Survey of Zebra Mussel (*Dreissena polymorpha*) Populations in Fourteen Lakes of Kosciusko County, Indiana

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Executive Summary

The freshwater mussel *Dreissena polymorpha*, more commonly known as the zebra mussel, is an aquatic invasive species that spreads quickly and damages lake infrastructure, ecology, and recreation. They spread passively by getting carried downstream in their larval stage or by hitching rides to new lakes on boats as larvae or attached mussels. As efficient and selective filter-feeders, zebra mussels impact water clarity and algae/cyanobacteria communities. This makes zebra mussels a topic of particular interest for Kosciusko County, Indiana lakes where cyanobacteria and their associated toxins are relevant to public health and research. In May-August 2019, we installed PVC multi-tiered samplers onto piers across fourteen major lakes in Kosciusko County, twelve of which have been infested with zebra mussels for multiple years and two of which have yet to be invaded. Monthly and summer counts of zebra mussels showed large variation between lakes and across the area of the largest lakes in the study, and some variation was present even between samplers installed on the same pier. These observations suggest zebra mussel settling and habitat on these lakes are complex, and lakes may vary drastically over a small geographic area in terms of the presence of mussels at this stage in their infestation. The largest zebra mussel counts occurred in July, and Lake Tippecanoe had the highest average zebra mussel counts per sampler, followed by Webster Lake. Previous reports indicate Yellow Creek and Beaver Dam lakes are not yet infested with zebra mussels, and no zebra mussels were observed on samplers from those lakes at any point in the study. Zebra mussels are present in many Kosciusko County lakes as observed here, but not all infestations or impacts by zebra mussels are equal. These data will also be analyzed for correlations between algae/cyanobacteria communities and cyanotoxin levels, which were also recorded the summer of 2019.

Introduction

The spread of invasive species is an ongoing issue in the environmental management of watersheds within the United States. One of the country's most notorious aquatic invaders, *Dreissena polymorpha*, or the zebra mussel, is one of these problematic introductions. These freshwater mussels have quickly spread from the Great Lakes to over 600 lakes and reservoirs within the United States

their larval stage floating downstream to other water bodies or in and on watercraft (Benson et al. 2020; Padilla et al. 1996). As they travel through waterways and are transported by watercrafts, the presence of zebra mussel populations greatly impacts the ecological systems of lakes, rivers, streams and reservoirs. These filter-feeders are incredibly efficient, processing up to one liter of water per day as adults (Benson et al. 2020). While filtering, zebra mussels recognize and reject sediment particles and inedible organisms, such as cyanobacteria (blue-green algae), spitting them back into the water (Benson et al. 2020). This selective feeding can alter the composition of algae and cyanobacteria populations within an infested body of water, though the relationship is complex and dependent on multiple factors (Fernald et al. 2007). Zebra mussels also have a high rate of reproduction, creating denser

(Benson 2020). Their invasion is primarily passive, with

Table 1. First recorded observations of zebra mussels in the fourteen major Kosciusko County lakes (USGS NAS 2016).

Lake	Year
Syracuse	1991
Wawasee	1991
Tippecanoe	1994
Dewart	1995
Big Chapman	1997
Pike	1997
Big Barbee	1998
James	1998
Oswego	1998
Webster	1998
Winona	2000
Center	2016

populations and more rapid infestations, multiplying the negative effects on the ecological quality of surrounding bodies of water . When lake water gets warm enough, zebra mussels release gametes freely into the water (Herman and Wickman 2014). Fertilized eggs will first develop into veligers, free-swimming larva which can populate a lake in the hundreds of thousands (Benson 2020). Veligers find places to settle and grow into juvenile mussels, which, along with adults, can move and migrate using their pseudopod. Juvenile and adult zebra mussels will immobilize and eventually suffocate a native mussel by settling on them directly as preferred habitat (Nalepa et al. 1996). Pipes, power plants and boats can become clogged by thousands of zebra mussels. Lake recreation becomes hazardous as their sharp shells cover ladders, pier posts, seawalls, and other recreational surfaces. This damage to infrastructure and the recreational capacity of lake communities can come at a great cost to local economies dependent on lake resources (Leung et al. 2002; Bingham and Bosch 2016).

On many Indiana lakes, zebra mussels are a regular sight. According to data from the United States Geological Survey, the earliest confirmed sighting of zebra mussels in Kosciusko County, IN, was in 1991 in Syracuse Lake, only three years after the species' first sighting within the U.S. (USGS NAS 2016). Since then, zebra mussels have been a familiar part of lake life for many local residents. As of 2019, twenty-one lakes within the county have confirmed established populations with the Department of Natural Resources (Fischer 2019). Perhaps due to their isolated location in the county and limited all-sport boating hours, Beaver Dam Lake and Yellow Creek Lake are two of the few non-infested lakes in the county. Most local lakes in the Winona-Warsaw, Wawasee-Syracuse, and Tippecanoe area, however, have had zebra mussels present for years (**Table 1**).

Kosciusko County lake residents reportedly experienced a boom in mussel population during the initial establishment of the species in the early nineties. Residents have observed variations in zebra mussel population size and intensity of infestation over the years, but no quantitative reports have been

made of these population changes. The Lilly Center for Lakes & Streams in Winona Lake, IN designed and conducted the following study to quantify current zebra mussel populations of fourteen major lakes relative to one another and around each lake. The interaction between zebra mussels and cyanobacteria/algae populations is of particular interest in Kosciusko County lakes where cyanobacteria and their associated toxins are relevant to public health and research (Bosch et al. 2019). This study will set a baseline for current populations for future studies monitoring changes in zebra mussel populations specifically for their impact on algae communities and cyanotoxin levels.

Study Design

This study took place on fourteen lakes in Kosciusko County, IN during May – August 2019: Beaver Dam Lake, Big Barbee Lake, Big Chapman Lake, Center Lake, Dewart Lake, the Tippecanoe **Table 2.** Sampled lakes and their pier and samplerdistributions. Asterisk denotes one pier wasequipped with three samplers on that lake.

	Surface		~ .
* 1	Area	Number	Samplers
Lake	(acres)	of Piers	Installed
Beaver Dam	155	2	2
Big Barbee	311	3	3
Big Chapman	504	3	3
Center	120	2	2
Dewart	554	3	3
James	278	3	3
Oswego	78	2	2
Pike	228	2	2
Syracuse	411	3	3
Tippecanoe	876	5	7*
Wawasee	3,006	6	8*
Webster	653	3	3
Winona	571	4	6*
Yellow Creek	155	2	2

chain (James Lake, Lake Tippecanoe, Oswego Lake), Pike Lake, Syracuse Lake, Lake Wawasee, Webster Lake, Winona Lake, and Yellow Creek Lake (**Table 2**). All lakes except Center and Pike are open for all-

watersport recreation at all or various times of day. All fourteen lakes are residential and have public boat ramps. Sampling occurred May – August 2019, which is both peak lake recreation season and the reproductive season for zebra mussels due to warm water temperatures (Benson et al. 2020). The method of sampling in this study provided an artificial habitat during the spawning season; if zebra mussel veligers were present in the water, on the PVC sampler served as definite habitat for them to colonize. Counting settled mussels gave an indication of the reproductive activity of adult populations not directly sampled in this study.

Methods

Sampler design: Zebra mussels were quantified using multi-tiered PVC samplers, which act as a substrate for new veligers and young mussels to settle and grow on (**Fig. 2**; WI DNR 2010). Tiered samplers were constructed out of uniformly sanded black PVC sheeting, a preferred material and texture for zebra mussel settling (Kilgour and Mackie 1993; Marsden 1991). The four plates of the samplers were 12×12 in, 10×10 in, 8×8 in, and 6×6 in, stacked on a 6 in long eye bolt with a 1 in PVC pipe between each plate to keep them uniformly separated, per WI Department of Natural Resources adult and juvenile zebra mussel sampler was strung on a cable with a carabiner on the end, which secured the sampler to the bottom of a pier. Each sampler was spray-painted with an identifying number on a small section of the bottom plate.

Sampler distribution across lakes: Samplers were hosted on the piers of lake homeowners, and sampling locations were chosen to be distributed across a lake as evenly as possible. To sample for



Fig. 2. Tiered PVC zebra mussel sampler.

variability in the colonization of zebra mussels on a single pier, three of the sampled lakes (Beaver Dam, Tippecanoe, Wawasee, Winona) hosted three samplers instead of one (**Table 2**). Two samplers on each of these piers were swapped out monthly for counting, and one was left in the lake for the duration of the summer.

Sampler installation: Researchers recorded the install date, water depth, sampler hanging depth, and sampler location on pier upon installation. All samplers were installed between May 14 and May 30, 2019. To fit zebra mussel preferences as closely as possible, samplers were hung in the deepest water possible accessible from the piers (Marsden 1991; WI DNR 2010; maximum hanging depth of 0.9 m, minimum hanging



Fig. 3. PVC samplers populated with adult zebra mussels (leftmost), juvenile zebra mussels (center), and large clusters of adults (rightmost) from this study.

depth of 0.25 m for this study). On piers with multiple samplers, the samplers were spaced evenly along the length of the pier.

Monthly sampler replacements: All samplers, excluding those that remained installed for the duration of the study, were swapped out with clean samplers once each month in the summer (June 17-20, July 22-24), and fully removed in August (Aug. 12-14). When swapped or uninstalled, each sampler was placed in its own garbage bag to avoid contamination by any other sampler during transportation back to the Lilly Center. A clean sampler was then installed exactly where the previous sampler was located on the pier. Because samplers were individually numbered, researchers were able to consistently use the same two samplers to replace each other every month at the pier. In this way, researchers could determine the sampler's lake, pier location and replacement date by only the number and avoid contaminating samplers between lakes. Samplers used at Beaver Dam and Yellow Creek were also handled with particular care to avoid potential cross-contamination from infested lakes. Samplers at Winona, Wawasee, Tippecanoe and Beaver Dam that remained for the duration of the study were monitored but not fully uninstalled and counted during the summer until August.

Counting and data analysis: Samplers were brought back to the Lilly Center for counting and cleaning. Researchers recorded the sampler number, counter's initials, date, and comments on a physical data sheet. The sampler was disassembled, and individual adult and juvenile zebra mussels were visually identified and counted on the top and bottom surface of each tier plate and the total for each plate surface was recorded on the data sheet (**Fig. 3**). Only live mussels were counted, and clusters of mussels were broken apart and individuals counted. Populations up to about 250 individuals were counted by hand, and

above this threshold, an area calculation was used. An area was selected haphazardly for what appeared to be an area of average population density for the plate. This total was multiplied by the total area of the plate surface minus 1-inch square (to account for the area in the middle of the plate that was covered by the PVC divider), and the results recorded as the total sum for the plate's surface. This process was repeated for all samplers.

Sampler cleaning: All samplers were cleaned after every counting. Zebra mussels and other material were scraped off each plate. All parts of the sampler were then scrubbed with a hard bristle brush and a mild dish detergent. Each sampler was placed in a bin designated by lake and reassembled using the same pieces. After reassembly, samplers were sprayed thoroughly with vinegar and placed on a rack to dry to ensure no viable zebra mussels veligers remained (WI DNR 2010). The samplers were then stored on racks for one month until their next deployment.

Results

Settled zebra mussels were observed on samplers from all lakes except Beaver Dam and Yellow Creek over the course of the summer (**Table 3**). During the study, four samplers were lost and could not be recovered from their piers during collection: one on an individual pier in Winona in July, one from an

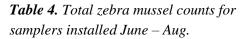
Lake	June	e	Jul	ly	Augu	ist
Lake	Total	Avg.	Total	Avg.	Total	Avg.
Beaver Dam	0	0	0	0	0	0
Big Barbee	3	1	30,693	10,231	12,041	4,014
Big Chapman	23	8	1	1	0	0
Center	0	0	4	2	0	0
Dewart	1	0	300	100	0	0
James	1	0	44,950	14,983	127	42
Oswego	8	4	35,841	17,921	161	81
Pike	0	0	50	25	0	0
Syracuse	1	0	0	0	0	0
Tippecanoe	8	1	228,699	38,117	6,597	1,319
Wawasee	1	0	51,814	8,636	106	21
Webster	53	20	70,230	35,082	2,148	1,074
Winona	8	2	74,006	18,502	1	0
Yellow Creek	0	0	0	0	0	0

Table 3. Monthly total adult and juvenile zebra mussel counts and average
counts per sampler. Counts do not include samplers installed the full summer.

individual pier in Webster in July, one from an individual pier in Tippecanoe in August, and the fullsummer install sampler in Tippecanoe.

Temporal variation: At all sites where zebra mussels were found, most population densities increased during the reproduction cycle of the summer, with the highest totals found in the month of July. Most populations consisted primarily of juvenile mussels until later during the study, in August, when large populations of adult mussels were found on samplers. The highest number of mussels on a sampler in June was 53, which increased to 228,699 in July and 12,041 in August. This trend was similar for most lakes, with the exception of Syracuse, which had 1 counted zebra mussel for the duration of the study, Beaver Dam and Yellow Creek, which both had no counted mussels, and Big Chapman, which peaked in

June at 23 mussels. For samplers left installed through the whole summer, Beaver Dam's sampler was retrieved with zero adult or juvenile zebra mussels, Tippecanoe's sampler went missing during the study and could not be retrieved, and Wawasee and Winona samplers had established colonies of zebra mussels present (**Table 4**). For both Wawasee and Winona, the samplers' totals were closest to the totals found in the month of July for that pier location and was much higher than the counts of samplers cleaned and reinstalled for August.



Lake	Total
Beaver Dam	0
Tippecanoe	*
Wawasee	23,862
Winona	4,463

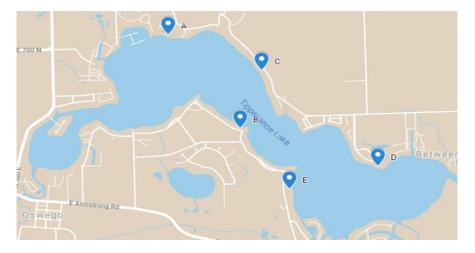


Fig. 4. Tippecanoe Lake pier locations. Pier A held two monthly samplers (A1 and A2 on Table 5) and one summer-long sampler.

Table 5. Total counts compared betweenpier locations on Tippecanoe Lake.Sampler on pier D detached in August, sono count could be performed for thatlocation that month.

Pier	June	July	August
A1	1	15,520	395
A2	0	8,684	4,140
В	0	75,754	114
С	4	72,980	1,915
D	3	28,034	*
Е	0	27,727	33

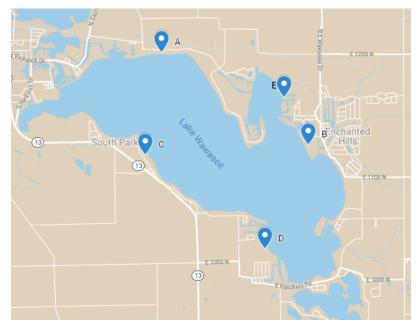


Fig. 5. Lake Wawasee pier locations. Pier A held two monthly samplers (A1 and A2 on Table 6) and one summer-long sampler.



Table 6. Total counts compared between pier locations on Lake Wawasee. Sampler on pier E detached in August, so no count could be performed for that location that month.

Pier	June	July	August
A1	0	9,458	0
A2	0	8,026	23
В	0	9,950	0
С	0	10,648	0
D	0	13,732	83
Е	1	0	*

Table 7. Total counts compared between pier locations on Winona Lake. Sampler on pier C detached in July, so no count could be performed for that location that month.

Pier	June	July	August
A1	0	24,980	0
A2	0	25,571	0
В	8	13,675	0
С	0	*	1
D	0	9,780	0

Fig. 6. Winona Lake pier locations. Pier A held two monthly samplers (A1 and A2 on Table 7) and one summer-long sampler.

Spatial variation: Multi-sampler piers on Winona and Wawasee showed some difference

between samplers in the same location each month (**Table 6, 7**). Tippecanoe's samplers on the same pier varied much more (**Table 5**). Sampler 1 on Tippecanoe pier A had almost twice as many adult and/or juvenile mussels than A2 in July, and A2 had over ten times more than A1 the following month. Counts also varied across lake area on each of these lakes (**Fig. 4, 5, 6**), most notably in July.

Comparison between lakes: Lake Tippecanoe had the highest counts total and on average, with an average of 38,117 zebra mussels found across its six samplers in July. The highest individual sampler total was also found on Tippecanoe that month, with 75,754 zebra mussels on one sampler. Yellow Creek and Beaver Dam were the only lakes to have no observed zebra mussels during the entirety of the study. A few of these lakes are directly connected to one another as lake chains: James, Tippecanoe, and Oswego form the Tippecanoe chain, and Wawasee and Syracuse are connected with a large wetland area called Mud Lake. The Tippecanoe chain counts were comparable to one another on average each month, but Wawasee and Syracuse differed greatly in their per sampler averages.

A few notable observations were made during installation and removal processes. Upon the first removal of samplers in late June, adult zebra mussels were observed on nearby rocks in Wawasee and Webster, while the sampler itself contained no adult or juvenile mussels. Some mussel veligers were observed on the samplers of multiple lakes in the July counting, though these were not identified to species (zebra mussel or otherwise) or quantified.

Discussion

Understanding the difference between zebra mussel expansions in different lakes is critical to developing a profile of the species' invasion in Kosciusko County and identifying potential correlations between zebra mussels, algae populations, and algal toxin levels in the county's lakes. Zebra mussels have had established populations in Kosciusko County since the early 1990s. This study, based on colonization of artificial substrate samplers on volunteer piers by juvenile and adult zebra mussels, aimed to quantify zebra mussel infestations to compare within and between lakes.

Zebra mussel counts were highest in July and moderately high in August, with veligers observed in June. This spawning time is related to water temperature, which is relatively consistent across these lakes in the summertime. Sources of heat pollution could potentially influence spawning habits in smaller areas of a lake (Benson 2020). Whether or not these sources exist in Kosciusko County lakes and whether or not they have an impact of zebra mussel populations could warrant future study.

The Tippecanoe chain, Wawasee, Big Barbee, Webster, and Winona appear to have large spawning and colonization events in the summertime, and likely have an established adult population that contributes to those reproductive events, though that population was not quantified here. Other lakes in the county – Big Chapman, Center, and Syracuse – have smaller spawning populations, or none at all, as

seems to be the case for Beaver Dam and Yellow Creek. It may be that the minimally infested lakes are in a "bust" portion of a "boom and bust" population cycle in which huge population growth causes a lack of food and habitat, decimating the population again until conditions are viable again. It could also be that other factors, such as bottom substrate or food availability, make certain lakes or lake areas poor habitat for colonization despite veligers likely being present from upstream sources (Benson 2020), and this can change in a lake or lake area over multiple years (Strayer and Malcom 2006). This may be the case for Syracuse, which is downstream of Wawasee. Population cycles or other factors in variation in zebra populations between lakes are reasons for continued monitoring of zebra mussel spawning and colonization in these lakes.

This variation in colonization also applies to a single lake and a single pier. Lakes with multisampler piers – Tippecanoe, Wawasee, and Winona – varied across their area. (Beaver Dam also had a multi-sampler pier, but had no zebra mussels.) This pattern could be related to the location of spawning adults, the availability of better habitat close by, or a number of other factors. These lakes likely contain many more appropriate settling habitats for zebra mussels as they can settle in water much deeper than piers, but zebra mussel habitat is greatly dependent on turbidity and algae availability (Benson 2020). Water clarity and algae populations are also known to vary greatly between these fourteen lakes (Bosch et al. 2019), adding to the complexity of local zebra mussel infestations. Piers themselves seem to be able to vary in colonization potential. More research is needed to determine what factors play into differences in colonization between samplers on the same pier or small areas along a shoreline. These observations suggest future monitoring that involves multiple piers across a lake for area and an investigation into which areas are most likely to be colonized. Future work could also involve the quantification of the settled adult population and survival rates of young zebra mussels.

As a baseline study, these data collected from this 2019 Lilly Center for Lakes & Streams zebra mussel survey will be beneficial in the next steps for investigation into local zebra mussel populations and the species' impact on multiple aspects of lake health. In addition to the research conducted on zebra mussels, key health parameters, such as algae and cyanobacteria identification and counts and cyanotoxin concentrations, were collected weekly throughout the summer. These data may offer further insights into the ecology and health of Kosciusko County lakes, and analysis of these data is ongoing. Continued summer monitoring of those parameters and others is also ongoing. In future studies, special attention will be paid to potential invasion of Beaver Dam and Yellow Creek by zebra mussels. If invasion is not properly prevented, these data and years of previous water quality measurements will act as a baseline for understanding the impact of zebra mussels on water quality, algae/cyanobacteria populations, and cyanotoxin production.

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