2015 Study of Golden Brown Algae on the Bottom of Torch Lake
by Professor R. Jan Stevenson, Michigan State University, March 29, 2016

Executive Summary & TLA's Water Quality Team's Comments
by TLA's Water Quality Team
April 15, 2016

In the spring of 2015, Three Lakes Association (TLA) retained the services of two professors (internationally recognized diatom experts), Rex Lowe and R. Jan Stevenson, to work with TLA's volunteers to design and conduct an investigation of golden brown benthic algae (GBA) in Torch Lake. Their complete scientific reports[1,] [2] are available on the TLA web site (3lakes.com). This paper summarizes the findings from Professor Stevenson's report, compares some of the current findings with earlier TLA work, and summarizes the TLA plans for further investigating the GBA in the watershed.

Professor Stevenson's report[1] includes the analysis of 140 water samples (nutrients in groundwater and surface water samples) and the characterizations of 20 samples of benthic algae;

Golden Brown Algae

Some of the benthic algae samples were sand with very visible assemblages of GBA (dense), other samples were sand without visible benthic algae (sparse), and some samples were the scrapings from rocks covered with GBA. The samples were collected by TLA's Water Quality Team under the direction of Professor Stevenson. The water samples were analyzed by researchers in MSU's Hydrogeology Department, and the algae samples were characterized by researchers in MSU's Zoology Department under the direction of Professor Stevenson.

The purpose of the 2015 investigation was to improve our understanding of possible factors that may be triggering and stimulating these near-shore outbreaks of GBA around Torch Lake in recent years. One of the most basic questions we were attempting to answer is "What's changed in the last 5 to 10 years that may have contributed to the GBA outbreaks?" It is generally acknowledged that GBA was far less apparent on the rock and sand around Torch Lake 10 years ago. It is also generally acknowledged that GBA is now being observed in the near-shore areas of most every lake in northwestern Michigan. The current study did not investigate the potential contributions of higher occupancy in lake shore residences or warmer near-shore water temperature to the growth of the GBA. We hope to obtain information that will lead to science-based strategies for reversing the occurrence of these GBA outbreaks and thereby return Torch Lake to its historical beautiful turquoise appearance.

The sampling strategy was designed to screen, among the various possible contributing factors that could help explain recent GBA observations, an increase in

available nutrients on the floor of the lake. Of particular interest were the large concentric patterns of GBA growth seen in aerial photographs.   The reasons for focusing the sampling strategy on the following three areas of Torch Lake were twofold, first to learn as much as possible about the relationship between groundwater nutrients and the GBA with the limited available funds and volunteer time, i.e. maximize TLA's "bang for the buck" and, second, to compare findings in more developed and less developed areas. We were looking for hints of noticeable differences in groundwater nutrients between, and within, areas with and without GBA, and over a summer growing season. The sites most heavily studied included:

1. **Petty property**…4 groundwater wells (piezometers) installed in a rectangle pattern, approximately 30 by 50 ft in water about 1 to 4 ft deep.
2. **Camp Hayo Went Ha property**….4 piezometers similarly installed in a rectangle pattern
3. **Gourley property**…1 piezometer.

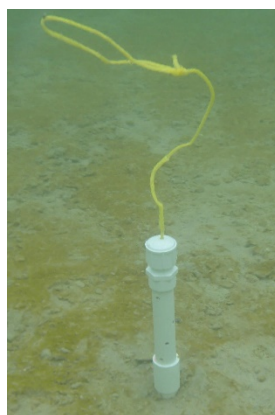A DEQ permit was obtained for these activities.

During the summer, in an effort to obtain more groundwater samples from different areas around the lake, we employed the following three additional techniques for collecting groundwater or interstitial (below the lake bottom) water samples:

(1) Peepers (passive dialysis tubing bags housed in perforated PVC pipes)
(2)   Reusable Temporary piezometer
(3) Turkey baster

In designing the sampling strategy, in conjunction with Professors Stevenson and Lowe, we were also researching the nutrient limitations on the GBA growth using the nutrient-diffusing substrate methodology developed by Dr. Lowe[2].
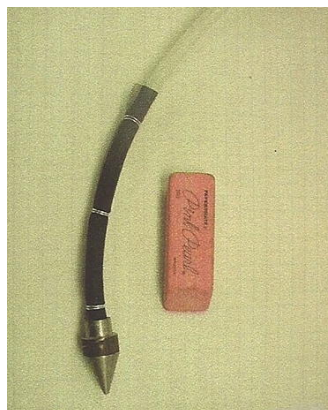
Piezometer point & screen   *Peeper in lake floor*   *Temporary piezometer point*

2015 Findings and some discussion of findings:

We learned that these GBA assemblages (collection of types of algae) are made up of more than 100 different species of diatoms[1].   At least eight of these diatom species have not previously been reported in the scientific literature[2]. It is our understanding that one of the visiting professors involved in this study is preparing a manuscript to a journal that specializes in diatoms. Although we have only a vague understanding of the role that each diatom species plays in these GBA

assemblages, it is our general understanding that some families of diatoms can fix nitrogen out of the atmosphere, others can extrude substances to help build the GBA assemblage structure, and still other types contain different photosynthetic chemicals and pigments[1].   Species of benthic diatoms may be similar to or different from planktonic algae that live suspended in lake water and are associated with algae blooms. We learned from sequential monthly aerial photographs that the patterns of GBA distribution expanded over the summer.

Microscopic view of some diatoms

We also learned a lot about efficient methods for collecting groundwater samples. A vacuum pump and a clean jug worked well to slowly collect groundwater from the piezometers. A conductivity meter was useful for determining in the field whether the collected sample was similar to or different from the lake water since higher values are found in groundwater than in lake water. Our (semi-permanent) piezometers were installed about two feet below the surface of the lake floor. "Peepers" (perforated PVC pipes with passive dialysis tubing bags) buried under the lake bottom, was an alternate method for collecting groundwater samples we also evaluated in this study.

We learned that the concentrations of phosphorus in groundwater were about two to ten times higher than in lake water. And we learned that these concentrations may be higher now than they were in the 2005 TLA groundwater study[3]. Based on measured concentrations of groundwater total phosphorus found in 14 groundwater wells in 2005[3] (average 21.6 ppb with range of 1.6 to 72 and median value of 22.8), the concentrations appear to be higher in 2015 (average 33.8 ppb with range 2.5 to 209.9 and median value 20.5). (We anticipate that the high variability in groundwater total phosphorus found both in 2005 and in 2015 will be reduced in future sampling by field filtering the samples, thereby eliminating potential contamination of samples with non-bioavailable phosphorus in sediment.) These findings are consistent with the hypothesis that some species of GBA diatoms may be more-or-less dormant at the lower and triggered to grow at these higher concentrations of phosphorus, thereby becoming visible on the surface. Once some of the dormant diatoms have been triggered to grow <u>on the sand and rocks</u>, rather than <u>in the sand</u>, additional growth may be encouraged by other species of diatoms with the capacity to increase nitrogen availability through fixing it directly from the atmosphere[1].

We observed that, despite the large variability within individual sites in groundwater phosphorus concentrations, there appeared to be no obvious differences in groundwater phosphorus concentrations among the locations studied. Additionally, no trends in changes over time were observed. With a single exception, the measured nitrogen concentrations were similar in groundwater and lake water.

Next Steps…Intriguing Questions to be addressed in a 2016 Study:

An intriguing finding in this study was the similarity of diatom cell densities in the sand samples collected from various locations around the lake; some with visible dense GBA, and others with sparse to no visible GBA growth[1]. One possible interpretation is that diatoms living in sand may be capable of becoming GBA on the sand surface when provided enough groundwater nutrients to trigger their movement to the surface of the sand. We intend to follow up on this observation by collecting and characterizing more samples of sand with and without visible GBA, which may include some core samples of sand collected at different depths.

We are intrigued by the possibility of comparing patterns of groundwater entering the lake with patterns of GBA growth. When the lake water is cold (40 to 45 deg F), and the groundwater entering the lake is about 5 to10 degrees warmer (50 to 55 deg F), and the lake is very calm (no wind or waves), we intend to capture infrared and visible light images of the lake with sensors & cameras mounted on a drone.

In the 2016 study, we plan to collect many additional surface water and groundwater samples as well as drinking well water samples for determination of phosphorus and nitrogen and also collect additional benthic algae samples for analysis. In addition we plan to measure the following substances, potential indicators of human source contribution of the nutrients stimulating the lake bottom GBA growth:
- ✓ Chloride
- ✓ Caffeine
- ✓ Boron

We will obtain aerial photographs to track the locations and spread of GBA and monitor lake floor temperature throughout the summer growing season. We are considering the possibilities of collecting lake floor core samples to study diatom deposition over time. We are also considering soil sample chemical analysis and dye tracking from septic distribution fields in selected locations.

References

1. Stevenson, R. Jan, "2015 Study of Golden Brown Algae on the Bottom of Torch Lake", professor of hydrogeology, Michigan State University Department of Integrative Biology and Center for Water Sciences, April 2016, beginning on page 5 of this document.
2. Lowe, R., "Investigation of Torch Lake Benthic Algal Outbreaks", visiting professor at the University of Michigan-Biological Research Station in Pellston, Michigan, October 2015; *Executive Summary* by Narwold, T. and Branson, D., Three Lakes Assn, Nov 2016: Investigation of Torch Lake Benthic Algal Outbreaks with Executive Summary
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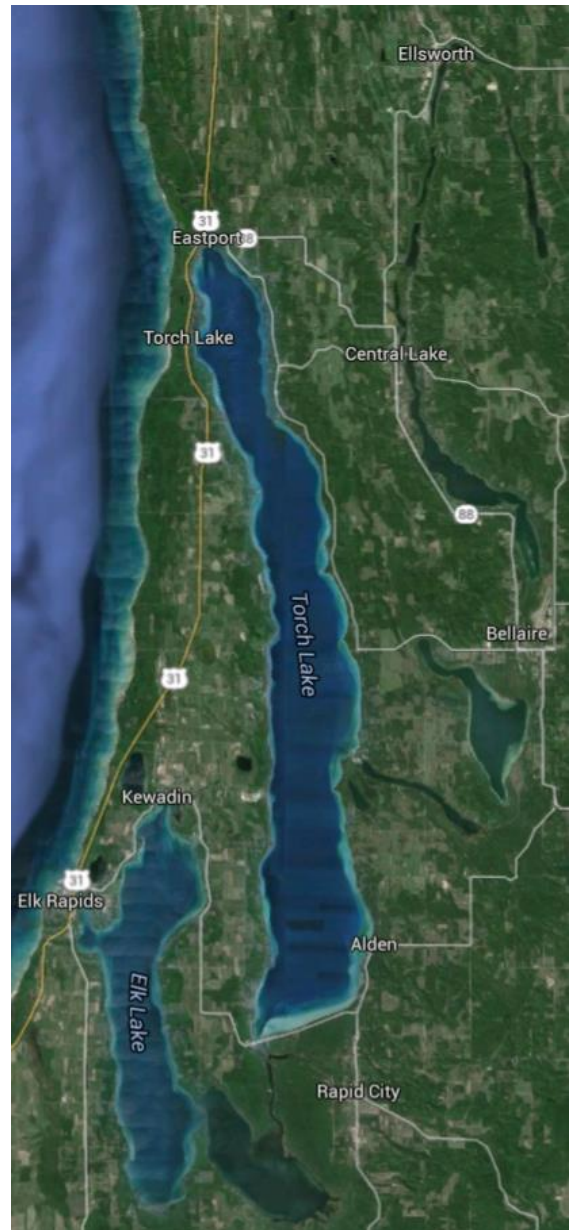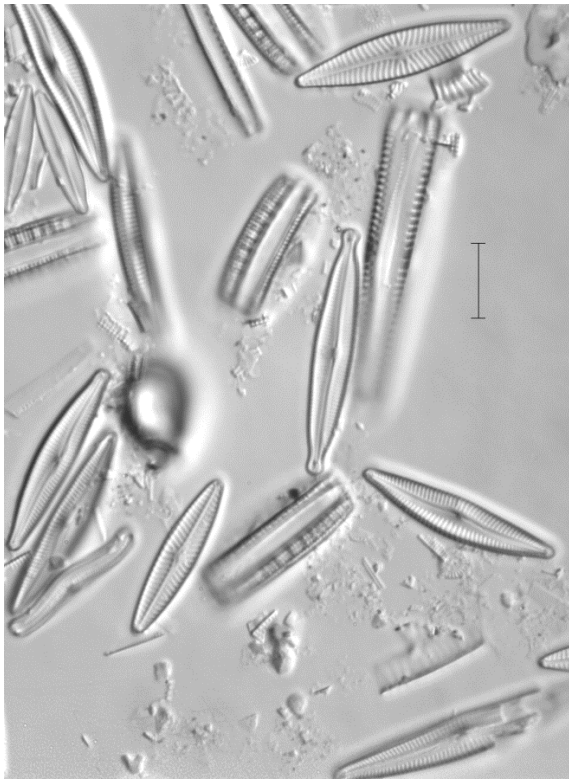
East Lansing, MI 48824

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Image Credits: Stevenson's image of common benthic diatoms in Torch lake (scale bar = 10 µm). Map of Torch Lake and surrounding watershed by Google®.

Summary

Groundwater and surface water can be sources of current and past nutrient contamination to lakes, streams, and wetlands. Changes in algae on the bottom of lakes, at the interface of the groundwater and surface water, may be an early warning indicator of lake contamination and the source of nuisance algae that affect aesthetics and potentially human health.

Increasing amounts of golden brown algae (GBA) have been observed in Torch Lake over the past 10 years. After alternative hypotheses were discussed, a study was initiated to evaluate the role of groundwater contamination and stimulation of golden brown algae.

Groundwater has higher phosphorus concentrations than surface water, but groundwater has lower nitrogen concentrations than surface water. Seasonal changes in groundwater were not observed. Little spatial difference was observed in groundwater because of the limited spatial extent of sampling groundwater.

Phosphorus and nitrogen concentrations in groundwater and surface water are likely sufficiently low that they independently and interactively control algal growth.

Cell densities of benthic algae in Torch Lake are as great in areas of sand that have a visual coating of golden brown algae as they are in areas with little visual coating of golden brown algae. The GBA visual effect is likely due to a change in location of algae from among and attached to sand grains to a matrix of algae on the sand surface.

Species composition of diatoms did not differ among samples from dense GBA sand, sparse GBA sand, and rock habitats.

Diatom metrics used in ecological assessment of nutrient conditions were related to shifts in species composition among samples, but were not related to cell densities of algae in and on sand or depth of water, which is an indicator of distance from shore and the potential sources of groundwater contamination.

Limited areas of sampling, small numbers of samples, seasonal variation in the development of GBA, and extensive areas of dense GBA during late summer are the likely reasons for uncertainty in relationships among visually apparent algal density, measured algal density, diatom metrics of nutrients, and groundwater chemistry.

Future studies should continue the effort to evaluate groundwater nutrient relationships to human sources, variation in groundwater nutrient concentrations in Torch Lake, and responses of algae on the bottom of Torch Lake to groundwater nutrient concentrations with experimental and survey approaches to establish cause-effect relationships for the dense accumulations of golden brown algae.

Introduction

Many Torch Lake residents are concerned about greater accumulation of golden brown algae (GBA) on the bottom of Torch Lake than in the past. The accumulation is reported to occur during summers and to be a relatively recent phenomenon. The accumulation is characteristically golden brown and seems to be accumulating on top of the sand bottom. Accumulations seem to be observed more in shallow than deep water, but they do extend to deeper water later in the summer. There are many hypotheses for why this could occur. Our initial task was to review some basic algal ecology, to develop a set of alternative hypotheses, to select the most likely hypotheses, and then to test as many as possible.

Algae occur naturally in the water column and on the surfaces underwater, including sand. All three of the major taxonomic groups of algae in freshwater ecosystems, cyanobacteria, green algae, and diatoms, are adapted to live in the water column (planktonic) or on surfaces under water (benthic). Within these major groups of algae, different species of these major groups are adapted to be planktonic and benthic. Of those species that live in benthic habitats, some species are better adapted to live on rocks, plants, sand, fine sediments, or plants. Therefore, the species that accumulate on top of the sand bottom may be different than those that live on sand grains and that are not visually apparent on the surface.

Algae are highly sensitive to environmental change. We know a lot about which species occur in different habitats as a result of almost 150 years of research on the taxonomy of algae and characterizing the habitats in which species occur. This research has been greatly advanced in the last 20 years by large scale surveys of the status and trends in water quality that have used algae, as well as other biota, to determine whether aquatic resources have changed and the human factors that could be responsible for those changes. Biological assessment is used in freshwater assessments because changes in biodiversity, fisheries productivity, or algal blooms are problems that we want to assess with direct measurement. In addition, species composition of assemblages changes slower than physical and chemical conditions. So the biota provide a temporally integrated assessment of water quality when we know species environmental sensitivities and tolerances. The USEPA and many states use algae in environmental assessment of streams, lakes, and wetlands. We will use results of one of these surveys, the 2007 National Lakes Survey, to provide diatom species environmental sensitivities which will be used to interpret changes in species composition of benthic algae in Torch Lake associated with the GBA.

Many environmental factors affect growth rates of algae and variation in those growth rates among species. Algal growth rates are most directly regulated by nutrient concentrations, light intensity, and temperature. Different species grow fastest in different nutrient, light, and temperature conditions. So changes in these conditions in Torch Lake could cause accumulation of different species that produce GBA. In addition, water chemistry, physical disturbance, and biota in lakes can affect algae growing on the bottom. pH and conductivity are very important factors regulating species composition of algae. Many fish and invertebrates eat algae, or disturb their accumulation on the bottom. Viruses and bacteria cause diseases in alga. Recent invasions of dreissenid mussels have been associated with increases of benthic macroalgae in lakes, like *Cladophora*, because they increase water clarity and light penetration to the bottom, which is particularly an important limiting factor for large accumulations of green macroalgae.

So what has likely changed in Torch Lake in the last 10 or 20 years that would increase the accumulation? We need to consider what has changed, the magnitude of change, and the likelihood

that it would generate the observed change in benthic algae. Many factors could cause the problem, but given likely changes in Torch Lake, what could cause GBA?

Our favorite hypothesis is that groundwater contamination with nutrients from septic tanks or agricultural activity would be causing increases in benthic algae. Groundwater enrichment would increase benthic algae before it would increase planktonic algae because of the proximity of benthic algae to exposure by groundwater, the concentration of groundwater at the sediment water interface, and dilution of groundwater when mixed in the water column.   Groundwater contamination would also be greater near shore than in deeper waters, so that it could cause faster accumulation in shallow than deeper water and lower abundances of GBA in deeper water. Changes in water transparency could cause the lower abundances of GBA in deeper water, but benthic microalgae grow well under low light levels. Changes in water temperature could also cause the problems. Many alternative hypotheses exist, but knowledge of relative levels of responses to likely changes in environmental conditions in Torch Lake leave increases in groundwater nutrient concentrations as the most likely cause of GBA.

Groundwater and surface water can be sources of past and current nutrient contamination to lakes, streams, and wetlands. Changes in algae on the bottom of lakes, at the interface of the groundwater and surface water, may be an early warning indicator of lake contamination that causes nuisance algae that affect aesthetics. In the worst cases, substantial macroalgal accumulations in recreational areas can threaten human health.

This study of golden brown algae on the bottom of Torch was designed and initiated to test the hypothesis that the quantity and species composition of benthic algae in Torch Lake were related to changes in groundwater chemistry. The overall research project on GBA in Torch Lake had two complementary efforts. The first was a survey of benthic algal quantity and species composition as well as the chemistry of groundwater and surface water which we expected to show two main relationships: 1) benthic algal quantity was positively related to groundwater nutrient concentrations and 2) benthic algal species changed from species able to grow in low P or N concentrations to species needing high P or N concentrations to grow with algal density on sand. The second research effort was an experiment that released P and N below sands (via nutrient diffusing substrata, NDS) which would enable determining: 1) whether benthic algal accumulation was limited by P, N, or both P and N supply and 2) which benthic algal species were limited by P, N, or both P and N enrichment to grow. Thus survey of benthic algae and nutrients around Torch Lake and the nutrient diffusing substrate experiment enabled causal analysis of whether groundwater nutrient enrichment could affect benthic algal accumulation and which species would respond to nutrient enrichment. The individual species responses in the experiment could then be used to corroborate and potential provide greater detail to existing knowledge of differences in algal species' nutrient requirements to help with diatom metrics of nutrient conditions in benthic habitats of Torch Lake.

This report is a review of the methods, results, and significance of the first of the two complementary 2015 Torch Lake GBA research efforts, the survey of the species composition and quantity of benthic algae and nutrient chemistry of groundwater and surface water in Torch Lake. Dr. Rex Lowe submitted a separate report for the NDS study.

Sampling and General Laboratory Methods

Benthic algae, groundwater, and surface water samples were collected at numerous locations around Torch Lake to relate benthic algal accumulation to differences in groundwater or surface water chemistry. In addition, we tested the hypothesis that recent GBA development could be related to changes in Torch Lake temperature using satellite imagery that measures surface water temperature.

Nutrient assays

Nutrient concentrations in groundwater and surface water chemistry were sampled and analyzed to determine whether nutrient concentrations were higher in areas with higher densities of homes and people. Groundwater was mostly sampled with piezometers (Table 1) and pumped into acid-rinsed polyethylene bottles. A Solinst temporary piezometer, and peepers were also used to sample groundwater. Surface waters were collected by subsurface grab samples in acid-rinsed polyethylene bottles. Often, pairs of filtered and unfiltered groundwater samples were collected. Most surface water samples were unfiltered. Four surface water samples were filtered. Filtering passes water through a 0.45 μm pore-size filter before placement water goes in polyethylene bottles, which removes bacteria, algae, small detritus particles, and inorganic sediments.

Samples were collected from 1 to 3 times during summer 2015 from nearshore zones at four locations in Torch Lake: Camp Hayo-Went-Ha, the Petty Property, the Gourley Property, and Cozy Point. Peeper samples were collected at Becky's Beach, the Ozanne Property, and the Gourley property. Nearshore areas at Camp Hayo-Went-Ha and Becky's Beach were expected to have relatively low nutrients in groundwater because of low density of people along the shoreline. We also expected to see changes in ground water nutrient concentrations during the summer due to seasonal changes in groundwater head pressure from groundwater sources around Torch Lake. In most cases, more than one sample was collected at a location to assess variation in conditions at a site and to evaluate differences among sites and times with replicate samples.

Groundwater and surface water samples were assayed for total phosphorus (TP), nitrate-nitrogen (NO_3-N), and ammonia-nitrogen (NH_3-N). Total phosphorus is usually measured in lake water as an indicator of bioavailable P, because P is rapidly taken up by algae and bacteria and then released from them when they die. So soluble reactive phosphorus concentrations usually underestimate P availability. In some cases, TP was measured on filtered lake water. In that case, the dissolved organic and dissolved inorganic fractions of the water were assayed, where dissolved fractions are defined as anything passing through the 0.45 μm filter. Total phosphorus assays in filtered water will be referred to as dissolved phosphorus (DP). Both nitrate and ammonia are bioavailable to algae as inorganic N source. They were added together and reported as dissolved inorganic nitrogen (DIN).

TP was measured as soluble reactive phosphorus concentration using a spectrophotometer, the standard ascorbic acid method, and after digestion of organic phosphorus (dissolved and particulate in unfiltered samples) with persulfate to soluble reactive phosphorus. Nitrate was measured using a standard cadmium reduction method on a Lachat® autoanalyzer. Ammonia was measured using the standard ammonia salicylate method on a Lachat® autoanalyzer.

Water chemistry analyses have not been completed for the peepers.

Benthic algal assays

45 benthic algal samples were collected from sand and rock bottom areas, at depths ranging usually from 3-6 feet, along the shorelines of Torch Lake (Table 2). 20 of these samples were identified as priority 1-3 samples because they would enable comparison of the quantity and species composition of algae in areas where golden brown benthic algae were dense and sparse as well as compare algae on sand versus rock. We could not analyze chlorophyll a in the samples, which is an indicator of algal quantity because samples thawed during shipping. We did use microscopic counts of algal cells measured as cells/cm^2 of lake bottom, which is another measure of algal quantity. Microscopic counts also provided measures of species relative abundances. Two types of microscopic counts were done. One was a detailed taxonomic analysis of diatoms, which are often the most abundant and diverse kinds of algae in freshwater habitats, which they were in Torch Lake. The second microscopic assay of benthic algae was a soft algal count, called that because diatoms (with "hard" glass cell walls) are only identified as living and dead in soft algal counts. Non-diatom algae are counted and identified to the lowest taxonomic level possible in soft algal counts. These microscopic assays of algae were done with methods used by the USEPA for their national assessments of benthic algae.

Nutrient diffusing substrata were employed for the experiment to determine whether benthic algal accumulation was limited by nitrogen, phosphorus, or both nitrogen and phosphorus. The results and report for the NDS experiment are complete. Dr. Rex Lowe provided a report for most of these results. Detailed assays of benthic diatoms from the NDS experiments were done after that report. They did not provide sufficient information about species nutrient requirements to aid with interpretation of the results of the survey of benthic algal patterns around Torch Lake. They will not be discussed further in this report.

The details of analysis methods, results, and discussion of the water chemistry and benthic algal survey are below. I have not embedded the many tables and figures of results in the report so the report would be easier to read. The tables and figures are presented at the end of the text of the report.

Data Analysis Methods, Results, and Discussion

Nutrient Chemistry of Torch Lake

I used a set of guidelines to interpret nutrient concentrations and two nutrient ratios that were calculated to characterize nutrient conditions. Algal growth rates have an asymptotic relationship with nutrients such that increases in nutrient concentrations in low concentration ranges produce proportionally great increases in growth rates until further increases in nutrient concentrations have no further positive effects on algal growth rates (Figure 1a). Relationships between benthic algae and water column nutrient concentrations are complicated by the mixing rates of surface and groundwater into the interstitial spaces within benthic algal mats. Nutrients in interstitial spaces within benthic algal mats are taken up more rapidly by algae and bacteria than they are replaced by nutrients leaking from cells or resupplied by diffusion or water mixing from groundwater and surface water sources. Thus, as benthic algal densities increase, higher and higher nutrient concentrations in surrounding waters are needed to produce maximum algal growth rates and a resulting maximum peak algal biomass (Figure 1b and c). Therefore, relationships between benthic algae and nutrient concentrations can characterized by the nutrient concentrations that are sufficient to produce maximum growth rates when algal biomass is low and the nutrient concentrations that are sufficient to produce peak algal biomass. Experiments in

recirculating artificial streams show growth rates of diatom dominated benthic algal assemblages reach 90% of their maximum value at 8 µg SRP/L and 86 µg DIN/L (Rier and Stevenson 2006), where SRP refers to soluble reactive phosphorus, which is a dissolved inorganic form of P and the most bioavailable form of P for algal uptake and growth. Peak algal biomass reaches 90% of its maximum when nutrient concentrations reach 38 µg SRP/L and 308 µg DIN/L. In this report, I use 8 µg P/L and 86 µg N/L as guidelines for the maximum nutrient concentrations that strongly affect benthic algal growth rates and 38 µg SRP/L and 308 µg DIN/L as guidelines for the maximum nutrient concentrations needed for producing peak benthic algal biomass.

There are issues for using these nutrient concentrations for guidelines when assessing benthic algal regulation in Torch Lake by surface water and groundwater supplies of nutrients. In general, these concentrations and the 15:1 molar ratio are probably lower than those that actually regulate benthic algae in lakes. The growth and peak algal biomass limiting nutrient concentrations above (Rier and Stevenson 2006) were determined in recirculating streams. Current increases the mixing of overlying water with interstitial water. Slower water currents in lakes than streams will likely increase the nutrient concentrations needed to produce maximum growth rates and peak biomass for microscopic benthic algae in lakes. There are no experiments similar to Rier and Stevenson (2006) that have been done for nutrient limitation of benthic algae in lakes. Another issue is we measured TP in unfiltered surface and groundwater samples, rather than SRP, so higher levels of TP are needed than SRP because particulate P is much less bioavailable in the short term than SRP. Total P is the sum of particulate P, dissolved organic P, and inorganic soluble reactive P. Dissolved P (DP), the dissolved organic P and inorganic soluble reactive P, was measured in filtered water samples. DP effects on algae are probably much more similar to those of SRP than TP because particulate P is usually a fairly large fraction of total P. So in general, my nutrient limitation guidelines are probably lower than those needed for lakes.

I also used 18:1 and 65:1 as molar ratios of N:P to indicate the boundary between N and P limitation. Experiments in lakes show that below the 18:1 molar ratio, N is in sufficiently short supply that phytoplankton growth is N-limited (Dzialowski et al. 2005).   When the molar ratio is greater than 65:1, P is in sufficiently short supply that phytoplankton growth is N-limited. Between the 18:1 and 65:1 molar N:P ratios, both N and P additions will stimulate algal growth. These guidelines were chosen because similar experiments have not been done in streams. The 15:1 molar ratio is the Redfield ratio, a ratio of the molar N and P concentrations in healthy algae. The fact that higher N:P ratios are needed to alleviate N limitation and generate P limitation may be related to the relatively higher retention of P in algal cells than N. Even in benthic algal assemblages, molar ratios of nutrients in benthic algal mats decrease over time, indicating algae leak more N than P.

I calculated an $NH_3:NO_3$ ratio to indicate low oxygen availability. NH_3 is usually converted relatively rapidly to NO_3 when O_2 is abundant in the water. So low $NH_3:NO_3$ in groundwater, for example, could indicate relatively anoxic conditions that is potentially caused by high biological oxygen demand and organic matter contamination.

The following presentation of results can get confusing without orientation, because I will present results of many chemical analyses with both filtered and unfiltered groundwater and surface water, in that order. First, I will compare filtered and unfiltered groundwater and surface water using average nutrient concentrations and nutrient ratios for a site for summer 2015. For each type of water chemistry sample, I will also evaluate whether N, P, or both N and P are below levels that would limit

algal growth. I will then evaluate whether nutrient concentrations, nutrient ratios, and relationship to guidelines for nutrient limitation differ among sites and times.

Boxplots (e.g. Figure 3) were used to evaluate differences in nutrient concentrations and ratios among sites and dates and to evaluate N and P limitation of benthic algae. In boxplots the median is the dark horizontal line in the box. The, 75th and 25th percentiles of observations at a site are the upper and lower boundaries of the box. The likely range of data is indicated by the whiskers, whose lengths are calculated as 1.5 times the distance from the median to the 25th and 75th percentiles of the data. Generally, if boxes of different groups of samples do not overlap, the differences are not likely to have occurred by chance, i.e. they are statistically reliable differences. Horizontal lines across the boxplots represent the nutrient limitation guidelines for P concentration (8 and 38 µg/L), N concentration (86 and 308 µg/L), and N:P ratios (18 and 65). Boxplot analyses were usually backed up by analyses or variance or linear regression to more quantitatively test hypotheses. These results were not presented to control complexity of the report and the time required to prepare it.

Comparing Groundwater from Piezometers and Surface Water. Average nutrient conditions were calculated per site and then the average across sites for each variable was calculated. This two-step averaging eliminated over influence of high number of samples from a single site on the comparison of groundwater and surface water. Samples with unusually high particulate P concentrations indicated contamination by sediments, so these samples were excluded from this analysis. Particulate P concentrations were estimated from differences in unfiltered and filtered samples from the same site, date, and water source.

Phosphorus concentrations were usually higher in unfiltered than filtered samples, but no significant differences were observed in DIN concentrations between filtered and unfiltered samples (Figure 3). The difference between phosphorus concentrations in unfiltered and filtered samples were the particulate phosphorus fraction (i.e. TP versus DP). The lack of difference in DIN concentrations in unfiltered and filtered samples indicated that particulate matter containing N was stable during preservation before analysis and no particulate matter fractions contributed to DIN.

Both TP (unfiltered) and DP (filtered) concentrations were higher in groundwater than surface water, and DIN concentrations were lower in groundwater than surface water (Figure 3). The relatively high $NH_3:NO_3$ ratio in groundwater indicated lower oxygen conditions in groundwater than surface water, which was likely related to organic matter and biological oxygen demand being higher in groundwater than surface water. These differences between groundwater and surface water could indicate contamination of groundwater by septic wastes, but it may also indicate natural background groundwater conditions and the accumulation and decomposition of planktonic nutrient sources in lake sediments. Distinguishing septic, natural groundwater, and planktonic sources of nutrients in sediments will be possible with future sampling of well water to assess background groundwater nutrient concentrations, assessment of high impact and low impact sites around Torch Lake, assessment of human contaminants in groundwater (i.e. caffeine), and assessment of nutrient gradients in sediments.

Nutrient concentrations and N:P ratios indicate that P supply from surface water would most severely limit benthic algal growth, with TP and DP concentrations well below the 8 µg/L growth limitation guideline. P concentrations in groundwater were higher and in the lower half of the range that would regulate peak biomass accumulation. DIN concentrations in both the groundwater and surface water were in the upper range of concentrations that would limit peak algal biomass accumulation based on

guidelines from the scientific literature. N:P ratios in both surface and groundwater were at or above the boundary that would indicate P limitation. Although the DIN:TP ratio in unfiltered groundwater had a mean close to the 65:1 N:P ratio indicating P limitation, if we account for leaving particulate N out of our N measurement, then actual TN:TP ratios should be greater than 65.

Unfiltered Surface Water. Measured nutrient conditions in Torch Lake surface water varied little among sites and dates sampled (Figures 4-7). Although replicate samples were not taken on many dates (indicated by solid lines rather than boxes in the figures), consistency in conditions among times and sites indicated little difference. Nutrient concentrations and ratios in samples taken in ankle (Ank) deep water were very similar to samples collected in deeper water at a location near the epicenter (Epi) of the piezometer grids at Camp Hayo-Went-Ha and the Petty and Gourley locations. The one notable exception was that "surface water" from a shoreline seep at Camp Hayo-Went-Ha had relatively higher TP concentrations, indicative of the characteristically higher TP concentrations in groundwater than surface water; but $NH_3:NO_3$ ratios were not conclusively higher than other surface water samples or as high as groundwater samples from piezometers.

Filtered Groundwater from Piezometers. Nutrient concentrations in filtered groundwater varied little among sites and dates, except for the Gourley location where DIN concentrations were very high (Figure 8). Nutrient concentrations were replotted without the Gourley location data to reduce the range of the Y-axis of graphs and thereby to increase visual evaluation of other differences among sites and dates and of relationships to nutrient limitation guidelines (Figure 9). Dissolved P concentrations in filtered groundwater were usually less than 8 µg/L at all sites and for all dates. DIN concentrations in filtered groundwater were usually less than 306 and greater than 86 µg/L at all sites and dates, which indicated moderate N limitation. Of course, DIN concentrations at the Gourley location were the exception because DIN concentrations were greater than 8000 µg/L. Ammonia composed almost all the DN at the Gourley location, therefore the $NH_3:NO_3$ ratio was very high at the Gourley location. The DIN:DP ratios in filtered groundwater were usually greater than 18:1 and sometimes greater than 65:1 at all sites and dates, indicating common N and P limitation and sometimes just P limitation of algal growth.

Unfiltered Groundwater from Piezometers. Nutrient concentrations in unfiltered groundwater varied little among sites and dates, except for the Gourley location where DN concentrations were very high and two early sample sets where TP concentrations were higher than usual (Figures 10-11). Again, nutrient concentrations were replotted without the Gourley location data to facilitate evaluating other differences among sites and dates, and relationships to nutrient limitation guidelines (Figure 12-13). Total P concentrations in unfiltered groundwater were usually greater than 8 and less than 38 µg/L at all sites and for all dates, except the higher values at Camp Hayo-Went-Ha and the Petty Property during the 7/22 and 8/15 sampling. Discussions with the field sampling team (usually Becky Norris, Dean Branson, and Trish Narwold) indicated these samples were likely contaminated by sediments that were taken up when pumping the groundwater.

DIN concentrations in unfiltered groundwater, like filtered groundwater, were usually less than 306 and greater an 86 µg/L at all sites and dates, except the Gourley location where concentrations were greater than 8000 µg/L. Again, ammonia composed almost all the DIN at the Gourley location, therefore the $NH_3:NO_3$ ratio was very high at the Gourley location. N:P ratios varied from less than 18:1 to greater than 63:1 for all sites and dates. If samples from Camp Hayo-Went-Ha and the Petty Property on 7/22 and 8/15 were excluded from the assessment and we lower our guidelines for N limitation because

particulate P was measured and particulate N was not in unfiltered water, N:P ratios in unfiltered groundwater indicate N-P colimitation or P limitation.

<u>*Benthic Algal Ecology of Torch Lake and Relationship to Nutrient Conditions*</u>

Extent of Golden Brown Algae. The spatial extent of GBA, the golden brown algae growing on top of sand, increased during the summer. GBA developed faster in shallow than deeper areas. The areal extent of GBA was not quantified well, but did not seem to be related simply to housing density along the shore. Other factors may be regulating groundwater paths and other conditions (sediment or water chemistry) that affect GBA development.

Benthic Algal Density. Although rocks and areas with dense golden brown algae covering the sand looked like they had relatively high amounts of algae, sparse sand habitats had as much or more algae in them (Figure 14). Cell densities of benthic algae in Torch Lake were as great in sandy areas that had little visual coating of golden brown algae as in areas with a dense visual coating of golden brown algae. This visual effect may be due to a change in location of algae from among sand grains below the interface of the sandy bottom and surface water to an algal-bacterial matrix accumulating on top of the sandy bottom. The GBA was dominated by diatoms with some cyanobacteria. More algae per unit area were observed on sand than rock. These results should be treated with caution because of the high variability of algal density in sand samples and lack of statistical significance in differences in cell densities among dense sand, sparse sand, and rock habitats.

Benthic algal density was not related to water depth of samples (Figure 15), an indicator of light availability and distance from shoreline contamination by septic wastes. Samples were collected from depths ranging from 2-6 feet, but cell density in them shows no relationship with depth.

Relating Patterns in Diatom Species Composition to Habitat, Cell Density, and Species N and P Preferences. Because we did not observe differences in nutrient conditions in groundwater or surface water between sites and dates, I did not try to relate changes in algal density or species composition to measured groundwater or surface water nutrient concentrations. However, I did use changes in diatom species composition and what is known about species nitrogen and phosphorus preferences to evaluate whether changes in species composition differed among habitat types and then if those changes were related to nutrient preferences of species.

There were over 100 species of diatoms in the 20 samples that MSU counted, in which we identified and counted at least 600 diatom valves. Diatoms are identified by characteristics of their cell walls. The glass cell wall of a diatom cell has two halves, called valves. The valves fit together like the two pieces of a Petri dish. Sometimes they come apart during sample preparation for counting, so we count valves instead of cells. 600 valves is equivalent to 300 cells, but we identify and observe more than 300 units because the valves of cells often come apart.

     Methods: Ordination methods. One of the first steps for identifying patterns in species composition in samples is a statistical procedure called ordination. Ordination allows us to map similarity in species composition in a two dimensional figure. Samples that have similar species composition are close together in this map, and vice versa for samples with the most different species composition. Then we can relate differences in species composition to environmental factors by relating environmental factors to the mapping space. The simplest way to think of this mapping is to

imagine a situation where we are only interested in two species in the samples. Each sample could then be located on the map if we use relative abundances of the two species (their proportions in multispecies assemblages) as the coordinate system or axes of the graph rather than north-south or east-west distance for cities on a map. Samples with high abundances of both species would be located in the northeast corner of the map. Samples with low abundances of both species would be located in the southwest corner of the map. Samples with low abundances of just one of the species would either be in the northwest or southeast corner of the map. In all cases, exact location of samples on the map are located based on specific relative abundances of the two species just as cities are located in geographic space based on latitude and longitude.

Of course, we are usually interested in more than two species. So ordination axes (or the map axes) are actually a combination of the relative abundances of multiple species with the species having the highest variability among samples also having the highest weight in defining the multispecies variables that form the ordination axes (labeled Dim for dimensions in Figure 16).

The map is rotated statistically so that the most different samples are oriented "east and west" or along the X-axis (Dim1, Figure 16) of the map. Then a second axis (Dim 2, Figure 16) is defined as different as possible (orthogonal) relative to the X-axis. Thus the axes (graphically referred to as dimensions 1 and 2, Figure 16) can be thought of as gradients in species composition based on weighted averages of species abundances. In this way ordination provides two-dimensional maps of samples in multidimensional species space.

I used an ordination method called non-metric multidimensional scaling (NMDS) to compare species composition among samples, and then relate differences in species composition among samples to habitat differences and diatom metrics of nutrient conditions. Ordination is usually used on data sets with 30 or more samples, so observations of patterns with the 17-20 samples that we had make it difficult to detect species-environment relationships.

Methods: Diatom metrics of nutrient conditions. Diatom metrics of relative nutrient concentrations were calculated by: 1) multiplying the species relative abundances (proportions of all diatoms) by species optimum TP or TN concentrations and 2) summing the products of species relative abundances and nutrient optima for each sample. This provides a weighted average metric of nutrient concentrations. Species TP and TN optima were determined using the diatom species composition of sediments and nutrient concentrations in approximately 1000 lakes that were sampled for the USEPA's 2007 National Lakes Assessment. When diatom metrics are calibrated for specific types of habitats and specific regions, they can be used to infer nutrient concentrations in streams, lakes, and wetlands rather than just relative concentrations as I have done for this report.

Because N and P concentration covary, problems exist for distinguishing a real causal relationship between diatom metrics of N conditions with N concentration versus the covarying P concentration; and vice versa for the diatom metrics of P conditions. Therefore, I combined these metrics into a diatom metric of general nutrient enrichment. In most lakes, P is the limiting factor, so this metric probably responds most to P enrichment.

In addition, I identified diatom species that can have endosymbiotic cyanobacteria in them, which allows these species to live in low N habitats, because the endosymbiotic cyanobacteria convert atmospheric nitrogen (N^2) that is dissolved in the water into ammonia. Ammonia is a bioavailable form of DIN for

algae. A diatom metric of N_2 fixers, i.e. nitrogen limitation, was calculated as the percent of diatoms in samples that can have endosymbiotic cyanobacteria.

Results: Diatom species composition of TLA030 sample was particularly different than others (Figure 16). TLA030 had a high score on Dimension 1 (Dim1, i.e. ordination axis 1) and low score on Dimension 2 (Dim2, i.e. ordination axis 2). Sample codes can be looked up in Table 2. TLA030 is one of two samples from Becky's Beach in Torch Lake, a site with sparse algae and gray color. This sample had high diatom density in the sand. Species of small diatoms that characteristically occur in sandy habitats were positively related to DIM1 and highly abundant in TLA030.

The diatom metric of NP enrichment was highly related to Dimension 2 of the NMDS plot, which was indicated by the direction of the blue arrow marked NP_Dia in Figure 16. The length of the arrow indicates the strength of correlation which changes in species composition. The shorter length of the arrow for the N_2 fixer metric indicates it was related less to variation in species composition among samples than the diatom metric for NP enrichment. The NP_Dia metric was related statistically and significantly (low probability of occurring by chance) to variation among samples, but the percent N2 fixers was not. Samples in the area of the NMDS plot indicated by the arrow have higher indications of nutrients in them than samples in the opposite direction of that arrow. Single samples from Lake Bellaire and Elk Lake (TLA034 and TLA044) scored highly on the second ordination axis, along with TLA024 and TL028. These samples were in line with the NP_Dia metric, suggesting that nutrients are higher in the habitats in which these samples were collected than in other habitats. In the opposite direction, species composition in TLA006, TLA029, and TLA030 indicated low nutrient concentrations because they were in the opposite direction of the NP_Dia arrow.

I removed the three "most" different samples from the ordination analysis so we could get more mapping (ordination-sample similarity/dissimilarity) detail for the remaining samples and perhaps distinguish differences between algae in sparse or dense habitats or in sand or rock habitats (Figure 17). In Figure 16, TLA030 from Becky's Beach sand and the samples from other lakes (TLA043 and TLA044) were arguably different than others, so they were removed from the ordination. You can see the changes in scales between Figures 16 & 17, decreasing from 3 to 2 as differences among all the samples in the respective analyses decreased. In addition, the axes were free to rotate for the second analysis without the three samples, so Figure 17 is not simply an enlargement of the "heart" of Figure 16.

Shifts in species composition were again related to the nutrient metrics (Figure 17), with NP_Dia indicating that habitats for TLA024 and TLA028 had highest nutrient concentrations and habitats for TLA006, TLA005, and TLA018 had lowest nutrient concentrations. The positive but statistically insignificant covariation between percent N_2 fixers and the NP_Dia metric indicates that N_2 fixers were increasing with nutrient enrichment, which is not what we would expect if nitrogen concentration was increasing. But it is possible if P concentration was increasing more than N relative to demand by algae, then N would become more limiting to algal growth and N_2 fixers would increase with NP_DIA, our indicator of nutrient enrichment. This hypothesis is even more likely if we add the factors that nutrient supply decreases to cells as benthic algal mats get thicker and N is lost from cells more rapidly than P. Algal density (Cells_cm2) was almost significantly related to changes in species composition, but not in a way that would be related to the diatom inferred nutrient gradient. Water depth was also not

statistically related to species composition, but ecologically it was consistent with expectations because it was oriented in the opposite direction of nutrient concentrations and algal density.

No difference in diatom species composition was observed between sparse sand, dense sand, and rock samples (Figure 18). Spider diagrams (Figure 18) were used to connect samples from sparse sand, dense sand, and rock. The ellipses delineate the species space in which we would expect sparse sand, dense sand, and rock to occur. The great overlap in the "webs" and ellipses for samples from each of these habitats indicates that little variation in diatom species composition at sites sampled in Torch Lake were related to these habitat characterizations.

Some trend, however, was observed when comparing diatom metrics indicating nutrient conditions across different habitat types (Figure 19), but these trends were not statistically significant. Therefore, diatom metrics showed no conclusive responses to the apparent density of algae on sand and whether samples were from sand or rock.

If groundwater nutrients were related to human sources, we'd expect decreasing concentrations with distance from shore. Our best indicator of distance from shore (which was not measured), is water depth. In the following analysis, I restricted the samples included in the analysis to sand samples from Torch Lake to control for between lake and between substrate (rock versus sand) differences. No relationship was observed between the diatom nutrient metric and distance from shore (Figure 20a).

The diatom metric for nutrient concentrations provides another method to assess nutrient conditions to which benthic algae were exposed. Thus, benthic algal cell density could be related to this indicator of nutrient concentrations. Again, I restricted the samples included in the analysis to sand samples from Torch Lake to control variability related to other factors. In this case, a very weak relationship was observed between benthic algal cell density and the benthic diatom indicator of nutrient concentrations (Figure 20b), but this relationship was not statistically significant.

Recent History of Torch Lake Water Temperature

Shengpan Lin, a graduate student at Michigan State University, used Landsat imagery and other sources of data to characterize temperature conditions in and around Torch Lake. Many satellites take images of earth with multiple sensors with each measuring a different wave length or band of electromagnetic reflectance. Most of the images that we see are composites of multiple bands, such as the "real color" image from Landsat 8 of the lower portion of Torch Lake composed of red, green and blue bands (Figure 21a). Landsat 8 images also measure surface water temperature with Band 11 (Figure 21b). The intensity of reflectance in Band 11 is used to measure and map variation in temperature by using a model to translate intensity into temperature, which is then represented by a color scale. In our case, the range in temperature on the map was from 16.6 to 21.7 °C, which is about 62 to 72 °F (Figure 21b-c). Landsat images taken over the last 3 years indicate that summer temperatures in Torch Lake reach their summer maximum in July and August, when the GBA was observed to grow and expand its coverage most (Figure 22).

Using Landsat 7, which has been in service longer than Landsat 8 and has a thermal band sensor like Landsat 8, surface water temperature was measured at two locations with different water depths at the southern end of the lake (Figure 23a), as well as with a lake-wide average using a set of cloud-free images that were downloaded from 1999-summer 2015 (Figure 23b). No long-term trends were

observed in summer water temperatures over the last 17 years when the GBA has become more apparent in Torch Lake. There is little evidence that temperatures varied more in the shallow regions of the lake than the deeper sections. Temperatures ranged from ice cover in the winter to about 294 °K, which is about 21 °C and 70 °F in the summer for the lake-wide average. The shallow regions of the lake were about 2° C warmer than the lake wide average. These results indicate that changes in lake temperature do not correspond with an increase in GBA during recent years in Torch Lake.

Review and Discussion

Groundwater phosphorus concentrations were approximately 5-10 times higher than surface water phosphorus concentrations. Groundwater nitrogen concentrations were about 20% lower than surface water phosphorus concentrations. This lead to a substantial shift in N:P ratios with those in groundwater lower than those in surface water. A much greater proportion of dissolved inorganic nitrogen (DIN) was ammonia in groundwater than in surface water. Without comparison to well water, contaminants unique to human waste, or gradient studies at different distances from septic tanks, differences in groundwater and surface water chemistry cannot be attributed to anything except to natural processes. The unusually high DIN concentrations near the Gourley location did indicate contamination from nearby sources. Future studies need to incorporate approaches to determine whether groundwater chemistry is related to human activities.

Phosphorus and nitrogen concentrations in both groundwater and surface water from Torch Lake were in ranges that limit growth of benthic algae in streams (Stevenson and Rier 2006). Because current reduces algal demand for nutrients by mixing surface water and nutrient poor water within algal mats, and currents are slower in lakes than streams, the limiting nutrient concentrations for benthic algae in lakes could be higher than for streams. So benthic algae in Torch Lake may be more limited by nutrients than estimated by the 86 and 306 µg DIN/L and 8 and 38 µg P/L. Any elevation in groundwater or surface water nutrient concentrations will increase benthic algal growth and accumulation because concentrations are below those that limit algal growth.

The relative high N:P ratios indicate that inorganic phosphorus is in more limiting supply than inorganic nitrogen. Because both nutrients are below levels that limit algal growth, increases in either nutrient should stimulate benthic algal growth. These results do not agree with the Lowe's (2015) finding with NDS experiments that nitrogen was the most limiting nutrient for benthic algal growth and phosphorus had no positive effect on benthic algal accumulation. The discrepancy in these results could be in the assumptions about levels of limiting nutrients from experimental streams and interpretation of nutrient ratios based on experiments with planktonic algae. Additional comparisons of results from experiments and surveys should be designed to reconcile cause-effect relationships between groundwater and surface water regulation of benthic algal growth.

Sand assemblages have remarkably high algal densities in them, even when they look relatively bare. Cell densities of algae on sand are usually between 100,000 and 1,000,000 cells/cm^2, unless waves or light limits algal accumulation (Stevenson and Stoermer 1981, Stevenson and Hashim 1989). The lack of variation in groundwater and surface water nutrient chemistry prevented testing the hypothesis that GBA development was related to enrichment of groundwater. Even differences between apparent dense and sparse areas of algae did not have different algal densities.

Benthic algae were dominated by diatoms, which is common in low nutrient waters. Species composition of the diatom assemblage was composed of both planktonic and benthic diatoms. The benthic diatoms included some diatoms in the genus *Epithemia* that can have endosymbiotic cyanobacteria in them that fix atmospheric nitrogen, which indicates that nitrogen was a limiting resource for benthic algae. Many of the diatoms in Torch Lake samples were characterized as low nutrient diatoms in the National Lakes Assessment.

The variation in species composition among samples was related to nutrient concentrations, as indicated by the diatom metric for nutrient concentrations. Therefore, variation in exposure of sampled diatoms to nutrients was indicated by the diatom metric. Species composition of diatom assemblages was not different between apparently densely and sparsely colonized sand, or between sand and rock. The diatom metric for nutrient concentrations did not differ with water depth, our indicator of distance from shore and septic tanks. And benthic algal densities were not related to our benthic diatom indicator of nutrient concentrations.

The lack of difference in species composition among habitat types may be related to characterization of areas with sparse and dense algae. Some sparse sand samples were taken in areas with dense GBA apparent in nearby habitats. It is possible that these sparse sand habitats developed into dense sand habitats as the summer progressed, so species composition in sparse habitats were just the early stages of succession for the species composition that would be found in dense habitats. As a result, water chemistry and resulting species composition were similar in some sparse and dense habitats. There were samples such as the one from Becky's Beach that was distinctly different. A wider range of sampling will help distinguish species composition of GBA and non-GBA assemblages.

The lack of difference in species composition among habitat types may be related to algal dispersal. Also, algae on and in surficial sands can occur in those habitats because they grew there, or they grew elsewhere and settled where sampled. Algae in the benthos disperse either actively by disassociating from the substratum and drifting, by sloughing when algal assemblages get dense and buoyant with oxygen bubbles, or by animals or waves disturbing and suspending them. These algae then settle in other areas where they can either grow, or die. Planktonic algae settle into the benthos and either grow slowly or die. In either case, the signal between species composition and its regulation by local environmental conditions can be masked by algal dispersal.

Diatoms are the most abundant and diverse algae in the Torch Lake benthos. Our methods for characterizing species composition of benthic diatoms did not distinguish between live and dead diatoms. We used a two-step process to characterize species composition of benthic algae that is a standard method for ecological assessments. First we characterized the species composition of non-diatom algae in a wet mount where we identified and counted cells of non-diatom algae and only distinguished and counted live and dead diatoms. Species of diatoms were identified in a separate preparation that removes all organic material samples from the glass cell walls of diatoms, which improves our ability to identify diatom species but prevents our ability to determine whether they were alive or dead when sampled. Once species composition of diatom assemblages of a habitat is pretty well known, it is possible to use different sample preparation methods for ecological studies, and these allow determining whether chloroplasts are inside the glass cell walls of diatoms, which we assume means they are alive. Distinguishing live and dead diatoms in future sample analyses s may help us understand the ecological differences between GBA and non-GBA assemblages and what controls GBA.

Changes in water temperature are not likely the reason for increased GBA in Torch Lake. GBA does develop during the warmest months of the year. But, satellite imagery indicates no change in surface water temperatures in Torch Lake, or shallow waters specifically, during the last 16 years when GBA has become apparent in Torch Lake.

<u>Conclusions</u>

We learned a lot about groundwater chemistry and benthic algal ecology in the first summer of research on Golden Brown Algae in Torch Lake. Groundwater and surface water chemistry differed. Benthic diatoms were related to changes in nutrient concentrations. But we were not able to relate benthic algal cell density and species composition to groundwater nutrient concentration due to a lack of differences in groundwater chemistry in the few areas sampled. The research conducted during the first summer provided data, a year of lessons learned about field methods, and a better understanding of how the data can be used to test hypotheses. All will help improve the research methods and approach for the future. There is still good reason to focus study on the hypothesis that groundwater enrichment by septic tanks is causing GBA. Better coordination of sampling water and algae, sampling a broad range of sites with expected differences in groundwater, and larger sample sizes with another year of data will increase chances of detecting relationships if they exist.

Recommendations for Future Research

The key questions remain the same.

Is GBA a recent development in the lake?
What regulates GBA development?
    Assuming that GBA is a recent development in the lake, what has changed in the lake? Answers
        to this question can provide hypotheses for what causes GBA.
    Does GBA develop with groundwater or surface water nutrient enrichment?
    Does GBA develop with changes in light or temperature, or other long term changes in lake?
Are human activities related to groundwater or surface water enrichment, groundwater flow paths, or other causes of GBA development?

The 2015 research approach was appropriate for an initial study, but needs to be revised based on what we learned and problems that we had.

We did learn that Torch Lake groundwater phosphorus concentrations are higher than surface water, but the opposite is true for N. Therefore, we should determine whether human activities cause enrichment of the groundwater. Comparisons of groundwater in drinking water wells which we assume are not contaminated with septic or agricultural nutrients and piezometer sampling along a gradient from septic tanks to the nearshore zone can determine whether human activities cause enrichment of the groundwater. In addition, measurement of human-specific contaminants such as caffeine can identify human contamination of groundwater.

The main issues with the 2015 results were:

1. not finding enough information about spatial variation in groundwater which prevented relating GBA to groundwater;

2. not distinguishing species composition of benthic algae between areas with visually apparent GBA and other areas;
3. not determining nutrient sensitivities and tolerances for benthic algae in Torch Lake with NDS experiments;
4. not determining what regulates GBA development in Torch Lake using experiments or surveys;
5. and not having enough resources to get everything done that we wanted to get done.

Regarding the last issue first. Not having enough resources to get everything done that we want to get done means we have to be strategic and patient. Being strategic means that all efforts should be targeted toward answering questions, whether they are existing questions or other questions that have enough priority that they warrant diverting resources and investigation.

We need to use either peepers or basters, or the Solinst temporary piezometer, to learn more about spatial variation in shallow and deep groundwater at different distances from the shore to relate variation in groundwater nutrients to GBA and human activities. Samples should be collected strategically by controlling as much as possible for natural sources of variation or expected levels of exposure to human activities (e.g. season (time), depth, distance from shore, land use on shore). In addition, a thermistor probe is recommended for measuring groundwater temperatures at depths up to 1 m.

More spatially extensive sampling, larger numbers of samples, and distinguishing live and dead diatoms in counts will help distinguish GBA from non-GBA algae, and thereby help determine causes of GBA. We learned that GBA is more spatially extensive than we thought, so we should expand the spatial range of sampling so sparse and dense GBA samples are not in the same location. Distinguishing live and dead diatoms in algal analyses from surveys and experiments will provide a refinement in characterizing species composition that could help distinguish GBA and non-GBA algae and their ecology. Ecological systems are naturally variable. Larger sample sizes help to account for that variability and resolve relationships among factors in ecological systems.

Classic nutrient diffusing substrate experiments as conducted last year will be important for determining whether benthic algae in those experiments respond most to N, P, or both N and P enrichment and which species are most regulated by N or P enrichment. Linking these results to surface and groundwater chemistry and relationships between GBA and water chemistry in natural habitats will be valuable first steps for establishing causal relationships for GBA development. Additional experiments will be necessary if NDS and survey results do not provide concordant lines of evidence for GBA regulation, but they will also be necessary to eliminate alternative hypotheses for GBA regulation. Experiments that independently and interactively manipulate surface and groundwater nutrient concentrations can determine the relative roles of nutrient supply from either habitat. Then experiments that manipulate nutrient concentrations at multiple levels could be used to determine the specific nutrient concentrations that regulate GBA development (see Figure 2), which will be valuable for refining cause-effect relationships and management strategies. Finally, it may not be nutrients alone that regulate GBA. So manipulations of temperature, light, water hardness, invertebrates or other factors may be necessary. These experiments will call for both in-lake and lakeside experimental approaches which are not technically difficult to conduct.

Acknowledgements

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Table 1. Chain of custody manifest for water samples. Epi stands for epicenter of the four-well configuration, two near shore and two farther out at about 4 ft depth, in a rough square. Ank stands for ankle depth lake water. Beach stands for groundwater seep at the beach, out of the water. Rock 1, 2, and 3 are groundwater samples collected where water motion was readily apparent by sight and touch; Rock tube was similar but used a tube to try and collect the groundwater. Weed 1, 2, and 3 are groundwater samples collected in weed beds where water motion was readily apparent by sight and touch. Drew spring 1, 2, and 3 are groundwater samples collected from a spring in the bed of the lake found by walking on it, in water about 8 - 12 inches deep just offshore and perhaps 6 - 8 feet north of the Beach sample at Hayo-Went-Ha (HWH). SW at Becky's Beach was collected at the same approximate depth as where the peepers were placed, a grab sample at arm's length depth. Peep 1, 2, and 3 are from about 3 inches deep in the floor of the lake at roughly 2, 4, and 6 feet from shore at the south end of Torch Lake. Solinst 1, 2, and 3 are groundwater samples collected at Gourley's, the only samples we obtained with the Solinst temporary peizometer.

Date	Site Code	Location	Well	SW	Filtered	TP/TN	SRP/N	Human	MSU	Priority	LakeSide	Distance
7/22/2015	L001AI	Hayo-Went-Ha	B		N	Y			1	1	E	13.9
7/22/2015	L001AI	Hayo-Went-Ha	B		N	Y			2	1	E	13.9
7/22/2015	L001AI	Hayo-Went-Ha	B		N	Y			3	1	E	13.9
7/22/2015	L001AI	Hayo-Went-Ha	B		Y		Y		4	1	E	13.9
7/22/2015	L001AI	Hayo-Went-Ha	B		Y		Y		5	1	E	13.9
7/22/2015	L001AI	Hayo-Went-Ha	B		Y		Y		6	1	E	13.9
7/22/2015	L001AI	Hayo-Went-Ha	D		N	Y			7	1	E	13.9
7/22/2015	L001AI	Hayo-Went-Ha	D		N	Y			9	1	E	13.9
7/22/2015	L001AI	Hayo-Went-Ha	D		Y		Y		8	1	E	13.9
7/22/2015	L001AI	Hayo-Went-Ha	D		Y		Y		10	1	E	13.9
7/22/2015	L001AI	Hayo-Went-Ha		Epi	N	Y			11	1	E	13.9

Date	Site Code	Location	Well	SW	Filtered	TP/TN	SRP/N	Human	MSU	Priority	LakeSide	Distance
7/22/2015	L001AI	Hayo-Went-Ha		Epi	N	Y			12	1	E	13.9
7/22/2015	L001AI	Hayo-Went-Ha		Epi	N	Y			13	1	E	13.9
7/22/2015	L001AI	Hayo-Went-Ha		Ank	N	Y			14	1	E	13.9
7/22/2015	L001AI	Hayo-Went-Ha		Ank	N	Y			15	1	E	13.9
7/22/2015	L001AI	Hayo-Went-Ha		Ank	N	Y			16	1	E	13.9
7/22/2015	L001AI	Hayo-Went-Ha		Beach	N	Y			17	0	E	13.9
8/18/2015	L001AI	Hayo-Went-Ha	Drew Spring 2		N	Y			55	0	E	13.9
8/18/2015	L001AI	Hayo-Went-Ha	Drew Spring 3		N	Y			56	0	E	13.9
8/18/2015	L001AI	Hayo-Went-Ha	Drew Spring1		N	Y			54	0	E	13.9
8/18/2015	L001AI	Hayo-Went-Ha		Beach	N	Y			57	0	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	A		N	Y			66	0	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	A		N	Y			67	0	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	A		N	Y			68	0	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	A		N			Y	72	1	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	A		Y		Y		69	1	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	A		Y		Y		70	1	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	A		Y		Y		71	1	E	13.9

Date	Site Code	Location	Well	SW	Filtered	TP/TN	SRP/N	Human	MSU	Priority	LakeSide	Distance
8/21/2015	L001AI	Hayo-Went-Ha	B		N	Y			73	1	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	B		N	Y			74	1	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	B		N	Y			75	1	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	B		N			Y	79	1	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	B		Y		Y		76	1	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	B		Y		Y		77	1	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	B		Y		Y		78	1	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	C		N	Y			80	1	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	C		N	Y			81	0	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	C		Y		Y		82	1	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	C		Y		Y		83	1	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	D		N	Y			84	0	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	D		N	Y			85	0	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	D		N	Y			86	0	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	D		N			Y	90	1	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	D		Y		Y		87	1	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	D		Y		Y		88	1	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha	D		Y		Y		89	1	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha		Epi	N	Y			91	1	E	13.9

Date	Site Code	Location	Well	SW	Filtered	TP/TN	SRP/N	Human	MSU	Priority	LakeSide	Distance
8/21/2015	L001AI	Hayo-Went-Ha		Ank	N	Y			93	0	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha		Epi	Y		Y		92	1	E	13.9
8/21/2015	L001AI	Hayo-Went-Ha		Ank	Y		Y		94	0	E	13.9
8/5/2015	L001AJ	2902 SETLD (Petty)	A		N	Y			18	0	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)	A		N	Y			19	0	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)	A		N	Y			20	0	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)	A		Y		Y		21	1	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)	A		Y		Y		22	1	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)	A		Y		Y		23	1	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)	B		N	Y			24	0	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)	B		N	Y			25	0	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)	B		Y		Y		26	1	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)	B		Y		Y		27	1	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)	B		Y		Y		28	1	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)	C		N	Y			29	0	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)	C		N	Y			30	0	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)	C		N	Y			31	0	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)	C		Y		Y		32	1	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)	C		Y		Y		33	1	E	10

Date	Site Code	Location	Well	SW	Filtered	TP/TN	SRP/N	Human	MSU	Priority	LakeSide	Distance
8/5/2015	L001AJ	2902 SETLD (Petty)	C		Y		Y		34	1	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)	D		N	Y			35	0	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)	D		N	Y			36	0	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)	D		N	Y			37	0	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)	D		Y		Y		38	1	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)	D		Y		Y		39	1	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)	D		Y		Y		40	1	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)		Epi	N	Y			41	1	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)		Epi	N	Y			42	1	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)		Epi	N	Y			43	1	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)		Ank	N	Y			44	1	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)		Ank	N	Y			45	0	E	10
8/5/2015	L001AJ	2902 SETLD (Petty)		Ank	N	Y			46	0	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	A		N	Y			95	0	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	A		N	Y			96	0	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	A		N	Y			97	0	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	A		Y		Y		98	1	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	A		Y		Y		99	1	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	A		Y		Y		100	1	E	10

Date	Site Code	Location	Well	SW	Filtered	TP/TN	SRP/N	Human	MSU	Priority	LakeSide	Distance
9/5/2015	L001AJ	2902 SETLD (Petty)	A		Y			Y	101	1	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	B		N	Y			102	1	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	B		N	Y			103	0	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	B		N	Y			104	0	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	B		Y		Y		105	1	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	B		Y		Y		106	1	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	B		Y		Y		107	1	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	B		Y			Y	108	1	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	C		N	Y			109	1	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	C		N	Y			110	0	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	C		N	Y			111	0	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	C		Y		Y		112	1	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	C		Y		Y		113	1	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	C		Y		Y		114	1	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	C		Y			Y	115	1	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	D		N	Y			116	1	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	D		N	Y			117	0	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	D		N	Y			118	0	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	D		Y		Y		119	1	E	10

Date	Site Code	Location	Well	SW	Filtered	TP/TN	SRP/N	Human	MSU	Priority	LakeSide	Distance
9/5/2015	L001AJ	2902 SETLD (Petty)	D		Y		Y		120	1	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	D		Y		Y		121	1	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)	D		Y			Y	122	1	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)		Epi	N	Y			123	1	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)		Ank	N	Y			125	0	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)		Epi	Y		Y		124	1	E	10
9/5/2015	L001AJ	2902 SETLD (Petty)		Ank	Y		Y		126	0	E	10
8/18/2015	L001AU	Cozy Point	Rock 1		N	Y			47	2	E	4.8
8/18/2015	L001AU	Cozy Point	Rock 2		N	Y			48	2	E	4.8
8/18/2015	L001AU	Cozy Point	Rock 3		N	Y			49	2	E	4.8
8/18/2015	L001AU	Cozy Point	Rock Tube		N	Y			50	2	E	4.8
8/18/2015	L001AU	Cozy Point	Weed 1		N	Y			51	2	E	4.8
8/18/2015	L001AU	Cozy Point	Weed 2		N	Y			52	2	E	4.8
8/18/2015	L001AU	Cozy Point	Weed 3		N	Y			53	2	E	4.8
8/19/2015	L001AW	6120 SETLD (Gourley)	A		N	Y			58	0	E	6.9
8/19/2015	L001AW	6120 SETLD (Gourley)	A		N	Y			59	0	E	6.9
8/19/2015	L001AW	6120 SETLD (Gourley)	A		N	Y			60	0	E	6.9
8/19/2015	L001AW	6120 SETLD (Gourley)	A		N			Y	64	1	E	6.9

Date	Site Code	Location	Well	SW	Filtered	TP/TN	SRP/N	Human	MSU	Priority	LakeSide	Distance
8/19/2015	L001AW	6120 SETLD (Gourley)	A		Y		Y		61	1	E	6.9
8/19/2015	L001AW	6120 SETLD (Gourley)	A		Y		Y		62	1	E	6.9
8/19/2015	L001AW	6120 SETLD (Gourley)	A		Y		Y		63	1	E	6.9
8/19/2015	L001AW	6120 SETLD (Gourley)		Epi	N	Y			65	1	E	6.9
9/5/2015	L001AW	6120 SETLD (Gourley)	Baster	pore	N	Y			207	1	E	6.9
9/5/2015	L001AW	6120 SETLD (Gourley)	Peep 1	benth		Y			201	1	E	6.9
9/5/2015	L001AW	6120 SETLD (Gourley)	Peep 1	sw		Y			202	1	E	6.9
9/5/2015	L001AW	6120 SETLD (Gourley)	Peep2	benth		Y			203	1	E	6.9
9/5/2015	L001AW	6120 SETLD (Gourley)	Peep2	sw		Y			204	1	E	6.9
9/5/2015	L001AW	6120 SETLD (Gourley)	Peep3	benth		Y			205	1	E	6.9
9/5/2015	L001AW	6120 SETLD (Gourley)	Peep3	sw		Y			206	1	E	6.9
9/5/2015	L001AW	6120 SETLD (Gourley)	Solinst		N	Y			218	1	E	6.9
9/5/2015	L001AW	6120 SETLD (Gourley)	Solinst		N	Y			219	1	E	6.9
9/5/2015	L001AW	6120 SETLD (Gourley)	Solinst		N	Y			220	1	E	6.9
9/5/2015	L001AY	Becky's Beach	Peep 1	benth deep		Y			208	1	W	17
9/5/2015	L001AY	Becky's Beach	Peep1	benth shallow		Y			209	1	W	17
9/5/2015	L001AY	Becky's Beach	Peep2	benth shallow		Y			210	1	W	17

Date	Site Code	Location	Well	SW	Filtered	TP/TN	SRP/N	Human	MSU	Priority	LakeSide	Distance
9/5/2015	L001AY	Becky's Beach	Peep3	benth deep		Y			211	1	W	17
9/5/2015	L001AY	Becky's Beach	Peep3	benth shallow		Y			212	1	W	17
9/5/2015	L001AY	Becky's Beach		sw	N	Y			213	1	W	17
9/9/2015	L001BF	6404 Crystal Beach Rd (Ozanne)	Peep 1	benth shallow		Y			214	2	W	0.7
9/9/2015	L001BF	6404 Crystal Beach Rd (Ozanne)	Peep 2	benth shallow		Y			215	2	W	0.7
9/9/2015	L001BF	6404 Crystal Beach Rd (Ozanne)	Peep 3	benth shallow		Y			216	2	W	0.7

Table 2. Chain of custody manifest for benthic algal samples.

MSU_ID	MSU_ID2	Habitat	Depth (ft)	Comments	Late Visual Assessments	Area Sampled (cm2)	Vol Left in Sample (mL)	U of M	MSU	Priority	LakeSide
TLA001	TLA-001	Sand	1.5 & 3	2 sand samples	Dense			Site 6		0	W
TLA002	TLA-002	Rock	2.5	Breakwater				Site 1		0	W
TLA003	TLA-003	Rock	2.5				69		4	1	E
TLA004	TLA-004	Rock	5			181	32		20	1	E
TLA005	TLA-005	Sand	6		Dense	66.18566	1		2	1	E
TLA006	TLA-006	Rock	6				80		3	1	E
TLA007	TLA-007	Rock	5			182	2		25	1	E
TLA008	TLA-008	Sand	5		Dense	66.18566	10		26	1	E
TLA009	TLA-009	Sand	1.5	2 rock samples, one sand	Dense			Site 2		1	E
TLA010	TLA-010	Sand	3	2 sand samples	Dense		62	Site 4		0	W
TLA011	TLA-011	Sand	5	Sparse growth	Sparse	66.18566	28		36	3	W
TLA012	TLA-012	Rock	4			124			21	0	E
TLA013	TLA-013	Sand	1.5		Dense		29	Site 3		0	E
TLA014	TLA-014	Sand	5		Dense	66.18566	15		18	3	E
TLA015	TLA-015	Sand	3	Sparse growth	Sparse	66.18566	53		19	3	E
TLA016	TLA-016	Sand	4.5		Dense	66.18566			1	1	W
TLA017	TLA-017	Rock	4	Pore water in & out of algae band			45	Site 5		0	W
TLA018	TLA-018	Sand	4.5		Dense	66.18566			34	1	W
TLA019	TLA-019	Sand	5		Dense	66.18566			12	0	E
TLA020	TLA-020	Rock	5			140			13	0	E
TLA021	TLA-021	Rock	3			100			30	0	W
TLA022	TLA-022	Sand	3		Dense	66.18566			31	0	W
TLA023	TLA-023	Rock	0.5	Darkish gray, not orange		124	63		32	0	W
TLA024	TLA-024	Sand	4	Dense growth	Dense	66.18566	23		5	1	E
TLA025	TLA-025	Sand	4	Sparse growth	Sparse	66.18566	65		6	1	E

MSU_ID	MSU_ID2	Habitat	Depth (ft)	Comments	Late Visual Assessments	Area Sampled (cm2)	Vol Left in Sample (mL)	U of M	MSU	Priority	LakeSide
TLA026	TLA-026	Sand	4	Dense growth	Dense	66.18566	22		23	1	E
TLA027	TLA-027	Sand	4	Sparse growth	Sparse	66.18566	36		24	1	E
TLA028	TLA-028	Rock	3			119	44		27	2	E
TLA029	TLA-029	Sand	3		Dense	66.18566	35		28	2	E
TLA030	TLA-030	Sand	5	Sparse growth	Sparse	66.18566			37	2	W
TLA031	TLA-031	Rock	5			176			35	2	W
TLA032	TLA-032	Rock	5			113			7	0	W
TLA033	TLA-033	Sand	5	Lots of pebbles in sand	Dense	66.18566			8	0	W
TLA034	TLA-034	Rock	5			107			9	0	W
TLA035	TLA-035	Sand	5	Dense growth	Dense	66.18566			10	0	W
TLA036	TLA-036	Sand	5	Sparse growth	Sparse	66.18566			11	0	W
TLA037	TLA-037	Sand	3.5	Sparse growth	Sparse	66.18566			14	0	W
TLA038	TLA-038	Rock	5			75			15	0	W
TLA039	TLA-039	Sand	5	Dense growth	Dense	66.18566			16	0	E
TLA040	TLA-040	Sand	5	Sparse growth		66.18566			17	0	E
TLA041	TLA-041	Rock	4.5			111			29	0	W
TLA042	TLA-042	Sand	4.5	Severe clay erosion, steep bank	Unknown	66.18566			33	0	W
TLA043	TLA-043	Sand	2		Sparse	66.18566	18		39	2	X
TLA044	TLA-044	Sand	2		Moderate	66.18566	59		38	2	X
TLA045	TLA-045	Rock	1.5	Breakwater				Site 1		0	X