental Quality (MDEQ), Water Resources Division (WRD), developed the following definition of a HAB (Kohlhepp, 2015[a]): "An algal bloom in recreational waters is harmful if microcystin levels are at or above the 20 µg/L World Health Organization (WHO) non-drinking water guideline, or other algal toxins are at or above appropriate guidelines that have been reviewed by MDEQ-WRD." A bloom should be considered *potentially* harmful when "the chlorophyll *a* level is greater than 30 micrograms per liter (µg/L) and visible surface accumulations/scum are present, or cells are visible throughout the water column." A key concept of this HAB definition is that while high chlorophyll *a* concentration and visible surface/water column algal accumulation can indicate potential problems, the WRD's focus is on the potential harm that toxins represent and so water samples must be analyzed for the presence of toxins to confirm that a bloom may, in fact, be harmful to humans or wildlife. Visible appearance of blooms cannot be used as a reliable predictor of toxin content.

The WRD receives reports each year about nuisance algal conditions, which may or may not be HABs, from district staff, lake associations, and the broader public. These reports can come in as concerns about algae or about suspected pollutants or toxic substances in the water, such as 'green paint' spills, which upon investigation, turn out to be algae. The number of such reports, particularly the occurrence of blue-green algal blooms and concern over the possible presence of algal toxins such as microcystin, appear to have increased in recent years, although this is difficult to quantitatively confirm (Parker, 2013 and Parker, 2014). As a result, the MDEQ-WRD established an internal work group in March 2013 to develop an approach to monitor, assess, and report on nuisance and harmful algal conditions, to improve our understanding of the nature, extent, and frequency of algal blooms in inland waters and nearshore Great Lakes. Since 2012, the WRD has been collecting and analyzing samples from 7 Lake Erie beaches, every-other-week from June through September. These data will be presented in separate reports. The need to understand and address HABs became more urgent in August 2014. Severe blooms were observed in the western basin of Lake Erie, and access to drinking water for hundreds of thousands of people served by the city of Toledo water treatment facility was temporarily interrupted due to elevated levels of an algal toxin associated with the bloom. This event caused the MDEQ-WRD to reexamine and expedite our efforts related to harmful algal blooms.

Historic monitoring in Michigan has found that microcystin concentrations in Michigan inland lakes are not typically very high across all lakes. Sarnelle et al. (2010) found that only 2 of the 77 inland lakes sampled by volunteers in August and September 2006 had microcystin concentrations greater than 20 µg/L. Recreational use of water with microcystin concentration greater than 20 ug/L is suggested to have high risk levels by the WHO. Rediske et al. (2007) also sampled 7 drowned-river mouth lakes in western Michigan in 2006 and did not find any

microcystin samples above 20 µg/L. During the United States Environmental Protection Agency's (USEPA), National Lake Assessments (NLA), in 2007 and 2012, no samples exceeded 10 µg/L (Kohlhepp, 2015[b]).

In 2015, the WRD expanded algal toxin monitoring to both targeted and randomly selected inland lakes and this report summarizes the results. This study was designed to allow the MDEQ to further understand: (1) the range of algal toxin concentrations across Michigan inland lakes; (2) how algal toxin concentrations change during a growing season in Michigan lakes; (3) if lake water chemistry parameters predictively correlate with algal toxin concentrations; and (4) how microcystin results compare using rapid field test strips and laboratory quantitative analysis.

# **Study Design**

To achieve the study objectives, WRD biologists collected qualitative algal bloom condition and water quality data at both randomly selected lakes and targeted inland lakes. The random sites were selected to represent a geographic range and to provide the ability to broadly understand conditions among Michigan's inland lakes during the summer growing season. The targeted lakes were selected because they are known to have high concentrations of nutrients and historic problems with algae blooms and were expected to represent some of the most productive lakes in Michigan.



Table 1. Michigan Department of Natural Resources (MDNR), Fisheries Division's (FD), randomly selected lakes sampled twice during summer 2015.

\*As part of this project, Round Lake was only sampled in August 2015.

The 22 randomly selected inland lakes (Table 1, Figure 1) included in this project were monitored in 2015 utilizing the MDNR-FD's and MDEQ-WRD's Status and Trends Programs. These lakes were sampled for HABs twice during the 2015 summer growing season; in July by MDEQ-WRD staff and in August by MDNR-FD staff. On both dates, field crews visually assessed whether an algal bloom was occurring in any portion of the lake, collected up to 4 surface water samples per lake, and used Abraxis test strips to estimate microcystin concentrations. General lake water chemistry samples were also collected at the center of the lake in July and August.



Figure 1. 2015 algal toxin monitoring locations. Lake Erie data are summarized in a separate report.

The targeted portion of this study identified lakes with a known history of cyanobacteria blooms that may produce harmful algal toxins. Ten lakes were identified with high potential for algal blooms and 7 of the lakes were sampled (Table 2, Figure 1). Lakes, with and without herbicide application permits, were intentionally chosen to capture possible relationships between herbicide treatment and microcystin concentrations. The MDEQ-WRD field crews sampled the targeted lakes when there was an algal bloom reported to the MDEQ by an arranged lake contact or when MDEQ staff visited a lake and observed a bloom.

Table 2. Targeted inland lakes with a history of algal blooms. Treated lakes had a current aquatic nuisance control permit with the MDEQ to apply herbicides for aquatic plant and algae management.

Lake	County	Sampled in 2015
Holloway Reservoir (untreated)	Genesee/Lapeer	
Lake Ovid (untreated)	Clinton	
Ford Lake (untreated)	Washtenaw	
Lake Macatawa (untreated)	Ottawa	
Lake Hudson (untreated)	Lenawee	
Morrison Lake (treated)	Ionia	
Jordan Lake (treated)	Barry	
Crockery Lake (treated)	Ottawa	
Mona Lake (treated)	Muskegon	
Bass Lake (treated)	Mason	

## Field Methods

Sampling occurred between late-June and early-September, with most monitoring occurring in July and August. Lakes were sampled repeatedly over varying time scales to assess the changes of algal toxins over the summer. During a monitoring event at a targeted lake, MDEQ-WRD staff took pictures of algal conditions, collected general water chemistry in the center of the lake, and collected water samples for algal toxin analysis from up to 4 locations around the lake. The algal toxin samples were analyzed using both Abraxis test strips to estimate microcystin concentration and High-Performance Liquid Chromatography with Mass Spectrometry (HPLC-MS) for quantitative assessment of a suite of algal toxins including microcystins, cylindrospermopsins, and anatoxins.

# *Survey Forms*

A field sheet was completed during every targeted lake survey to document shoreline algae levels and accumulation.

## *Water Samples - General Chemistry*

Water sample parameters collected at the Status and Trend lakes and the targeted lakes were generally similar, except for quantitative algal toxins. At all lakes, temperature, dissolved oxygen, conductivity, and pH were measured using a YSI sonde at the mid-lake location. At the same location as sonde sampling, water samples were collected for nutrient analyses and a secchi disk reading was taken.

At all lakes, surface water samples were collected at an approximate 1-foot depth using new 250 milliliter (ml) polypropylene sample bottles that were triple-rinsed with site water. At targeted lakes, the following samples were collected: total phosphorus, total Kjeldahl nitrogen, ammonia, nitrate+nitrite, ortho-phosphate, and chlorophyll *a*. The samples were analyzed at the MDEQ Environmental Laboratory (Table 3). At Status and Trend lakes the same nutrient samples were collected, excluding ortho-phosphate.

Following sampling, preservatives were added if required by MDEQ protocols and sample bottles were placed on ice or refrigerated for transport and storage prior to delivery to the laboratory. At targeted lakes, the nutrient samples were not collected at every sampling event if sampling occurred several times over a week. The August Status and Trend water chemistry samples were collected by MDNR-FD staff and analyzed by the Great Lakes Environmental Center.

#### *Water Samples - Algal Toxins*

At all lakes, 1 mid-lake sample and 3 shoreline samples were collected in 250 ml polyethylene terephthalate sample bottles. Water was collected in the top foot of water, at locations in the lake approximately 2-to 6-feet deep. Surface algae was neither targeted nor avoided while sampling. The shoreline sampling locations were distributed approximately evenly around the shoreline of the lake. However, downwind locations, bays that may be used for recreation, areas impacted by river outlets, or beaches were preferentially targeted.

Sample bottles were placed on ice or refrigerated for transport and storage. The quantitative cyanotoxin samples were frozen and batch shipped to the Wisconsin State Lab of Hygiene for HPLC-MS analysis of 8 algal toxins: anatoxin-a, homoanatoxin-a, cylindrospermopsin, deoxycylindrospermopsin, microcystin-LR, microcystin-YR, microcystin-RR, and microcystin-LA (Table 3).

Qualitative microcystin samples were held on ice or refrigerated for no more than 5 days prior to analysis. If microcystin samples were held longer than 5 days, they were frozen with care taken to reduce volume to allow for expansion. Qualitative microcystin samples were tested using Abraxis test strips (Product Number #52022) by either MDEQ-WRD or Great Lakes Environmental Center staff following Abraxis protocols. Abraxis microcystin test strips were selected for this project because the procedure includes a cell lysis step, which was more consistent with other MDEQ algal toxin monitoring for total microcystin (both free in the water column and intra-cellular). MDEQ-WRD staff analyzed the July Status and Trend samples and all targeted lake samples using the test strips. The August Status and Trend samples were analyzed by staff of the Great Lakes Environmental Center.



Table 3. Analytical methods and reporting limits.

# **RESULTS**

# *Status and Trend Lakes*

Over the course of the July and August 2015 sampling, 168 Abraxis microcystin test strips were run on discrete samples from the Status and Trend Lakes. All but 4 of the sample results were non-detect (less than 1 µg/L). All 4 samples collected in Rush Lake in July had positive test strip results for total microcystin in the 1 to 10 µg/L range. Although testing only occurred on 2 sampling dates, these results suggest that algal toxins are not routinely present at high levels in most Michigan lakes.

## *Targeted Lakes*

Targeted lake sample dates are presented in Table 4 and Appendix 1, along with herbicide treatment dates. Sampling began in late June to collect data before and after planned herbicide treatments on Jordan and Morrison Lakes. Sampling at each lake was initiated based on reports of blooms, planned herbicide treatments, or MDEQ staff visual assessment of algae blooms. Upon notification of a planned herbicide treatment, sampling was attempted before treatment and generally again after treatment.



Table 4. 2015 sample and permitted chemical treatment dates. Of the targeted lakes, only Crockery, Jordan, and Morrison Lakes were regularly treated with algal herbicides in 2015.

General lake chemistry in the targeted lakes is summarized in Table 5 and Appendix 1. The data collected are indicative of eutrophic to hypereutrophic lakes, as expected. Across all 7 lakes the total phosphorus concentration ranged from 0.019 to 0.220 milligrams per liter (mg/L). Chlorophyll *a* ranged from 7 to 350 µg/L. Total nitrogen ranged from 0.730 to 5.0 mg/L. Secchi depth ranged from 0.9 to 7.8 feet.

Table 5. Targeted lake chemistry data from June to September 2015. Total phosphorus, chlorophyll a, and total nitrogen are presented in µg/L and secchi depth is presented in feet. Total nitrogen was calculated as the sum of kjeldahl nitrogen and ammonia.



# *Test Strips*

Abraxis field test strip results for the targeted lakes are presented in Table 6. Out of 145 tests, only 10 (all from Mona Lake) had a result greater than 10 µg/L total microcystin. Crockery Lake, Mona Lake, and Lake Macatawa had some test results between 1 and 10 µg/L. Ford Lake, Jordan Lake, Lake Hudson, and Morrison Lake did not have any samples with detectable concentrations of microcystin using the field test strips. Over 20% of Mona Lakes samples exceeded 10 µg/L. Approximately 53% of Mona Lake samples were in the 1-10 µg/L range, while Crockery Lake and Lake Macatawa had approximately 40% and 23% of samples in the 1-10 µg/L range, respectively.



Table 6. 2015 targeted lake information and Abraxis field test results. Abraxis microcystin test strip results are presented as number of tests within ranges (non-detect  $= 0.1$  µg/L range; positive test results  $= 1-10$   $\mu$ g/L or greater than 10  $\mu$ g/L.)

# *HPLC-MS*

Quantitative analysis of 8 algal toxins (Table 3) was conducted using HPLC-MS by the Wisconsin State Laboratory of Hygiene. Total microcystin was calculated as the sum of the concentrations of 4 microcystin congeners (LR, YR, RR, and LA). Minimum and maximum algal toxin concentrations per lake are presented in Table 7. Microsystin LR, microcystin RR, and anatoxin-a were the most commonly quantified toxins. Each of the 8 algal toxins was quantified at least once in the 145 samples.

Seven samples, collected from Hudson, Jordan, and Morrison Lakes, did not have quantifiable concentrations of any toxin. Twenty-eight samples did not have quantifiable levels of any of the 4 microcystin congeners; however, 10 of these had quantifiable levels of deoxycylindrospermopsin, and 13 had quantifiable levels of anatoxin-a.

Table 7. 2015 HPLC-MS algal toxin data summary of 145 inland lake samples. All data are presented in µg/L. The minimum for each parameter is not quantified (NQ); the lowest quantified value is presented in the minimum row in parentheses. Total microcystin (MC) is a sum of the 4 microcystin congeners (LR, YR, RR, and LA). For reference, various health guidelines for drinking water and recreational water use are included in the bottom portion of the table. These data were collected from the following USEPA Web sites: *(The link provided was broken and has been removed)* and



https://www.epa.gov/sites/default/files/2017-06/documents/microcystins-report-2015.pdf.

The 2 samples with quantifiable concentration of cylindrospermopsin also had higher concentrations of microcystin LR, RR, and LA. All 3 samples with quantifiable concentrations of microcystin LA had higher concentrations of total microcystin (9.4, 29.6, and 193 µg/L). Based on these limited data, it appears that these toxins may only be present when algal toxin production is higher.

The concentrations of the cylindrospermopsins and anatoxins were all low compared to all available drinking and recreational health values (Table 7). Only 3 samples had total microcystin concentrations greater than the WHO provisional recreation microcystin LR guidance value of 20 µg/L. All of these samples were collected in Mona Lake (Figure 2).

The total concentrations of the 4 microcystin toxins are presented in Figure 2. The concentrations in Jordan, Morrison, Macatawa, Ford, and Hudson Lakes were all less than 2 µg/L. Concentrations of total microcystin in Crockery Lake were all less than 4 µg/L. All of the samples collected in Mona Lake had quantifiable concentrations of total microcystin and only 2 samples did not have concentrations of anatoxin-a. Although total microcystin was consistently present in Mona Lake, the concentrations were not consistently high; 14 samples out of 47 (30%) exceeded 4.0 µg/L, 6 samples (13%) exceeded 10 µg/L, and only 3 (6%) exceeded 20 µg/L.



concentrations) from 7 targeted lakes. Note: Concentration scale varies on each graph.

## *Comparisons*

## *HPLC-MS vs Abraxis Test Strips*

The test strip data are presented in Table 6 as binned concentrations; non-detect (assumed to be less than 1  $\mu q/L$ ), detectable but less than 10  $\mu q/L$ , and greater than 10  $\mu q/L$ . Using the Abraxis test strips, it is possible to estimate a value between 1 and 10, but there is a good amount of user interpretation required. Generally, the MDEQ-WRD will use the categorical data for assessing microcystin concentrations, but we did also estimate a specific value within the 1 to 10 µg/L range and are using those estimates in this comparison analysis.

The total microcystin results using Abraxis test strips and HPLC-MS are moderately correlated (Figure 3) when using the samples with HPLC-MS results less than 10  $\mu$ g/L (R<sup>2</sup> of 0.6). Because the resolution of the test strip ends at 10 µg/L, including the test results with

microcystin concentrations greater than 10  $\mu$ g/L, the R<sup>2</sup> value is reduced to 0.45 when the 5 HPLC-MS samples between 11 and 40 ug/L are included.

Overall, the data show that the test strips do not always accurately detect very low concentrations of microcystin, between approximately 4 and 9 µg/L, the test strips appear to overestimate the concentration of microcystin, and the test strips accurately characterized the sites with HPLC-MS concentrations greater than 10 µg/L in 6 of 10 samples. The 10 Abraxis test strips that showed microcystin concentrations of 10 ug/L or greater had corresponding HPLC-MS results ranging from 5 to 193 µg/L, 4 of which were less than 10 µg/L and 6 were greater than 10 µg/L.



Figure 3. Abraxis test strip microcystin results compared to HPLC-MS microcystin results. Line is the  $1:1$  line.

# HPLC vs water chemistry

A variety of standard water chemistry parameters were collected on most sampling trips, but not all. Generally, these parameters were only collected at 1 station, usually the center of the lake. To examine the relationship between water chemistry components and microcystin concentrations, the average microcystin concentration on a date (across 4 samples) was compared to total phosphorus, ortho-phosphorus, total nitrogen, chlorophyll a, and secchi depth.

Table 8. The correlation coefficients (r) of the average total microcystin concentration (across 4 samples) and water chemistry data. Total nitrogen was calculated as the sum of Kjeldahl nitrogen and ammonia.



When using data from all of the lakes, there are no strong correlations between water chemistry and microcystin levels. Looking at Mona and Crockery Lakes independently, the 2 lakes that had more than 5 sample events and higher concentrations of microcystin, there are some stronger correlations between water chemistry and microcystin concentration (Table 8 and Figure 4). Mona Lake shows some positive correlations with total phosphorus, chlorophyll a,

and total nitrogen. Crockery Lake has strong positive correlations with total phosphorus, ortho-phosphorus, chlorophyll a, total nitrogen, and secchi depth.

The correlations in Mona Lake are driven by the heavy weight of the 1 high microcystin concentration >50 µg/L. The high concentration of microcystin occurred when there were also high concentrations of phosphorus, chlorophyll *a*, and nitrogen and shallow secchi depth, but the overall data are not as strongly correlated absent 1 high data point.



Figure 4(a). Total phosphorus (mg/L) and chlorophyll *a* vs average microcystin (µg/L) concentrations. Each data point is 1 sample date. The top graphs present the data for all lakes, followed by the data for Crockery and Mona Lakes. NOTE: One outlier chlorophyll a data point (Morrison Lake: 350 µg/L chlorophyll *a* and 0.26 µg/L total microcystin) was removed from the all lakes graph.



Figure 4(b). Total nitrogen (mg/L) and secchi depth (feet) vs average microcystin (µg/L) concentrations. Each data point is 1 sample date. The top graphs present the data for all lakes, followed by the data for Crockery and Mona Lakes.

Conditions in Crockery Lake changed in late July (Figure 2). In mid-July, the lake had a heavy algal bloom with microcystin concentrations between 2.0 and 4.0 µg/L and associated higher concentrations of phosphorus, nitrogen, and chlorophyll *a*, and shallower Secchi depths. All of the samples collected from late July through early September had microcystin concentrations in the 0 to 1 range, with associated lower concentrations in nutrients, chlorophyll *a*, and greater Secchi depths. The reduction in algae and algal toxins in Crockery Lake may have been caused by weather events or lake management treatments or a combination of several factors. The lake was treated with herbicides on 5 dates, including July 21 and August 4 to reduce algal growth. It is not clear if a single factor caused the apparent reduction in algal biomass from the data collected in the study.

## *Shoreline versus center toxin samples*

Algal toxin concentrations have the potential to be greater along the shoreline than the center of a lake (Sarnelle and Wandell, 2008). Elevated toxin shoreline concentrations may be occur when algal scums accumulate along the shoreline, or in bays or canals, due to wind and lake water movement. The lakes sampled for targeted monitoring in 2015 had higher nutrient concentrations and more algae (as seen through higher chlororphyll *a* concentrations) than the lakes in Sarnelle and Wandell (2008). Although the 8 highest total microcystin samples in 2015 were collected from shoreline samples, there was not a statistically significant difference in the medians of the shoreline versus the lake center microcystin samples (Mann Whitney U test P value 0.39). Because algae was present at higher concentrations and more distributed in the water column in the 2015 targeted lakes, as compared to the Cooperative Lakes Monitoring Program lakes, there may have been more homogeneous toxin concentrations than in less productive lakes. Heiskary et al. (2014) also found that there was no meaningful difference between center and shoreline microcystin concentration in the 2007 and 2012 Minnesota NLA lakes.

# **DISCUSSION**

Monitoring data collected in 2015 from Michigan inland lakes with known histories of algal blooms, found microcystin concentrations (assessed using HPLC-MS) only exceeded the WHO recreation target of 20 ug/L in 3 out of 145 samples. Anatoxin concentrations were all less than 3.1 µg/L and cylindrospermopsin concentrations were all less than 0.1 µg/L, both well below any associated guidelines (Table 7).

Microcystin concentrations, when present in the targeted lakes, did not show consistent patterns over the summer (Figure 2). Several lakes had consistently low or no levels of microcystin, including the Ford, Macatawa, Hudson, and Jordan Lakes. Mona Lake had the highest concentrations of microcystin, but also had high variability at different locations on a given date and across sample dates. Crockery Lake microcystin concentrations were more consistent across sites and showed a decrease over the course of the summer, possibly due to algaecide treatments or weather events.

Across the targeted lakes, there was no broad relationship between water chemistry parameters and microcystin concentrations. However, in Crockery Lake when the microcystin concentrations decreased, there was a corresponding decrease in nutrients and chlorophyll *a* and an increase in Secchi depth. These data indicate there likely was not only a reduction in toxin production within algal cells, but that the amount of algae suspended in the water column decreased.

Using HPLC-MS analysis for algal toxins is advantageous for generating precise quantitative data for many algal toxins. However, the cost and time lag between sample collection and results can be a drawback when you need the results to inform recreation risks in a lake or to make decisions on continued monitoring. The Abraxis test strips used in this study did not consistently detect low concentrations of microsystin and overestimated microcystin in samples at concentrations greater than 4 µg/L of total microcystin, which makes them a conservative screening tool. One potential drawback to using test strips that detect only 1 algal toxin is that it may suggest no health risks for using a body of water after a low or negative result for microcystin, but does not account for other toxins such as anatoxin. In Minnesota, in 2015, there were dog illnesses, dog deaths, and human illness associated with HABs, but follow-up

water chemistry did not always confirm high microcystin concentrations and in 1 dog death anatoxin was found to be the likely cause (Heiskary, 2016).

Michigan, like other states in the upper midwestern United States, has found that algal toxins are produced in inland lakes at variable concentrations (Graham et al., 2004; Graham and Loftin, 2014). In Minnesota, microcystin monitoring in 2004-2007 showed that 80% of the 133 samples were less than 10 µg/L (Linden and Heiskary, 2009), while Wisconsin found that approximately 70% of 43 samples collected from targeted lakes in 2009-2013 were less than 10 µg/L (LaLiberte, 2014). The data collected in 2015, from 22 Status and Trend and 7 targeted lakes, lend support to previous studies (Sarnelle et al., 2010) that found that Michigan inland lakes do not regularly have high concentrations of toxins, although unpredictable and rare occurrences of high concentrations have been, and will be, detected in Michigan lakes.

MDEQ monitoring in 2016 will include sampling lakes with expected, or reported, large algal blooms to further understand the magnitude of toxin levels and how toxins vary throughout and across summers. Microcystin monitoring will continue in randomly selected Status and Trend Program lakes to provide more data on statewide HAB conditions. In 2016, phycocyanin monitoring and dominant algal species identification will provide more information on algae bloom composition. To date, monitoring in Michigan does not indicate that algal toxins are a wide-spread problem in Michigan's inland lakes. However, at any point in time a localized problem can occur and it is warranted to recommend that people limit exposure of themselves, children, pets, livestock, and irrigated crops to water that is very green or very hard to see through because of the risk of algal toxins.



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