#### **Executive Summary**

Professor Rex Lowe's Report, October 2015

#### **Investigation of Torch Lake Benthic Algal Outbreaks**

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In the summer of 2015, Three Lakes Association initiated two studies to investigate the recent appearance of golden brown benthic algae in Torch Lake, which were partially funded by Torch Lake Protection Alliance. This is a summary of the findings from one of those studies, as conducted by a team from the University of Michigan Biological Station Experiment Research; Dr. Rex L. Lowe, Professor Emeritus of Biology, Bowling Green State University, Dr. Pat Kociolek, Professor of Biology, University of Colorado, and Kristel Sanchez, University of Michigan Graduate Student.

The research team conducted experiments to determine the nutrients associated with the growth of benthic (bottom) algae. This study employed a method called Nutrient Diffusing Substrate to answer the question "How does benthic algae respond to different nutrients, phosphorus and nitrogen?"

Known amounts of nutrients (nitrate, phosphate, nitrate plus phosphate) were added to agar (a gelatin substance) preparations along with a control agar that contained no nutrients. The agar was placed in plastic cylinder containers which were deployed for 20 days on the lake bottom in 4 feet water depth at two sites on the eastern shore of Torch Lake (Pettys and YMCA Hayo-Went-Ha.)

The concentration of chlorophyll and cell densities were measured on each agar preparation, as indicators of algal growth. The nitrate-enriched agar (highlighted in yellow) had a significantly higher level of chlorophyll when compared to the others (Table AA.) Cell density in the agar samples followed a similar trend as the chlorophyll values; both showed higher algal growth in the nitrate-enriched agar and the nitrate plus phosphate enriched agar (Table AB). Both parameters show that the addition of nitrogen near the lake bottom sands in Torch Lake stimulated more benthic algal growth.

Agar Enrichment	Nitrate	Phosphate	Nitrate and Phosphate	Control- No nutrients
Average ug of	29.24	11.46	8.87	9.15
chlorophyll at Pettys				
site				
Average ug of	49.14	24.72	28.93	18.76
chlorophyll at Hayo-				
Went-Ha site				

Table AA: Comparison of chlorophyll in different nutrient enriched agar at 2 sites in Torch Lake.

Agar	Nitrate	Phosphate	Nitrate and Phosphate	Control- No nutrients
Combined Average	100,827	53,981	129,979	57,369
Total Cells				

Table AB: Comparison of cell density in different nutrient enriched agar.

The ideal growing conditions in water bodies occur when there are 16 nitrogen atoms to 1 phosphorus atom. Torch Lake, a traditionally clear oligotrophic lake typically has low amounts of phosphate available to grow aquatic plants and algae. This condition is called phosphate-limiting. However, when phosphate levels increase, nitrogen can become limiting to algal growth.

The algae on the nutrient-diffusing substrates were identified and counted under the microscope, the dominant organisms were diatoms. The five most common algae species identified on the substrates are listed in (Table AC.) Of these, *Epithemia* is one species of diatoms that can provide its own nitrogen (by nitrogen fixation) and can grow well under nitrogen-limiting conditions or conditions where there is not enough nitrogen available to use all the phosphorus. Additionally, 8 diatom species in the Torch Lake algae samples are new to science and will be described in a subsequent publication.

Agar	Nitrate		Phosphate		Nitrate and Phosphate		Control-No nutrients	
Number of Diatoms								
Achnanthidium	14,892	15%	14,472	27%	33,032	25%	14,165	25%
Centric	4,159	4%	3,745	7%	24,899	19%	2,421	4%
Naviculoid	16,877	17%	6,820	13%	4,403	3%	8,153	14%
Nitzschia	14,870	15%	11,027	20%	16,361	13%	9,790	17%
Ulnaria					1,163	0.9%	1,466	2.6%
Mastogloia	4,150	4%						
Pseudoanabaena			1,201	2%				
Epithemia	1,128	1%	154	0.3%	188	1%	253	0.4%
Total cells	100,827		53,981		129,979		57,369	

Table AC: Comparison of predominant algal genera in different nutrient enriched agar. All are diatoms except *Pseudanabaena* which is a cyanobacterium. Photo AB is an image of *Nitzschia*.

As increases in algal growth appears to be benthic (growing on the bottom) in Torch Lake rather than planktonic (growing on the water surface or in the water column), the nutrient source does not appear to be coming from storm water runoff. Instead, the source of the nutrient phosphorus could be the interstitial water or water between the grains of sands. To explore where the phosphorus may be coming from, water samples were taken of the surface water and between the sand grains using a hand vacuum suction method at 5 sites around the lake. At all sites, phosphate was higher in the interstitial water than in the surface water (Table AD.)

Water Sample Location	Phosphate (P0 <sub>4</sub> -P) ug/L Surface Water	Phosphate (P0 <sub>4</sub> -P) ug/L Interstitial Water
Clam River Dockside	3.28	7.79
Alden Harbor	4.26	13.48
Lake Ave	4.28	5.76
Sand Point	4.35	9.98
Eastport DNR	3.37	17.52
Average	3.91	10.91

Table AD: Comparison of Surface water and interstitial water at 5 sites around Torch Lake.

The larger quantity of algae in the nitrate-enriched agar, the diatom community containing nitrogen-fixing species and the higher amounts of phosphate in the interstitial water compared to surface water, all point to the conclusion that phosphorus is entering Torch

# Lake, changing the lake bottom environment from a phosphate-limited condition to a nitrogen-limited one.

The second Three Lake Association sponsored experiment is still on-going and in the laboratory analysis stage with Dr. Jan Stevenson at Michigan State University. Questions asked in this study are: 1) Are the benthic algae communities at different locations around Torch Lake the same?

2) Is groundwater entering Torch Lake a potential source of benthic algae nutrients?

Results are expected later this year and will be summarized and posted on the Three Lakes Association website: http://3lakes.com/. A presentation of the 2015 Golden Algae Study is planned for June 2016.

# OCTOBER, 2015

# **Investigation of Torch Lake Benthic Algal Outbreaks**

## Dr. Rex L. Lowe Professor Emeritus of Biology Bowling Green State University & Visiting Professor, University of Michigan Biological Station

## **INTRODUCTION**

In the summer of 2014 I was contacted by a resident of Torch Lake, Trish Narwold, asking if I could help in the identification of a local algal outbreak that was being referred to as brown crud (Figure 1).



Figure 1. Benthic algal growth in Torch Lake, summer 2014.

This outbreak was apparently a relatively new phenomenon on the normally pristine white sands of Torch Lake. I examined several of the samples under the microscope and determined that it was a healthy diatom community with none of the species obviously indicating gross pollution. However, there were large populations of the diatom *Epithemia*. This diatom is one of few that has internal endosymbiotic blue-green algae and is capable of fixing elemental nitrogen (Figure 2).



Figure 2. Benthic algae, 2014 with 5 large cells of *Epithemia*, other diatoms and the desmid *Cosmarium*.

*Epithemia* is usually most abundant in habitats where the atomic ratio of nitrogen/phosphorus is less than 16 and is an indicator of nitrogen limitation of algal growth. The abundance of this diatom on the bottom of Torch Lake led me to believe that the benthic algal growth was being stimulated by phosphorus-rich groundwater. Phosphorus-rich groundwater would lower the nitrogen/phosphorus ratio and select for nitrogen fixing diatoms. We assumed groundwater was the source of these nutrients since the diatom growth appeared in patchy strips across the bottom of the lake and there seemed to be no increase in planktonic algal

growth which might indicate that the source of nutrients was coming from surface runoff or precipitation. With this information we designed an experiment employing nutrient diffusing substrates (NDS) to be deployed on the bottom of Torch Lake in two different areas. The experiment was designed to test the hypothesis that with increased phosphorus fluxes on the bottom of Torch Lake, nitrogen had become limiting to benthic algal growth.

### **MATERIALS AND METHODS**

A research team from the University of Michigan Biological Station (UMBS), Dr. Rex Lowe, Dr. Pat Kociolek and Kristel Sanchez (a University of Michigan graduate student) scouted Torch Lake on July 1, 2015 to determine an ideal location to deploy the NDS. Two areas were chosen that were likely to be protected from potential vandalism, a site in front of the home of cooperating landowners, the Pettys and a site at a YMCA Hayo-Went-Ha summer camp.

Substrates (NDS) were constructed using 6 inch pieces of 4-inch diameter PVC pipe. The PVC pipe was sealed at one end with a petri plate and filled with a solution of 2% agar containing nutrients. Four treatments were constructed; 2% agar with 0.5 molar sodium nitrate, 2% agar with 0.5 molar sodium phosphate, 2% agar with 0.5 molar sodium phosphate and 0.5 molar sodium nitrate, and a control containing just 2% agar. An open space at the top of each substrate was filled with 2 cm of sand collected from the bottom of Torch Lake. This was done to conform with the natural sand substrate in Torch Lake such that nutrients defusing from the NDS would influence growth on the sand. Three replicates of each of the four treatments were constructed resulting in 12 NDS at each site. The substrates were placed in wire mesh baskets and deployed on the bottom of Torch Lake on July 6, 2015 in approximately 1.5 m of water (Figure 3).



Figure 3. NDS deployed at "Pettys" site in Torch Lake, July 6, 2015.

The NDS were retrieved after 20 days of exposure and taken back to the laboratory at UMBS for analysis. A strong storm with winds from the north had disturbed the substrates previously to the collection day rendering some of the replicates compromised. The sand on some of the substrates had been pushed off by high wave action during the storm. The substrates were still analyzed with the understanding that this disturbance may have altered the results. Quantitative measures of chlorophyll density were obtained by standard methods from each of the substrates. Chlorophyll is used as a proxy measure of biomass accumulation on the NDS. In addition, quantitative subsamples of the periphyton community from each substrate were quantified at 400 X magnification on a research grade compound microscope to determine community structure and cell density. Subsequently, on August 6, 2015 water samples from surface and interstitial benthic sand were collected and analyzed at UMBS by standard methods.

### RESULTS

At both locations, Pettys and YMCA Hayo-Went-Ha, the sodium nitrate substrates

had significantly higher levels of chlorophyll than the other three treatments (Table 1, Figure 4).

	µg of chlorophyll a by treatment								
	Petty				YMCA				
	N+	P+	N+P	Contro		N+	P+	N+P	Control
	38.07	19.69	21.04	18.69		74.97	35.84	30.06	11.43
	29.17	3.01	0.22	5.70		39.41	33.96	5.89	31.05
	20.49	11.69	5.35	3.05		33.04	4.37	50.84	13.81
Average	29.24	11.46	8.87	9.15		49 14	24.72	28.93	18.76
SD	8.79	8.34		8.37			17.65	22.50	10.70

Table 1.  $\mu$ g of chlorophyll (a)/cm<sup>2</sup> on the NDS from Pettys and YMCA Hayo-Went-Ha following 20 days of exposure on the lake bottom. Mean and standard deviation of chlorophyll density are listed at the bottom of the table.

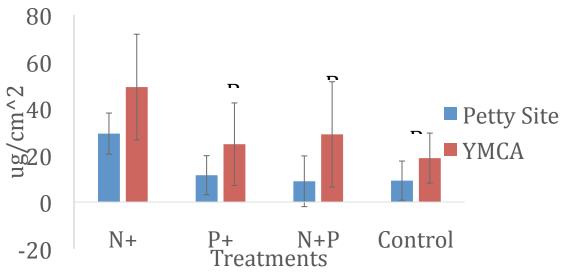


Figure 4. Chlorophyll response to NDS at Pettys & YMCA Hayo-Went-Ha sites.

This indicates that nitrogen now appears to be limiting for the growth of benthic algae in Torch Lake. The microscopic analysis of the samples showed a similar trend. The community was extremely species rich. We observed over 140 diatom species, eight of which are new to science and will be described in a subsequent publication. The nitrogen treatments with or without phosphorus generated higher Cell densities (Table 2).

Averages across b	ooth sites			
	N+P	P+	N+	Control
Achnanthidium	33032	14472	14892	14165
Centric	24899	3745	4159	2421
Cymbelloid	640	114	664	1023
Epithemia	188	154	1128	253
Eutonoid		44	73	
Fragilaria	708		662	547
Gomphomena	175		112	188
Mastogloia	139	31	4150	106
Naviculoid	4403	6820	16877	8153
Nitzschia	16361	11027	14870	9790
Ulnaria	1163	527	1941	1466
Other Diatoms	33736	14590	26271	18878
Other algae	13606	1299	13159	
Pseudoanabaena	930	1201	1944	379
Total cells	129979	53981	100827	57369

Table 2. Means of cell densities by species on NDS across both site, Pettys and YMCA Hayo-Went-Ha. Means of total cell densities appear at the bottom of the table. Images of 2 Genus can be found in Photo A and Photo B at the end of the report.

Because of storm-induced variability among the replicates these differences are not statistically significant. However, the trend is clear that nitrogen addition leads to higher Cell densities.

Chemistry data are presented in table 3.

Sample ID	PO4-P	NO3-N	NH4-N
L001AF-Clam Dockside surface water	3.28	177.2	19.1
L001AF-Clam Dockside interstitial water	7.79	92.1	172.2
R004AA- Clam River surface water	3.84	136.2	24.7
L001AL- Alden Harbor Creek surface water	4.26	358.9	18.8
L001AL- Alden Harbor Creek interstitial water	13.48	146.8	119.2
L001AP- Lake Ave. surface water	4.28	195.1	19.5
L001AP- Lake Ave. interstitial water	5.76	176	188.2
L001AT- Sand Point surface water	4.35	189.1	42.2
L001AT- Sand Point band ground water	9.980	138.90	1175.00
L001AT- Sand Point NO band ground water	10.570	65.80	1055.00
L001AC- East Port DNR surface water	3.370	194.00	21.50
L001AC- East Port DNR interstitial water	17.52	60.6	12800
L001AJ-Petty surface water (08/05/15)	3.84	210.4	20
L001AJ-B Petty well water (08/05/15)	5.9	29	32.8
Cozy Cottage (08/07/15)	4.73	ND	14.5

Table 3. Chemical data from Torch Lake benthic areas, summer 2015 reported as  $\mu g/L$ .

It is interesting to note that in every instance interstitial phosphate is higher than surface water phosphate. This indicates that as phosphorus enters the lake from groundwater it is being sequestered by the benthic algal growth.

# CONCLUSIONS

We feel we have shown that phosphorus entering Torch Lake from interstitial water on the sandy lake bottom is stimulating benthic algal growth. Benthic diatoms and other algae have become dense enough that nitrogen appears to have become the limiting element at the surface water sediment interface. Both chlorophyll data and cell density data support this conclusion. This is unusual for large oligotrophic lakes in northern Michigan which are traditionally phosphorus limited. Many questions remain unanswered. What is the source of this phosphorous? Why is this happening now? What has changed? Is this a Torch Lake phenomenon or is it more wide-spread in other lakes in the area?

#### **FUTURE RESEARCH**

If funding is available we propose to repeat this experiment in Torch Lake at different sites at a depth below the wave disturbance zone using a more stable NDS device. In addition we would like to export our experiments to other nearby lakes experiencing the same phenomenon.

#### **ACKNOWLEDGEMENTS**

I thank hard-working members of the algae team from UMBS, Kristel Sanchez and Pat Kociolek and the Pettys and Jim Austin for providing access to research sites. Also thanks to Becky Norris for insight into algal issues, Drew Narwold and the Hoadley brothers for field assistance. Finally, and especially thanks to Trish Narwold for enthusiastic support and seemingly tireless energy devoted to environmental issues on Torch Lake.

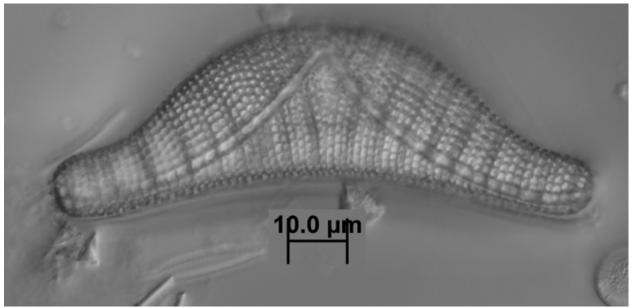


Photo A: Microscopic image of *Epithemia smithii* identified in a sample collected from YMCA Hayo-Went-Ha.

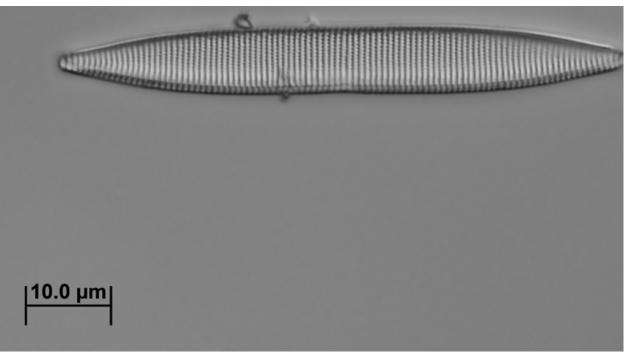


Photo B: Microscopic image of *Nitzschia bruno* identified in a sample collected from Pettys.