

# **Blue-green algae in northern Indiana lakes: An analysis of the algal toxin, microcystin, over 2010-2013 in lakes of Kosciusko County, Ind.**

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## ***Executive Summary***

One of the most alarming current human health concerns related to lakes is the presence of blue-green algae toxins. Health guidelines have been established which are based on concentrations of the common blue-green algae toxin called microcystin as well as on algae counts which quantify and identify individual types of algae and groups such as blue-green algae. Alarming results from a small 2010 pilot study including a few Kosciusko County lakes prompted the Center for Lakes & Streams at Grace College to launch a comprehensive research project to determine the extent and severity of the common blue-green algae toxin, microcystin, in local lakes. Observed microcystin concentrations over 2010-2013 indicate that microcystin does not pose a consistent health threat at the present time in Kosciusko County lakes. However, algae count results may indicate higher human health threats from other blue-green algae toxins which were not tested for in the present study. Furthermore, our research indicates that microcystin detections are more common but currently at lower levels in Kosciusko County lakes compared to national and regional data which demonstrates a potential for major microcystin problems in the future under the right conditions. Visual cues of blue-green algae (such as water clarity as measured by secchi disk) are helpful but not reliable indicating that a better method of determining the safety of the water is necessary. A unique pigment to blue-green algae, phycocyanin, can be measured by a simple hand-held device in the field and was found to be the most promising predictor for both microcystin and algae counts. The present study warrants further research with Kosciusko County lakes in order to protect human health, including ongoing monitoring, testing of new prediction techniques, and exploration of additional toxin threats.

**Keywords:** blue-green algae, Kosciusko County, nutrients, streams, lakes, microcystin

## ***Introduction***

One of the most alarming current human health concerns related to lakes is the presence of blue-green algae toxins. Funari and Testai (2008) review the state of present knowledge in the area of blue-green algae toxins as a human health threat. They describe several different classes of toxins which can be hepatotoxins (affecting liver), neurotoxins (affecting nervous system), cytotoxins (affecting specialized cells of the body), gastrointestinal toxins (affecting digestive system), and skin irritants. Research has shown that high levels of these toxins can be lethal to humans, and emerging research is indicating that chronic low-level toxin exposure can also lead to terminal health problems. Toxin exposure can come from less serious skin exposure to more serious ingestion (Funari and Testai, 2008). Research on two lakes in California has even shown that exposure can occur through inhalation of toxin aerosols near lakes with high blue-green algae populations (Backer et al., 2009). More recent research by Manganelli et al. (2010) has also demonstrated that more toxic blue-green algae can live farther below the water surface making visual detection difficult.

To protect human health, the World Health Organization and the Indiana Department of Environmental Management have set human health guidelines related to blue-green algae in recreational waters. These guidelines are based on concentrations of the common blue-green algae toxin called microcystin as well as on algae counts which quantify and identify individual types of algae and groups such as blue-green algae. The guidelines prescribe low, medium, and high health risks based on either microcystin or blue-green algae counts, and they suggest precautions people should take based on each health risk category. At high risk levels, for example, both organizations suggest avoiding contact with the water.

Blue-green algae toxins have been identified as a human health problem recently in Kosciusko County. In 2007, the U.S. EPA conducted an assessment of the nation's lakes and found detectable levels of algal toxins in 68 percent of Indiana lakes, compared to the national average of 32 percent. A pilot study including seven Kosciusko County lakes was conducted in September 2009 and found moderate blue-green algae toxin levels in four of the lakes sampled and high levels in Lake Wawasee according to guidelines published by the World Health Organization and the Indiana Department of Environmental Management.

Early research by the Center for Lakes & Streams, partially funded by K21 Health Foundation, in 2010 and 2011 found detectable levels of blue-green algae toxins in 40 of the 44 lakes sampled. Four lakes, Big Chapman, Dewart, Muskelunge, and Wawasee, had toxin levels above the Indiana Department of Environmental Management safety threshold, advising the public to reduce recreational contact with water from those lakes. At that time, the center found some promising results with predicting toxin levels and potentially reducing these levels in the future, but these relationships were inconsistent among some lakes and prompted further study.

The present study focuses on the human health threat of blue-green algae through the production of the common toxin microcystin in the lakes of Kosciusko County. Data from 2010-2013 was used in this analysis, including in-lake measurements at all depths, inflowing stream measurements, and near shore and open lake sampling. The goals of the study were to accurately characterize the human health threat of microcystin in local lakes and explore ways to predict toxin occurrence. Part of this study included identification and quantification of

algae species in a small subset of the lakes to further aid in characterization and prediction of microcystin threats.

## **Methods**

### **Study area**

Kosciusko County contains 104 lakes greater than 5 acres in surface area (Figure 1). Most of these lakes are glacially formed and have varying lake bottom substrates (see [www.lakes.grace.edu](http://www.lakes.grace.edu) for individual lake details). All lakes with a surface area greater than 25 acres were selected for near shore sampling in the present study by the Center for Lakes & Streams at Grace College (Table 1). The 13 all-sports lakes had two sampling sites – one near shore and one off shore. The near shore sampling sites were at representative swimming areas. The off shore, or open lake, sampling sites were at the deepest point of the lake. Syracuse Lake had two public swimming beaches, so both of these near shore sites were sampled as well as an off shore site.

### **Field sampling**

Lake sampling was conducted at monthly intervals during August-September in 2010 and May-September in 2011 and then at biweekly intervals during May-September in both 2012 and 2013. All lakes were sampled within the same week to allow comparisons between lakes at a given time.

Near shore sampling occurred at all 44 lakes included in the study (Table 1), including two near shore sites at the public swimming beaches of Syracuse Lake. At each site a Secchi depth measurement was made to determine water clarity. Next, dissolved oxygen, conductivity, pH, and water temperature were measured at the surface, 0.5 m, and 1 m depths using a Hydrolab Quanta meter. Finally, an integrated water sample from the surface to bottom was taken using a vertical Van Dorn sampler. This integrated water sample was used for the blue-green algae toxin (microcystin) samples for later lab analysis as well as blue-green algae pigment (phycocyanin) analyses (only during 2013) using a Turner Designs AquaFluor fluorometer in the field.

Open lake sampling occurred at thirteen all-sports lakes included in the study (Table 1). At each site a Secchi depth measurement was made to determine water clarity. Next, dissolved oxygen, conductivity, pH, and water temperature were measured at each meter from the surface to the lake bottom. An integrated water sample from the surface to 2 m depth was taken using a vertical Van Dorn sampler. This integrated water sample was used for the blue-green algae toxin (microcystin) samples for later lab analysis as well as blue-green algae pigment (phycocyanin) analyses (only during 2013) using a Turner Designs AquaFluor fluorometer in the field. In 2012 and 2013, this integrated water sample was also used for algae count samples for Dewart, James, Wawasee, Winona, and Yellow Creek lakes which were collected for later lab analysis.

Beginning in 2011, a multi-parameter sample was added which included suspended solids, conductivity, chloride, fluoride, sulfate, silicon dioxide, total phosphorus (TP), soluble reactive phosphorus, total nitrogen (TN), total Kjeldahl nitrogen, ammonia, nitrate, and nitrite.

Samples were taken at open lake sites at a depth of 1 m below the lake surface and 1 m above the lake bottom using a vertical Van Dorn sampler.

Stream sampling occurred in the week preceding lake sampling during 2012 and 2013 for inflowing streams to Dewart, James, Wawasee, Winona, and Yellow Creek lakes (Table 2). A multi-parameter sample was taken from the center of the stream which included same parameters as listed above for lake samples. Next, dissolved oxygen, conductivity, pH, and water temperature were measured in the center of the stream using a Hydrolab Quanta meter. Total stream flow was calculated from measurements across the entire cross section of the stream with an OTT MF Pro Flow Meter.

## **Lab analysis**

Water samples obtained in the field for later lab analysis were stored in the dark near 2° C until returning to Grace College each day. Microcystin, multi-parameter, and algae count samples were kept at 2° C. All samples were transported to the National Center for Water Quality Research at Heidelberg University for analysis within 7 days of field sampling.

## **Data analysis**

Data was collected in the field and immediately recorded electronically throughout project to avoid any loss of data. Data was always checked twice for accuracy when transferred from field log sheets to computer spreadsheets. Several microcystin samples yielded concentrations below detection limits of 0.15 ppb. To aid in data summarization and statistical analyses, these data values were recorded as 0.075 ppb.

## ***Results and Discussion***

### **Algae toxin (microcystin)**

Results for the blue-green algae toxin microcystin showed several interesting patterns and some sporadic and unexpected levels in certain lakes. Over the four-year study, there were 1,068 samples collected for microcystin analysis. Of these, 468 (44%) samples had detectable levels of microcystin found in them. By comparison, a 2007 study of 1,028 lakes across the U.S. found that 38% and 23% of lakes had detectable levels of microcystin nationally and in the Upper Midwest region, respectively (EPA, 2009). Kosciusko County lakes in the present study had an average microcystin concentration of 0.32 ppb, while the 2007 nation-wide study found averages of 1.00 ppb nationally and 0.56 ppb in the Upper Midwest region. From these overall microcystin results, we find that microcystin is more common but at lower levels in Kosciusko County lakes compared to other lakes in our region or nationally.

Average microcystin levels in Kosciusko County lakes varied over time during the four-year study. There was wide variation among the years of the study (Table 3). It was expected that 2012 would be lower compared to other years since it was a drought year which would lead to stream nutrient loads to these lakes being relatively small in 2012. There is also variation among the five months that were sampled for this study (Table 4). June and July microcystin concentrations were approximately double those observed in the other three months on average.

Microcystin levels also widely varied across lakes during the study period. Among the 13 all-sport lakes, Lake Wawasee and Dewart Lake had the highest average microcystin concentrations at 0.67 and 0.66 ppb, respectively, while Winona and Camelot lakes had the lowest average concentrations at 0.10 ppb for both lakes (Figure 2). Low toxin levels in Camelot was expected due to the low productivity of that lake in general and the lack of inflowing streams to bring in nutrients to promote algae growth. However, lower toxin levels in Winona compared to Dewart and Wawasee was surprising as the latter two lakes are typically less productive with relatively less nutrient inputs from inflowing streams and thus lower productivity of algae and other plants.

The Indiana Department of Environmental Management has set a threshold of 4 ppb of microcystin for reducing recreational contact with water for human health concerns. The World Health Organization has set a threshold for moderate health risk of 10 ppb and high health risk of 20 ppb. Lake Wawasee had a toxin concentration of 63 ppb in 2009 which prompted the present study. During 2010-2013, there were seven lakes which exceeded this 4 ppb threshold briefly – Big Chapman (4.5 ppb in July 2011), Dewart (5.9 ppb in June 2011), Diamond (5.3 ppb in May 2011), McClure's (4.2 ppb in May 2013), Muskellunge (12 ppb in August 2010), Syracuse (5.0 ppb in June 2013), and Wawasee (5.1 ppb in July 2011). According to the World Health Organization guidelines, only Muskellunge Lake was in the moderate health risk range, and none of the 1,068 samples taken during the 2010-2013 study period were in the high health risk range.

Microcystin concentrations were tested against several potential predictor variables with varying success. Relationships varied widely among lakes, indicating additional factors or combinations of factors were influencing blue-green algae toxins which were not included in the present study. For example, stream inflow measurements and most in-lake variables had no correlation with microcystin levels in the lakes, but there appears to be some influence of nitrate and phycocyanin concentrations in the epilimnion, dissolved oxygen in the hypolimnion, and Secchi disk measurements. Results from 13 all-sports lakes where near surface nitrate concentrations were measured show that there seems to be a higher likelihood of elevated microcystin levels with lower nitrate levels in these lakes (Figure 3). This is contrary to findings from a recent experimental lab study which showed blue-green algae produce less toxin at low nitrate concentrations (Harke and Gobler, 2013). New work by Molot et al. (2014) demonstrates that blue-green algae are controlled by ferrous iron coming from anoxic sediments since these algae have the ability to migrate down near the bottom to acquire this form of iron. Data from the present study on Kosciusko County lakes supports this mechanism as elevated microcystin concentrations consistently follow near-zero dissolved oxygen levels at the lake bottoms. Secchi disk measurements seem to show a threshold of 8 ft such that there are no instances where microcystin levels were over 2 ppb when Secchi disk measurements were greater than 8 ft (Figure 4).

The most promising predictor variable for microcystin from the present study was phycocyanin as quantified by the hand-held fluorometer in the field. Statistical analysis of the data using Partial Least Squares Regression showed that phycocyanin has a predictive nature to microcystin but that more data was needed to confirm this relationship. This relationship seems to be lake specific, and results are particularly promising for certain lakes like Beaver Dam Lake with an  $R^2$  of 84.8% (Figure 5). These results are as expected since blue-green algae

uniquely contain this phycocyanin pigment which would lead to a good relationship between these two variables. Researchers in Ohio have found phycocyanin as a good indicator of microcystin threats as well (Marion et al., 2012).

## Algae counts

Compared to the blue-green algae toxin results for these Kosciusko County lakes, the algae count results are potentially more concerning for human health considerations. Of the 92 algae count samples taken over 2012 and 2013, 90 (98%) of the samples contained blue-green algae. For reference, the Indiana Department of Environmental Management and the World Health Organization both consider 20,000-100,000 cells/ml of blue-green algae in the moderate human health risk zone and over 100,000 cells/ml as high risk for human health. Using these guidelines, the number of samples in the present study falling into the moderate or high risk categories together was 38 (41%) and the national reference from the 2007 EPA study was only 27% (EPA, 2009). Clearly, blue-green algae are more common at higher health risk levels in Kosciusko County lakes compared to national statistics.

Like microcystin results, algae counts are sporadic over time. The average blue-green algae count across the five lakes where measurements were taken was 51,014 cells/ml for 2012 and increased to 241,066 cells/ml for 2013. Algae counts were more than over four times higher in 2013 compared to the previous drought year of 2012. Monthly averages also show wide variation of strongly increasing blue-green algae counts over May thru September (Table 5).

There was also wide variation even among the subset of five lakes where algae counting was done (Figure 6). Winona Lake had the most high risk blue-green algae counts of the five lakes, and Wawasee and Dewart lakes only had moderate risk counts over 2012 and 2013. In fact, Winona Lake samples were in moderate or high health risk category 56% of the time. Both Winona and Yellow Creek lakes had blue-green algae cell counts of over 1,000,000 cells/ml in September 2012. These results show that microcystin concentrations and blue-green algae counts are not necessarily related. For example, Lake Wawasee has a much higher average microcystin concentration than Winona Lake even though Winona Lake has a much higher incidence of high blue-green algae counts. The reason for this is seen in the specific types of algae that are found in each of these lakes. Even though Lake Wawasee has a relatively low blue-green algae count, the dominant algae during the higher microcystin concentration that we found was *Microcystis aeruginosa* which is a known microcystin producer. However, Winona Lake has high blue-green algae counts with low microcystin concentrations because three of the four dominant algae species do not even produce microcystin – they produce other toxins that were not measured in this present study (Figure 7).

Algae count results were also tested against several other variables with varying success. Algae counts were somewhat surprisingly not a good predictor for microcystin. This is reasonable upon further analysis since blue-green algae make several other toxins in addition to microcystin, and certain species of blue-green algae cannot make microcystin at all. To predict blue-green algae counts, water clarity as measured by Secchi disk was helpful in some cases. Overall, there seems to be a threshold relationship where Secchi disk measurements over 10 ft leave little likelihood for moderate risk based on blue-green algae counts and World Health Organization guidelines, and Secchi disk measurements over 5 ft leave little likelihood

for high risk based on blue-green algae counts (Figure 8). For Winona Lake specifically, Secchi disk measurements do not seem to be a good predictor of blue-green algae counts (Figure 9). Nutrient ratios based on nitrogen to phosphorus (N:P) showed a range from 30-60 where blue-green algae counts had the potential to achieve high levels (Figure 10). This result does not support previous work done by Smith (1983) who found global blue-green algae populations were controlled by N:P ratios such that blue-green algae were rare when  $N:P > 29$ . More recent work by Downing et al. (2001) suggests that N:P ratios are not as important in controlling blue-green algae populations compared to nutrient concentrations and total algal biomass, but our data suggests that N:P ratios are the most important of these variables to blue-green algae counts. These comparisons across studies indicate that these relationships are likely lake- and region-specific.

The most promising predictor variable for blue-green algae counts from the present study was phycocyanin as quantified by the hand-held fluorometer in the field (Figure 11). With more data to allow for these relationships to be developed for individual lakes, this has strong possibilities for predicting blue-green algae counts in local lakes.

## **Implications**

These research results have several important implications for Kosciusko County and other Midwestern U.S. counties with lakes. First, observed microcystin concentrations indicate that microcystin does not pose a consistent health threat at the present time in Kosciusko County lakes. However, algae count results may indicate higher human health threats from other blue-green algae toxins which were not tested for in the present study. Furthermore, our research indicates that microcystin detections are more common but currently at lower levels in Kosciusko County lakes compared to national and regional data, which demonstrates a potential for major microcystin problems in the future under the right conditions.

Next, this study confirms that prediction of blue-green algae counts or microcystin concentrations is difficult but potentially possible in the future. Visual cues of blue-green algae (such as water clarity as measured by Secchi disk) are helpful but not reliable. The unique pigment in blue-green algae, phycocyanin, is the most promising predictor for both microcystin and algae counts, although more data will need to be collected to confirm this relationship.

The present study also demonstrates that each lake needs to be studied and managed separately. Data collected to date on Kosciusko County lakes show wide variation in algae counts and microcystin levels that do not correlate well with the predictor variables that were tested across all the lakes together. Solutions to human health problems in one lake may not be valid for another adjacent lake, and it is not enough to just study one lake in the county to get a general synopsis of Kosciusko County lakes overall.

Finally, the present study implies that further work needs to be done with Kosciusko County lakes in order to protect human health. These lakes need to be monitored with an ongoing effort, new prediction techniques need to be tested, and additional toxin threats need to be explored.

## **Future work**

This research has prompted several immediate actions. First, the Center for Lakes & Streams at Grace College is sending a lab technician for training to give Grace College the algae counting capacity on campus for quick response to algae health threats. Second, the Kosciusko Health Department has already acquired laboratory equipment with the capacity to test for several toxins locally, including microcystin and other toxins known to be produced by types of algae identified in the present study in our lakes. Finally, the center will work with the K21 Health Foundation and other stakeholders to notify the general public about these research findings and to further educate the public about blue-green algae.

In addition to these immediate actions, this present study also demonstrates that additional monitoring and research is needed in Kosciusko County lakes into the future. Some level of microcystin monitoring with rapid results is critical going forward to alert county residents of potential health threats. A focused study on 5-10 lakes to further test the predictive ability of the phycocyanin pigment using a hand-held fluorometer is warranted based on the promising results in the present study. With the discovery of high algae counts in lakes like Winona and Yellow Creek that are made up of species known to produce additional toxins beyond microcystin, an exploratory survey of a broad cross-section of lakes in Kosciusko County is likely needed to test for additional blue-green algae toxins which may be present.

## ***Conclusion***

Observed microcystin concentrations over 2010-2013 indicate that microcystin does not pose a consistent health threat at the present time in Kosciusko County lakes. However, algae count results may indicate higher human health threats from other blue-green algae toxins which were not tested for in the present study. Furthermore, our research indicates that microcystin detections are more common but currently at lower levels in Kosciusko County lakes compared to national and regional data, which demonstrates a potential for major microcystin problems in the future under the right conditions. Visual cues of blue-green algae (such as water clarity as measured by Secchi disk) are helpful but not reliable. The unique pigment in blue-green algae, phycocyanin, as measured by a hand-held fluorometer in the field is the most promising predictor for both microcystin and algae counts. The present study warrants further research with Kosciusko County lakes in order to protect human health, including ongoing monitoring, testing of new prediction techniques, and exploration of additional toxin threats.

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## Tables

Table 1. Lake sampling sites included in present study.

Lake Name	Site ID	Site Type	Latitude (degrees)	Longitude (degrees)	Lake Size (acres)
Backwater Lake	BAC	Near shore	41.31615	-85.67	189
Beaver Dam Lake	BEA1	Near shore	41.09133	-85.972	146
Beaver Dam Lake	BEA2	Open lake	41.09157	-85.9752	146
Big Barbee Lake	BBA1	Near shore	41.28802	-85.7024	304
Big Barbee Lake	BBA2	Open lake	41.28647	-85.7063	304
Big Chapman Lake	BCH1	Near shore	41.2875	-85.7784	414
Big Chapman Lake	BCH2	Open lake	41.28149	-85.7937	414
Boner Lake	BON	Near shore	41.43108	-85.7209	40
Caldwell Lake	CAL	Near shore	41.12424	-85.9023	45
Camelot Lake	CAM1	Near shore	41.41694	-85.7934	30
Camelot Lake	CAM2	Open lake	41.41126	-85.7934	30
Carr Lake	CAR	Near shore	41.15819	-85.8603	79
Center Lake	CEN	Near shore	41.2431	-85.8555	120
Crystal Lake	CRY	Near shore	41.23516	-85.9884	68
Dewart Lake	DEW1	Near shore	41.37385	-85.7827	551
Dewart Lake	DEW2	Open lake	41.37209	-85.7759	551
Diamond Lake	DIA	Near shore	41.10541	-85.9338	79
Goose Lake	GOO	Near shore	41.18977	-85.883	28
Hill Lake	HIL	Near shore	41.10752	-85.9088	70
Hoffman Lake	HOF	Near shore	41.27538	-85.99	180
Irish Lake	IRI	Near shore	41.29276	-85.7331	182
James Lake	JAM1	Near shore	41.31745	-85.7231	282
James Lake	JAM2	Open lake	41.32078	-85.7259	182
Kuhn Lake	KUH	Near shore	41.28099	-85.6942	137
Lake Tippecanoe	TIP1	Near shore	41.32446	-85.742	768
Lake Tippecanoe	TIP2	Open lake	41.33408	-85.7688	768
Lake Wawasee	WAW1	Near shore	41.38576	-85.6848	3410
Lake Wawasee	WAW2	Open lake	41.24249	-85.4251	3410
Little Barbee Lake	LBA	Near shore	41.28867	-85.7205	74
Little Chapman Lake	LCH	Near shore	41.27188	-85.787	121
Loon Lake	LOO	Near shore	41.08316	-85.9666	40
McClure's Lake	MCL	Near shore	41.08123	-85.9511	34
Muskellunge Lake	MUS	Near shore	41.17076	-85.853	32
Oswego Lake	OSW1	Near shore	41.3299	-85.7837	83
Oswego Lake	OSW2	Open lake	41.32506	-85.7856	83

Lake Name	Site ID	Site Type	Latitude (degrees)	Longitude (degrees)	Lake Size (acres)
Palestine Lake	PAL	Near shore	41.16908	-85.9397	290
Papakeechee Lake	PAP	Near shore	41.3716	-85.6667	189
Pike Lake	PIK	Near shore	41.24788	-85.8421	230
Ridinger Lake	RID	Near shore	41.26077	-85.6644	141
Rock Lake	ROC	Near shore	41.04129	-85.9761	56
Sawmill Lake	SAW	Near shore	41.30028	-85.7294	56
Sechrist Lake	SEC	Near shore	41.29752	-85.7169	105
Sellers Lake	SEL	Near shore	41.15295	-85.7474	32
Shock Lake	SHK	Near shore	41.35548	-85.6928	32
Shoe Lake	SHO	Near shore	41.3065	-85.7495	43
Silver Lake	SIL	Near shore	41.07897	-85.8988	102
Spear Lake	SPE	Near shore	41.35709	-85.6648	40
Stanton Lake	STA	Near shore	41.32293	-85.7699	30
Syracuse Lake	SYR1A	Near shore	41.42664	-85.747	414
Syracuse Lake	SYR1B	Near shore	41.43001	-85.7493	414
Syracuse Lake	SYR2	Open lake	41.42848	-85.7379	414
Waubee Lake	WAU	Near shore	41.39492	-85.8357	187
Webster Lake	WEB1	Near shore	41.32259	-85.6932	585
Webster Lake	WEB2	Open lake	41.32858	-85.6933	585
Winona Lake	WIN1	Near shore	41.22225	-85.8224	562
Winona Lake	WIN2	Open lake	41.22232	-85.8306	562
Yellow Creek Lake	YEL1	Near shore	41.10729	-85.9521	150
Yellow Creek Lake	YEL2	Open lake	41.1037	-85.9573	150

Table 2. Stream sampling sites included in present study.

Stream Name	Site ID	Lake Connection	Latitude (degrees)	Longitude (degrees)
Cherry Creek	CHE	Winona Lake	41.22136	-85.8207
Keefer-Evans	KEE	Winona Lake	41.21385	-85.8231
Peterson Ditch	PET	Winona Lake	41.21259	-85.8289
Diamond Creek	DIA	Yellow Creek	41.10406	-85.9451
Yellow Creek Inflow	YCI	Yellow Creek	41.10812	-85.9559
Cable Run	CAB	Dewart Lake	41.35754	-85.7492
Tippecanoe River	TIP	James Lake	41.31472	-85.7143
Papakeechee Outlet	PAP	Lake Wawasee	41.37658	-85.6706
Turkey Creek Inflow	TUR	Lake Wawasee	41.38268	-85.6670
Dillan Creek	DIL	Lake Wawasee	41.39501	-85.6617
Launer Creek	LAU	Lake Wawasee	41.40445	-85.6638

Table 3. Average microcystin (MC) concentrations (in ppb) for each year of the four-year study across all Kosciusko County lakes.

Year	MC (ppb)
2010	0.37
2011	0.47
2012	0.18
2013	0.33

Table 4. Average microcystin (MC) concentrations (in ppb) by month during the four-year study across all Kosciusko County lakes.

Month	MC (ppb)
May	0.23
June	0.48
July	0.45
August	0.27
September	0.22

Table 5. Average blue-green algae counts (in cells/ml) by month during 2012 and 2013 for Dewart, James, Wawasee, Winona, and Yellow Creek lakes.

Month	Blue-green algae counts (cells/ml)
May	29,263
June	49,221
July	133,653
August	228,030
Sept.	368,918

## Figures

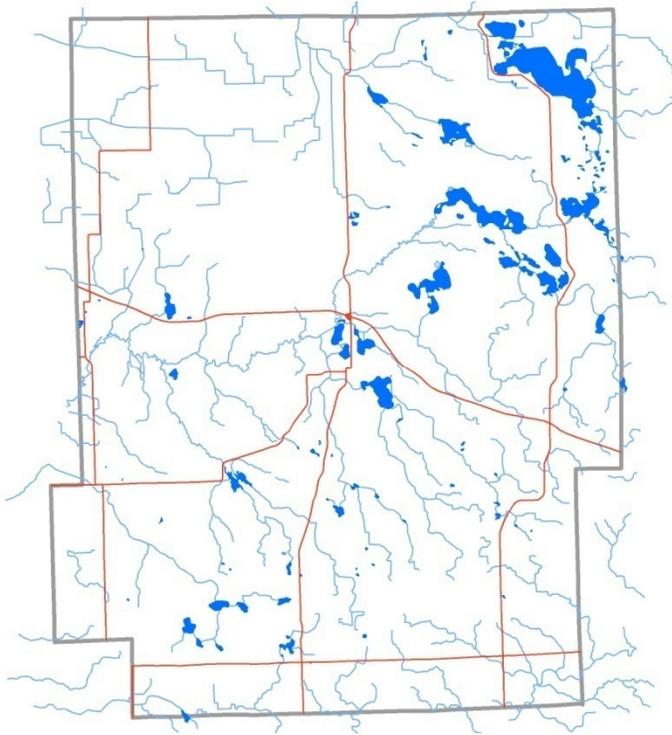


Figure 1. Map of Kosciusko County showing locations of lakes.

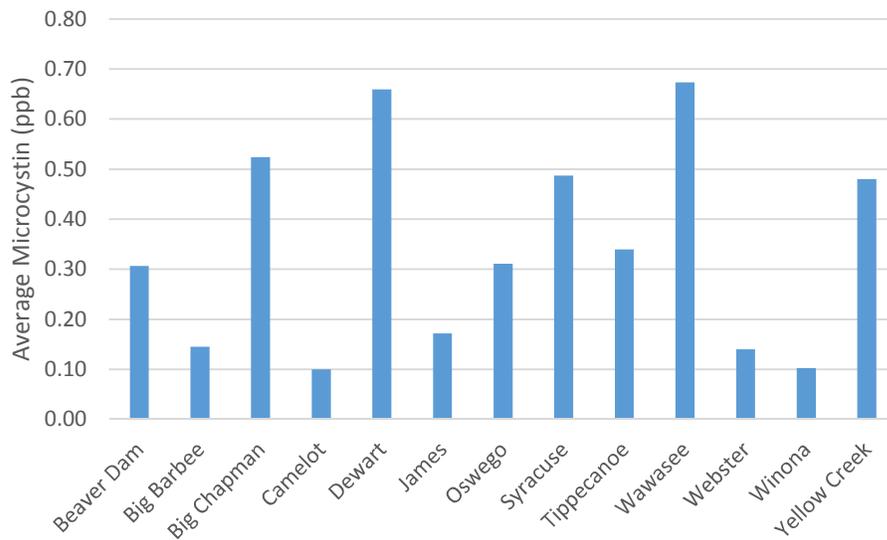


Figure 2. Average microcystin concentrations (in ppb) for 13 all-sports lakes in Kosciusko County over study period of 2010-2013.

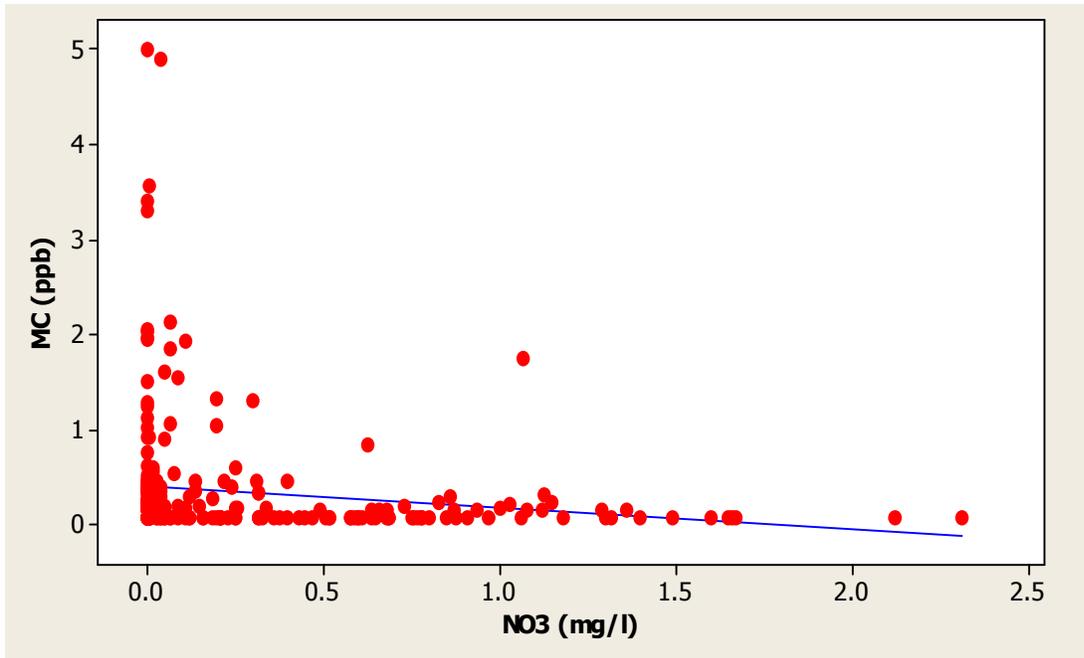


Figure 3. Microcystin (MC) concentrations (in ppb) and nitrate (NO3) concentrations (in mg/l) near surface for 13 all-sports lakes in Kosciusko County over 2010-2013. Blue line shows negative trend of data indicating lower microcystin levels with increasing nitrate levels.

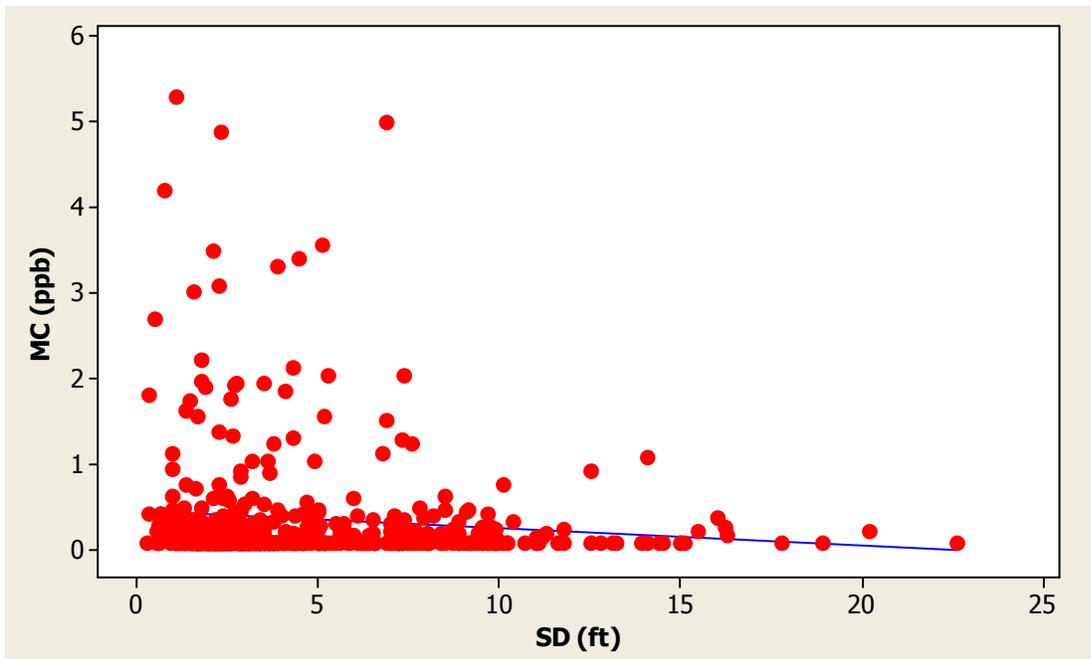


Figure 4. Microcystin (MC) concentrations (in ppb) and Secchi disk (SD) measurements (in ft) for 13 all-sports lakes in Kosciusko County over 2010-2013. Blue line shows negative trend of data indicating lower microcystin levels with increasing water clarity.

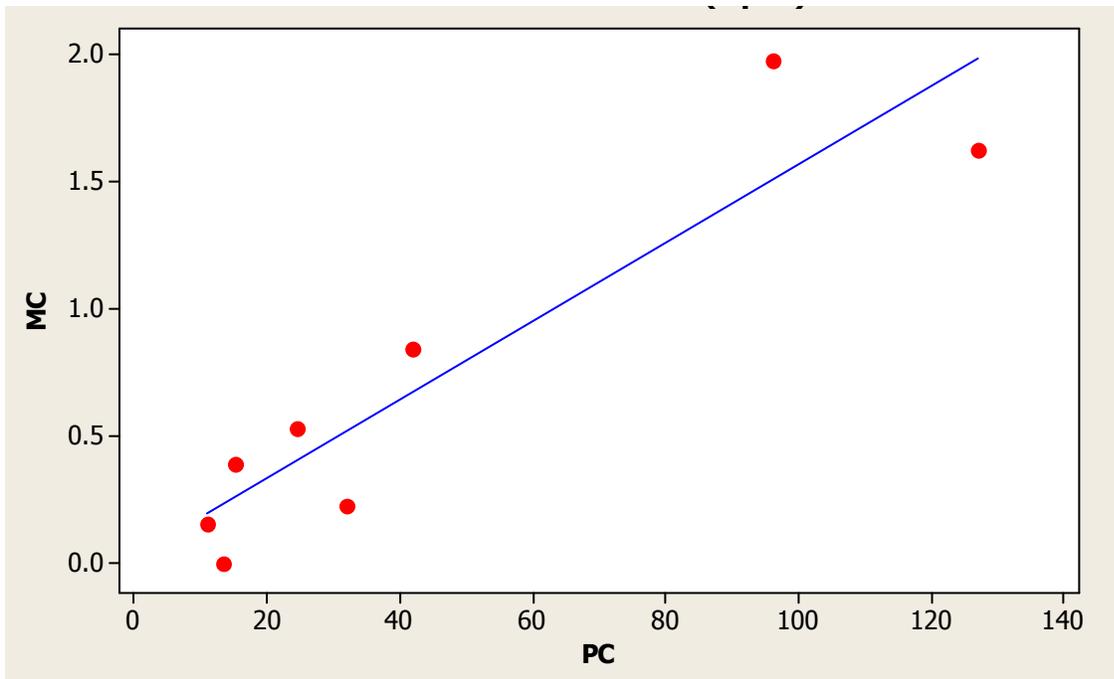


Figure 5. Microcystin (MC) concentrations (in ppb) and phycocyanin (PC) concentrations (in ppb) for Beaver Dam Lake in 2013. Blue line shows positive trend ( $R^2 = 84.8\%$ ) of data indicating prediction of increased microcystin levels with increasing phycocyanin.

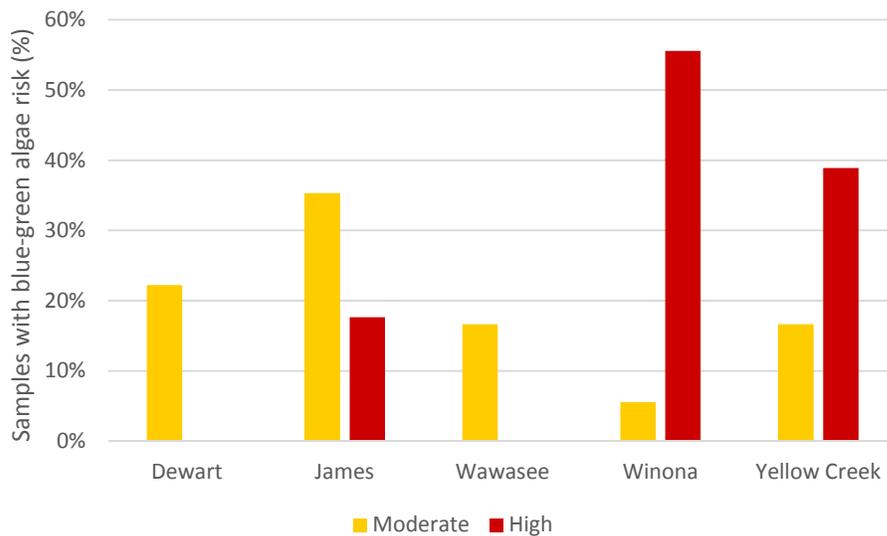


Figure 6. Portion (in %) of samples taken that fell within moderate and high human health risk guidelines for five lakes in Kosciusko County over study period of 2012-2013. Human health risk guidelines set by World Health Organization and the Indiana Department of Environmental Management.

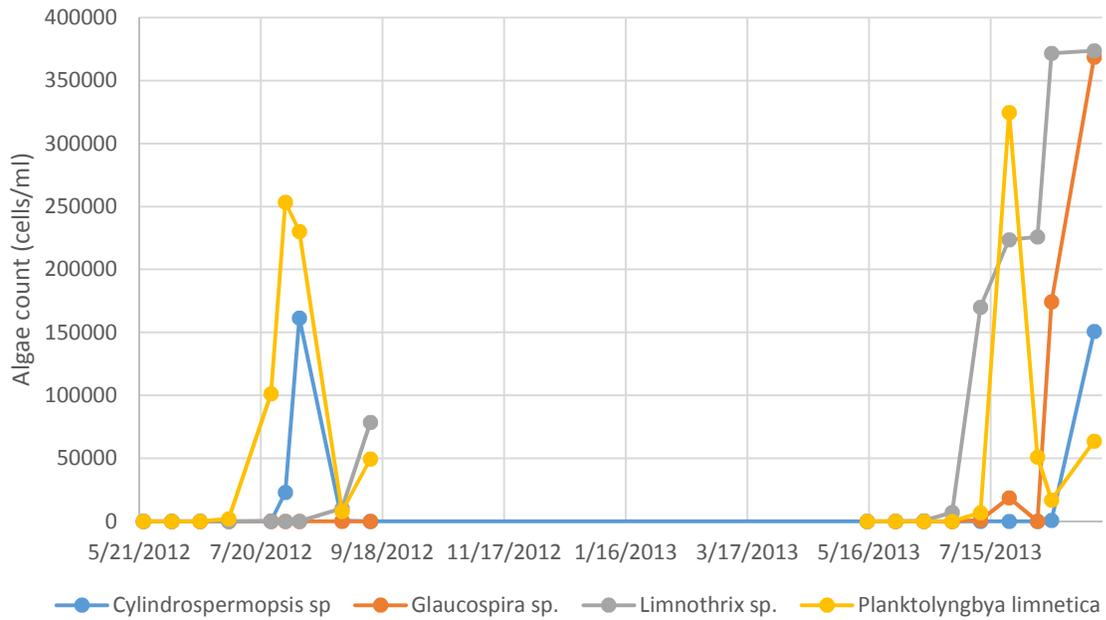


Figure 7. Blue-green algae counts (in cells/ml) for the four most dominant algae types in Winona Lake in Kosciusko County over study period of 2012-2013.

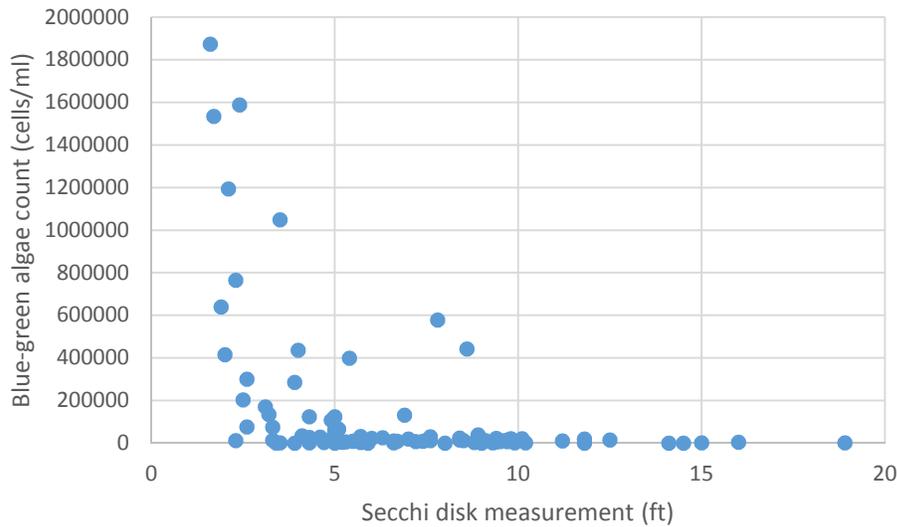


Figure 8. Blue-green algae counts (in cells/ml) and Secchi disk (SD) measurements (in ft) for Dewart, James, Wawasee, Winona, and Yellow Creek lakes in Kosciusko County over study period of 2012-2013.

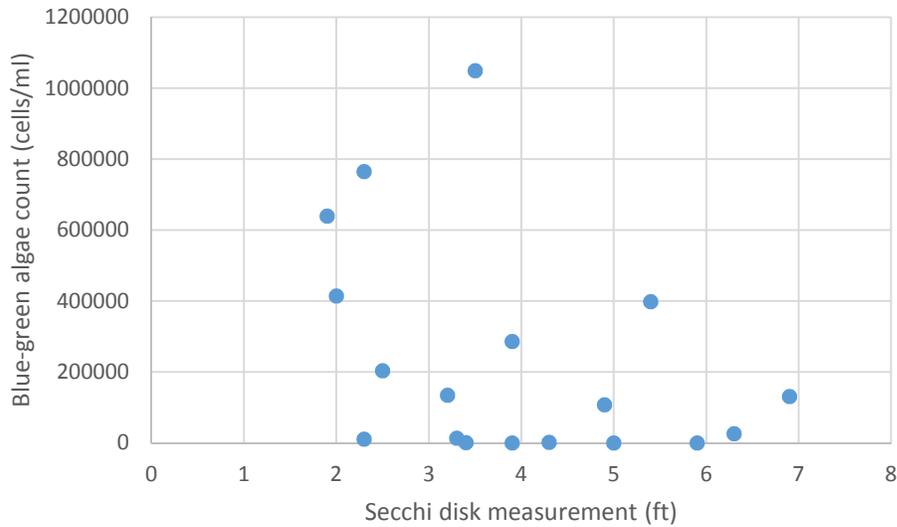


Figure 9. Blue-green algae counts (in cells/ml) and Secchi disk (SD) measurements (in ft) for Winona Lake in Kosciusko County over study period of 2012-2013.

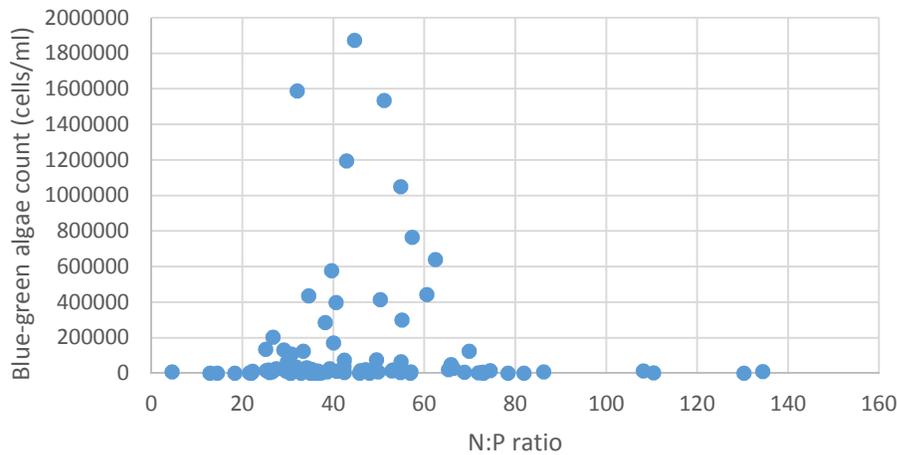


Figure 10. Blue-green algae counts (in cells/ml) and nitrogen to phosphorus ratios (N:P) for Dewart, James, Wawasee, Winona, and Yellow Creek lakes in Kosciusko County over study period of 2012-2013.

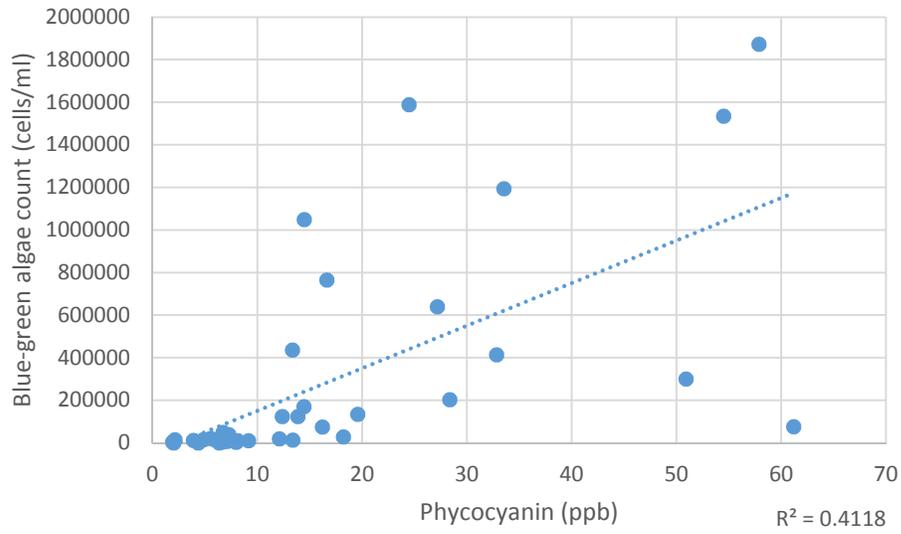


Figure 11. Blue-green algae counts (in cells/ml) and phycocyanin concentrations (in ppb) for Dewart, James, Wawasee, Winona, and Yellow Creek lakes in Kosciusko County over study period of 2012-2013. Linear regression fit to data shown as dotted line with  $R^2$  of 41.2%.