The Impacts of Herbicides on the Diatom Communities Found Within the Burt Lake Channel

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Abstract

A study was done of a channel off of Burt Lake to study the impacts of the herbicides being dumped into the area for human recreational purposes. Diatoms in particular were studied due to their ability to indicate environmental conditions of a system. Samples were done utilizing a plant squeezing technique and cellular counts. Shifts of multiple genera were discovered throughout the channel as the lake was approached. These shifts suggest a change in nutrient and pH levels from the first site to the last.

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Signed, Michelle Busch

Introduction

In many ecosystems, arguably the most important organisms for overall ecosystem health are the primary producers (Stiling, 2012). The factors which typically impact the primary productivity of aquatic ecosystems are nutrient variability and light penetration (Townsend et. al, 2008). Light is quickly absorbed by water, by a depth of one meter, half of the intensity has already been absorbed (Stiling, 2002). When light is lacking, photosynthesis is not able to take place, greatly limiting the primary productivity possible in a system. The two nutrients which tend to limit primary productivity in aquatic systems tend to be phosphorus and nitrogen (Townsend et. al, 2008; Stiling, 2002). Light intensity and nutrient composition, along with other factors such as temperature and flow velocity, play a large role in determining what primary producers can live in an area. The primary producers in most aquatic systems are typically either aquatic plants and algae, including diatoms (Stiling, 2012).

Diatoms are crucial to better understanding an aquatic ecosystem. Not only are they vital to food webs (Mayer & Likens, 1987), able to oxygenate surface water (J. P. Descy, communication), supply links in biogeochemical cycles (Newbold et al., 1981), and provide a system with much of its "biodiversity and genetic resources" (Patrick, 1961), assessing diatoms is one of the best tools in order to assess the environmental issues in a specific area. In fact, "people began to compile and speculate upon the relationships between the occurrence of certain diatoms and other things which were useful to know almost as soon as optical microscopes were developed" (Stoermer & Smol, 1999). Diatoms are excellent indicators of the chemical and nutrient conditions of a system, and easy to study (Stoermer & Smol, 1999). After any sort of physical, chemical, and biological changes take place in an environment, diatom communities respond quickly and sensitively to such changes, demonstrating how the system as a whole is

impacted (Squires et al., 1979). "[Diatoms] reproduce and respond rapidly to environmental change and provide early warning indicators of both pollution increases and habitat restoration success" (Stoermer & Smol, 1999).

While diatoms are able to demonstrate the impacts of pollution, different diatoms have distinct niches. Diatoms found in lakes tends to be benthic and epiphytic. Benthic algae is attached to hard substrate or fine sediment and is known as epilithic (Round, 1965). Diatoms are also able to grow on plants and are known as epiphytic (Bold & Wynne, 1985). Diatoms found in rivers, however, are mainly found along edges and floating debris or on large beds of floating plants. Areas where the river water is shallow and less turbid provide great environment for diatoms. Similar to species found in lakes, diatoms living in rivers are usually epipelic (attached to mud or sand), epiphytic (growing on plants), or benthic (bottom) species (Bold & Wynne, 1985). River diatoms depend on the river's watershed for nutrients. No matter where diatoms are found, however, "the structure of diatom communities...seems to be largely determined by available species pool, invasion rate, density-independent factors, density-dependent factors, predator pressure, parasitism, and competition" (Werner, 1977).

"Diatoms are affected by many different parameters in the environment and it is probably a combination of factors that limit their distribution" (Lowe, 1974). These factors include flow velocity, light, temperature, turbidity, pH, and many distinct nutrient levels. Flow velocity is one of the most important factors in diatom growth and community (Ruttner, 1940) (Kolbe, 1932). This factor plays an important role in determining the algal community by washing away detritus and nutrients, determining the stability of the substrate, and impacting algal immigration to an area (Stevenson, 1983). Distinct species of diatoms prefer unique light intensities in order to better undergo the processes of photosynthesis. Temperature can either directly or indirectly

impact the diatom population of an area. In one study, it was found that "general diatom diversity seemed to increase as one approached the optimum range of temperature within the range of tolerance for most of the species of a community" (Stevenson, 1983), leading to the conclusion that species dominance can be highly related to temperatures. Turbidity also impacts a diatom community. As the turbidity increases, the sediments get homogenized which can impact light penetration. The hydrogen ion concentration impacts what form of carbon is available to the diatom community, impacting which species can thrive. Many ions, nutrients, trace elements, and organic compounds can also influence a diatom community. These factors typically impact metabolism or the photosynthesis of the communities, affecting diatom growth. Due to the specific conditions which impact diatom growth, these organisms can provide a plethora of insights into the condition of an environment.

In Cheboygan County, Michigan lies Burt Lake. In the late seventies, a channel was dug alongside Burt Lake in order to provide more housing with lake front property for residents in the area. The channel was filled in with lake water. As the channel became more populated, sea walls were installed and some private docks were placed in the channel. In order to get their boats into the channel and thus into the lake more easily, the residents began to spray herbicides. These sprays occur every month. In order to better assess the health of the channel's ecosystem and the impacts of the herbicide sprays, a study was done to observe and identify the different diatom communities found within the system. Along with a study of the diatom community, mussel and amphibian surveys were completed. Land studies were done of both the natural and human impacted area surrounding the channel in order to better understand every aspect of the Burt Lake channel.

Methods

To test the growth and community composition of diatoms, a handful of rooted plants were taken. These plants were then rinsed and squeezed three times over an enamel pan to collect the algae and diatoms growing on the plant. Plants were utilized due to their abundance throughout the channel, allowing for consistency in data. The diatoms collected were then flushed into a wide neck bottle. These bottles were placed in a cooler to keep the samples cold and in the dark. The same process was repeated at each of the ten sites along the Burt Lake channel. When present, hard substrate such as rocks were taken to collect algae. Using a toothbrush, the algae was removed from a measured area of the substrate. Rocks were sampled when possible to get a better understanding of the primary productivity of the sites where larger rocks could be found. Water samples were also taken at each of the sites along with pH levels to get a full understanding of the channel system.

Plant samples were also taken for diatom counts within Maple Bay of Burt Lake and Carp Creek, a creek found within the same water shed as the Burt Lake channel, in order to provide samples from non-impacted environments. The same process utilized in the channel was put into practice at these two sites.

Using an eyedropper, one or two drops of each sample was placed onto a cover slip after the samples were mixed to homogenize the solution. The number of droplets of sample was determined by the density of the diatoms. This droplet was then diluted with six drops of water in order to spread out the sample. Using a low temperature on a hot plate, the water on the coverslips was evaporated. Once all of the water was gone from the slides, the temperature of the hot plate was increased to about 225 degrees Fahrenheit. The coverslips were then allowed to burn in order to get rid of any extra organic material. This temperature was maintained for

about an hour. Once the coverslips had cooled, naphrax was added to a slide, the coverslips were attached and heated at about 175 degrees Fahrenheit for as long as it took for the naphrax to cease bubbling (Stevenson et. al, 1996; Pillsbury, Personal Communication).

These slides were then observed and cells were counted and identified. The counting process took place by placing each slide under a scope and recording the first three hundred cells observed. These identifications were done with the help of Robert Pillsbury and the taxonomic book *The Diatoms of the United States: Volumes I and II* (Patrick & Reimer, 1966; Patrick & Reimer, 1975). Each diatom family observed was researched in order to conclude the environmental conditions necessary for their growth. Species specific counts from the first two sites was done. The most common species found there allowed for extrapolation to a family level in order to determine niche needs for the diatom families.

Once the counts were completed, the percentage of each genus was found in the samples by taking the number of individuals of the genus, dividing them by the total counted, and multiplying by 300. This was done in order to normalize the data so that each site had the same amount of cells counted. A Friedman's ANOVA test was done in order to compare the data across all eight sites. A Simpson's Diversity Index was completed to get a better understanding of the diversity at each site.

Results

The Friedman ANOVA test had an n of 8 and 14 degrees freedom. The Friedman ANOVA, ANOVA Chi Square test came up with a number of 69.4. The p-value was less than 0.00001. Simpson's Diversity Index (figure 1) increased at the first three sites from 0.4301 to 0.816; the third site having the highest diversity index. The next highest in the Burt Lake

Channel was site nine with an index of 0.8121. The site at Maple Bay had an index of 0.8513 and the site at Carp Creek had an index of 0.7792 (figure 2).

$$\hat{D} = \frac{\sum_{i=1}^{S} n_i (n_i - 1)}{N(N - 1)}$$
 Fig. 1

Site	Burt Lake Channel 1	Burt Lake Channel 2	Burt Lake Channel 3	Burt Lake Channel 5
Simpson's Value	0.4301	0.6917	0.8156	0.7450
Site	Burt Lake Channel 7	Burt Lake Channel 9	Maple Bay Site 1	Carp Creek Site 2
Simpson's Value	0.7297	0.8121	0.8513	0.7792

Fig. 2

The diatom genus *Achnanthes* was most plentiful at the first site of the channel; this was also the point in the channel where there was the most of any single genus of diatoms. These numbers decreased throughout the channel, except between the third and forth site where there was a slight increase. The control sties of Maple Bay and Carp Creek had around the same cell count as the fifth site, between 50 and 60.

Denticula was not present in the channel until the ninth and final site. This genus was also present in Maple Bay and Carp Creek, however, it was not as prevalent at these sites than the channel. The count in the channel was 41. Maple Bay had a total cell count of 8 while the Carp Creek sample contained 5 Denticula cells.

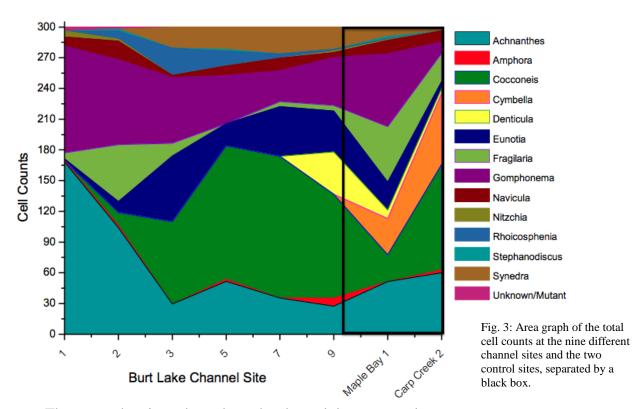
The third site of the Burt Lake Channel had the highest amount of *Eunotia* diatoms. This increase was extremely steep, beginning with only two *Eunotias* at site one and ending up with 64 at site three. *Eunotia* counts were also high in the last two sites (48 and 40, respectively). Numbers of this genus dropped in the control sites, with only 27 found in Maple Bay and 7 found in Carp Creek.

The amount of *Fragiliaria* in the channel increased from six to 55 between the first two sites and dropped back off to 13 at the third. The numbers decreased to zero at the fifth site of

the channel and remained constant until the last site at a count of five. Maple Bay had a high count of 54, and Carp Creek had a count of 28.

The first site of the Burt Lake Channel contained 105 *Gomphonema* cells. This number decreased throughout the channel, reaching a count of 30 at site seven. The number then increased to 47 at site nine. Maple Bay had a 71 *Gomphonemas*, a count closest to the third site with a number of 64. Carp Creek contained the least amount of cells with a count of 11.

Cocconeis numbers increased from no cells at the first site to a count of 138 at the seventh site. This number dropped back down to 102 at the last site; a number similar to the count found at Carp Creek (103). Maple Bay contained a total of 26 cells, a number most similar to that of the second site of the channel which had a count of 12 (figure 3).



The water chemistry throughout the channel demonstrated a

general decrease in nitrogen concentration the closer the sites got to Burt Lake, with some spikes

at different sites. These levels began at 0.550 mg/L of N and decreased to 0.453 mg/L of N at site nine. The highest spike in the levels occurred at site three when nitrogen levels increased to 0.563 mg/L of N. The level of nitrogen in Maple Bay was 0.248 mg/L of N and Carp Creek had a level of 0.174 mg/L of N. Silica levels increased and peaked at the fourth site, and remained around 12 mg/L SiO₂ until the eighth site when the levels decreased to around 9.4 mg/L SiO₂. Silica levels at Maple Bay and Carp Creek were 5.7 and 5.6 mg/L SiO₂, respectively. Chloride levels decreased throughout the channel, beginning at a level of 71.2 mg/L Cl and ending at the ninth site at a level of 24.7 mg/L Cl. The pH level remained pretty consistent throughout the channel, at a value of 8.47 (figure 4).

ID	NO3-N	NH4-N	SRP	TN	SiO2	Cl
	μg N / L	μg N / L	μg P / L	mg N / L	mg SiO2/L	mg Cl / L
1	< 1.0	< 1.0	5.7	0.550	11.3	71.2
2	< 1.0	< 1.0	4.6	0.521	11.8	68.5
3	< 1.0	< 1.0	4.4	0.563	12.2	66.4
4	< 1.0	2.7	4.4	0.440	12.6	64.6
5	< 1.0	< 1.0	4.2	0.507	11.9	58.2
6	< 1.0	2.7	4.2	0.444	12.2	57.1
7	< 1.0	< 1.0	4.8	0.399	12.1	52.0
8	19.6	17.6	3.4	0.365	9.6	34.9
8b	23.3	23.7	3.3	0.554	9.2	32.5
9	29.8	43.4	2.9	0.453	9.2	24.7
Maple Bay	8.0	9.1	2.0	0.248	5.7	10.3
Carp Creek	43.15	4.5	2.1	0.174	5.6	7.775

Fig. 4

Discussion

The significance value for the Friedman value (0.00001) suggests that the cells counted at each of the eight sites are significantly different. Based on this ANOVA Chi Square test, each site's distribution of diatoms is not equal to any other site's diatom population. The Simpson's Diversity Index demonstrated that Maple Bay has the most diverse group of diatoms, followed by site three and site nine. Maple Bay was one of the control sites (not impacted heavily upon by humans), thus, more diatoms were expected there. The third site had the highest levels of silica (12.6 mg SiO₂/L), possibly contributing to the high diversity found there. However, Maple Bay

and the ninth site of the channel had the next highest diversity indices, yet both had lower levels of silica (5.7 mg SiO₂/L and 9.2 mg SiO₂/L, respectively), suggesting that more than just high silica levels are needed in order to have a diverse community of diatoms.

The *Achnanthes* population decreased rapidly from the first to third site, increasing at the fifth and then decreasing until the end of the channel. Maple Bay and Carp Creek had numbers similar to those found at the fifth site (around 55). These species are typically found in a pH range of 4.3 to 9.2, with an optimum range of 7.5 - 7.8. This genus can tolerate a small amounts of salt, and are able to live where inorganic nutrient levels are high (Lowe, 1974). Similarly to the distribution of *Achnanthes*, the *Gomphonema* population decreased throughout the channel. This genera has an optimal pH level of 7.8 to 8.2, can tolerate low levels of salt, and prefer to be in zones where the oxidation of organic material is currently taking place (Lowe, 1974). Based on the numbers of *Achnanthes* and *Gomphonema* present, one can assume that the pH level was more neutral at the first site of the channel, and became more or less acidic as the sites continued. Also, because *Achnanthes* are able to live where inorganic nutrient levels are high, any pollution from the herbicides and fertilizers used in the channel would theoretically not have much of an impact on this particular population.

Cell counts of *Eunotia* and *Cocconeis* were also similar; increasing and then decreasing within the channel. The amount of *Cocconeis* increased at a much more dramatic rate, however, and did not decrease in numbers until the last site was examined. Both genera had lower counts in the control sites; *Cocconeis* had higher numbers in Carp Creek and *Eunotia* had higher numbers in Maple Bay. *Eunotia* have been found to be associated with acidic water (Pierre, 1996), suggesting that where the numbers increase, sites three and seven especially, the acidity of the water should also increase. *Cocconeis* has an optimal pH above 7, which matches the

data; at *Eunotia* numbers increase and acidity increases, the number of *Cocconeis* decrease. Similar to *Achnanthes* and *Gomphonema*, *Eunotia* and *Cocconeis* are both able to tolerate low amounts of salt. *Eunotia* are able to survive in areas of inorganic nutrient levels are high, and *Cocconeis* can tolerate areas of high nitrogen pollution (Lowe, 1974).

Fragilaria have optimal growth in areas of a pH around 7.7 and 7.8, need high nutrient concentration, can tolerate small amounts of salt, and are able to grow where the concentration of inorganics is high (Lowe, 1974). These cell counts increased from the first to second site and decreased at the third. The count dropped off to zero at the fifth site and remained consistent for the rest of the channel. In comparison to the channel, both Maple Bay and Carp Creek had high counts of this genera. However, once the ninth site was reached, Fragilaria were seen that are planktonic species which were also found in Maple Bay, suggesting that the channel becomes more lake-like as it enters Burt Lake. This is probably due to the high amounts of mixing that occur at the mouth of the channel between the channel water and the lake water.

Once site nine was reached, a new genera was observed in the counts: *Denticula*.

Denticula were also found in Maple Bay and Carp Creek, though at much lower counts. This genera is commonly found in carbonate rich waters with moderate conductivity. It is typical to find this genera in oligotrophic lakes (Spaulding et al, 2009).

In general, the diatom genera observed in the Burt Lake Channel are able to survive with low levels of salt, yet the chloride levels found in the channel were higher than those observed at Maple Bay or Carp Creek, and these diatoms were still able to thrive. *Denticula*, however, has been found to increase in density as salinity increases (Herbst & Blinn, 1998), yet was only found in areas where the salinity levels were lower, suggesting another factor which would impact their dispersal. The drop of *Cocconeis* cells found in the control sites suggest that there is

some pollution in the Burt Lake Channel which does not exist in either Maple Bay or Carp Creek.

Overall, the diatoms found in the channel offer a multitude of information on the Burt Lake Channel and the shifts that the sites go through. The first site was found to have higher levels of nutrients based on the water chemistry. This is also supported by the number of *Nitzchia* that were found in the area. These diatoms thrive in high nutrient conditions (Patrick & Reimer, 1966). Multiple *Achnanthes* were also found at the first site of the channel, while these diatoms can live in many different environments, they also prefer habitats with higher concentrations of nutrients, supporting the *Nitzchia* population found there. As the second site is approached, *Achnanthes* numbers drop along with the number of *Nitchia*; illustrating a decrease in nutrient availability. *Fragilaria* numbers increased slightly, the species found were those that are epiphytic.

Once the third site was reached, *Cocconeis* numbers increased dramatically, implying a large amount of filamentous algae present in the area. *Eunotia* numbers also increased, demonstrating an increase in acidity of the channel. *Rhicosphenia* numbers also increased in this section of the channel. These diatoms prefer water with a greater flow velocity, suggesting that there is more mixing at this site than the others. At the fifth site, *Achnanthes* numbers increased from the third site, displaying a spike in nutrient levels. *Cocconeis* numbers were the largest at this site than any other, suggesting masses of filamentous algae present here.

Achnanthes numbers decreased from the fifth to seventh site, suggesting a decrease in available nutrients. The seventh site also demonstrated an increase in *Eunotia*, illustrating a decrease in pH levels towards the end of the channel, a trend which continued until the ninth site. The ninth site had a sudden addition of *Denticula* species, which grow well in warmer waters

with high levels of conductivity. This site saw another increase in *Fragiliaria* species, however, this sample had a large increase in planktonic species. *Eunotia* numbers also decreased at this site, illustrating a drop in pH levels. *Cocconeis* levels also decreased, demonstrating a lower number of filamentous algae in the last site of the channel, a trait common to most lentic environments.

In general, the pH levels were low in the beginning, and then increased until the end of the channel based on the counts of *Eunotia*. Nutrient levels and the amount of filamentous algae also decreased as the lake was approached, and *Fragiliaria* species shifted to planktonic as the lake was reached. This datum suggests that there may be more lake water mixing than what was previously hypothesized. This will have an impact on the ecosystem as a whole. The decrease in nutrients will impact the fauna and flora of the system; altering what plants will be able to grow in different areas. The large amounts of *Eunotia* found suggest the acidity of the water is high, preventing commonly found fish from inhabiting the area. Large amounts of filamentous algae suggested by the *Cocconeis* counts could also be choking out the natural flora that could be growing. In conclusion, diatom community structures shift from the first site of the canal to the lake due to their changing environment. These populations found in the Burt Lake channel suggest that while there is some pollution in the canal, the effects of the inorganic and organic matter found there is not that detrimental to the diatom communities in Burt Lake.

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