Factors Contributing to Cyanobacteria Blooms in Upper Saint Croix Lake, WI

A Thesis
SUBMITTED TO THE FACULTY OF
UNIVERSITY OF MINNESOTA
BY

Thomas William Pevan

IN PARTIAL FULFILLMENT OF THE REQUIREMENTS
FOR THE DEGREE OF
MASTER OF SCIENCE

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December 2012

Acknowledgements

I would like to thank my advisors Stephanie Guildford and Robert Hecky, and my committee members, Erik Brown and Tom Hrabik for their assistance, time, and support. I am grateful for the assistance provided by Tom Mowbray for the cyanobacteria identification and counts and Hedy Kling for the algal identification. I am especially appreciative of the assistance of Jim Heim for providing his boat, lake level data, time, personal knowledge, and support.

This research was funded by the Vice-Chancellor Award and startup funds from the University of Minnesota – Duluth.

Dedication

This thesis is dedicated to my lovely wife Kathryn Mullenberg. Thank you for your love and patience while I completed this research.

Abstract

Upper Saint Croix Lake is a small, shallow, eutrophic lake that has experienced anthropogenic eutrophication. Nutrients, and in particular phosphorus availability, have long been recognized as a factor influencing the water quality of lakes. If phosphorus is excessive, cyanobacteria are often favored, leading to significant negative implications for the overall water quality and biodiversity of the lake, recreational enjoyment, and human and animal health. Nutrient concentrations, environmental factors, and phytoplankton efficiency were assessed to determine the role of seasonality and associated factors in the development of cyanobacteria blooms, and to examine the influence of year to year variability on the seasonal dynamics of the phytoplankton community. Light measurements, water temperature, and water samples were collected during the summers of 2008 (only year with a significant cyanobacteria bloom), 2009, and 2010, along with some winter sampling in 2010. Chemical analyses included chlorophyll *a*, particulate phosphorus, soluble reactive phosphorus, total and total dissolved phosphorus, total and total dissolved nitrogen, ammonia, nitrate, and soluble reactive silica in addition to active fluorometry (Phyto-PAM and Fluoroprobe). Results indicate that cyanobacteria in Upper Saint Croix Lake are controlled ultimately by phosphorus, and to a lesser degree, nitrogen and water temperature. Phosphorus concentrations, are in turn, controlled by environmental factors (precipitation, outflow, and inflow) that manipulate the dilution and flushing rate of the phosphorus present in the lake.

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Chapter 1: Eutrophication

What is eutrophication?

The term “eutrophication” was first coined by Weber (1907) to describe the nutrient conditions determining the vegetation of peat bogs. By the mid-1960s, the term eutrophication was used to describe changes within an aquatic ecosystem as well. As it became possible to directly measure the primary productivity of lakes and the role of human activities affecting the lake’s watershed, eutrophication became a term used to imply changes within the lake and land-water interactions (Schindler, 2006). Eutrophication is currently used to describe the biological reaction of aquatic systems to an increase in the inputs of nutrients, especially phosphorus and nitrogen, which encourages the growth of planktonic algae to nuisance proportions (Schindler, 1977). Depending on the degree of eutrophication, environmental effects can develop and degrade water quality (Vollenweider, 1976).

The process of eutrophication is a natural occurrence in lakes (Greeson, 1969). Eutrophication is an aspect of aging; it increases the rate at which lakes can fill with sediment and senesce. As lakes age, there is an accumulation of nutrients, sediments, silt, and organic matter in the lake from the surrounding watershed. Layers of sediments, silt, and organic matter produce successively shallower depths and can eventually reduce a water body to a marsh or bog. Once the process ends, the lake basin is completely filled and colonized by terrestrial vegetation (Schindler, 2006). The timing of natural eutrophication is highly variable and depends on the characteristics of the basin, watershed, and climate (Wetzel, 2001). However, humans have altered the rate and quantity of nutrient inputs for many lakes, increasing the pace at which eutrophication

would occur naturally in a process called cultural eutrophication (Rast and Thornton, 1996).

Causes of eutrophication:

The main cause of eutrophication is increased loading of phosphorus and nitrogen concentrations, which result in high algal biomass, often dominated by cyanobacteria (Marsden, 1989). Eutrophication can be accelerated as a result of human activity within the watershed. Humans have been altering the structure and function of the environment to suit their needs for thousands of years. However, since the industrial revolution, human activities and rapid population growth has lead to dramatic increases on the demands from both aquatic and terrestrial ecosystems. Approximately one-third to one-half of the land's surface has been transformed through land clearing, agriculture, forestry, and urbanization (Vitousek et al., 1997). Altered land use has led to changes in hydrological cycles, significant changes in population composition and abundance of biological communities, and unintentional and deliberate introduction of non-native species (Dobson et al., 1997, Matson et al., 1997). These human activities have also had profound impacts on the biogeochemical cycles of nitrogen and phosphorus (Vitousek et al., 1997). Excessive amounts of nitrogen and phosphorus are applied to croplands through fertilizers and animal manure with surplus nutrients migrating into surface waters and groundwater (Carpenter et al., 1998). Additionally, fluxes of nutrients can enter water bodies through point sources, such as municipal and industrial wastewater effluent, overflows of combined storm and sanitary sewers, runoff and infiltration from animal feedlots, and untreated sewage; and nonpoint sources, such as runoff from agriculture and

irrigation, urban runoff, septic tank leakage, and atmospheric deposition (Carpenter et al., 1998).

Substances other than inorganic phosphorus and nitrogen compounds can also contribute to eutrophication. Vitamins, growth hormones, amino acids, and trace elements can be a factor in eutrophication (National Academy of Sciences, 1969). Some of these substances are synthesized in the biological treatment of sewage.

Consequences of eutrophication:

Excessive nutrient inputs have various effects on the biology, chemistry, and human use of lakes and reservoirs. Effects include increased biomass of freshwater phytoplankton, shifts in phytoplankton species composition taxa to bloom-forming, frequently toxic, cyanobacteria, reduction in water clarity, elevated pH, dissolved oxygen depletion, increased probability of fish kills, and decreased aesthetic value (Smith et al., 1999). During the eutrophication process, the density of cyprinid fish generally increases. Because cyprinids are zooplanktivorous, the abundance and size of grazing zooplankton decreases, leading to changes in the structure of the lake ecosystem (Hrbáček et al., 1961; Jeppesen, 1997). The abundance and species composition of planktonic, bacterial, benthic and fish populations change as eutrophication progresses, and changes of this nature may be used detect and measure the degree and rate of eutrophication (National Academy of Sciences, 1969).

A common outcome of eutrophication is the mass occurrence and dominance of cyanobacteria in freshwater and estuarine ecosystems throughout the world. Cyanobacteria blooms can cause a variety of water quality problems, which include

aesthetic nuisances (e.g. odor, scum, unsightliness) dissolved oxygen depletion and subsequent fish kills, and unsafe drinking water (Paerl, 1988). Such problems caused by cyanobacteria blooms can severely impact aquatic habitat, recreational activities, and fisheries. Many species of bloom-forming cyanobacteria also produce a diverse range of toxins, which are released in the water following death and senescence of the bloom (Namikoshi and Rinehart, 1996) (Codd et al. 1999). These toxins include human and animal health hazards that can cause illness and mortality (Falconer, 1996) (Codd et al. 1999).

Nutrients and eutrophication:

The Experimental Lakes Area (ELA) is a permanent field station located in northwestern Ontario that uses a whole ecosystem approach, along with long-term and whole-lake investigations, to focus on cultural eutrophication. 46 small, deep, and pristine lakes were chosen due to being relatively unaffected by external human influences within the catchment. Beginning in 1969, phosphorus and nitrogen were added to one lake determine if would become eutrophic due to the nutrient inputs (Schindler, 2006). The lake quickly became eutrophic, despite having low concentrations of inorganic carbon (Schindler, 1971). Another lake was separated with heavy vinyl-covered nylon curtain to divide two basins. Both halves were fertilized with nitrogen and carbon, but only the north basin received phosphorus in addition to the other nutrients. The basin that did not receive phosphorus remained near-pristine, while the basin that received phosphorus rapidly developed algal blooms (Schindler, 1974). As a result of these two whole-lake studies, it was determined that, not only was phosphorus limiting algal growth

in many lakes, but low N:P ratios magnified eutrophication by favoring cyanobacteria that were capable of fixing atmospheric nitrogen (Smith, 1983).

Since the 1960s, it has been recognized that Lake Erie has been sensitive to accelerated eutrophication caused by elevated nutrient loading from human activities (Beeton, 1961). Lake Erie is the shallowest and warmest of the Great Lakes, and the basin has been intensively developed with agriculture, urban areas, industries, and sewage treatment plants. Decades of nutrient pollution lead to algal blooms covering large areas of the lake during the summer months, cyanobacteria causing taste and odor problems in some municipal water supplies, and decomposing algae depleting dissolved oxygen in some of the deeper areas of the lake (Mortimer, 1987). The problems caused by eutrophication became so severe that Lake Erie was declared dead in the 1960s. Joint collaboration between the United States and Canada led to decreased phosphorus loading into the Great Lakes (IJC, 1986). Despite reductions in nutrient loading and improved water quality, Lake Erie is still susceptible to cyanobacterial blooms during the summer (Ouellette et al., 2006). Lake Erie demonstrates the challenge of managing non-point source pollution in addressing cultural eutrophication.

Upper Saint Croix Lake Eutrophication

Upper Saint Croix Lake is a small and shallow drainage lake located in Douglas County, Wisconsin. Water quality monitoring of nutrient concentrations and the presence of cyanobacteria suggests that the lake is eutrophic during the later part of the summer. Water quality monitoring of the lake did not begin until 1995, but historical information and accounts suggest that the lake has gone through dramatic eutrophication in the past.

The village of Solon Springs surrounds most of Upper Saint Croix Lake. The first written record of a permanent settlement in the area that is now Solon Springs is in 1832. When a railroad was built in 1882, settlers and loggers built a logging camp at what is now the Main Street area off Solon Springs. Upper Saint Croix Lake was used as a giant holding pond for logs before they were floated down the Saint Croix River. Logging continued in the area for several decades. After World War II, much of the land around the lake was used to construct weekend/vacation homes. Plots of land were even given to new subscribers of the Duluth News Tribune newspaper (Solon Springs Historical Society). This led to rapid construction and soil erosion around the lake. Homes around the lake also relied on septic tanks, often placed near the shoreline, until the water treatment plant was built in the 1980s in response to concerns about the lake's health and aesthetics.

Citizens that have lived on the lake shoreline since the 1950s have given personal accounts on how the lake was in pristine condition in their youth, but degraded in condition until large algal blooms became a common occurrence. While the source for the blooms was unknown at the time, the blooms diminished in frequency after the construction of the water treatment plant and the removal of septic tanks in the 1980s. While blooms still occasionally appeared during the late summer, citizens did not take much notice, as it was an improvement over the large blooms that occurred previously. However, in late July of 2006, a large algal bloom formed over much of the lake that continued throughout much of the summer, and even forced the closure of the public beaches due to cyanobacteria toxin levels. Concerned citizens, worried that the lake might become as degraded as it was years earlier, began to look for answers.

While water quality monitoring did not begin until 1995, a sediment core was taken from the lake in 1997 for historical data. The core indicated that soil erosion increased slightly in accordance to the founding of the village and the completion of the railroad around 1880, but soil erosion started to increase dramatically around 1940 and continued to rise until around 1990 before stabilizing. The result of the soil erosion was an elevated sedimentation rate that increased nutrients entering the lake (Garrison, 2004). The core implies that Upper Saint Croix Lake has gone through cultural eutrophication due to human activity within the watershed. Recent water quality monitoring also suggests that Upper Saint Croix Lake is susceptible to relatively rapid changes in nutrient concentrations (Figure 1). Total phosphorus was the highest on record in 2006, which corresponds to the exceptional cyanobacteria blooms that also occurred that year (Figure 1).

Importance of Research:

Cultural eutrophication is a major threat to water bodies in many parts of the world, although recovery and nutrient loading has progressed in several regions. Eutrophication can lead to considerable changes in the structure of water bodies' ecosystem, which in turn reduces the possibilities of the use of the water as a source of drinking water, fishing, and recreational use. Of 215 lakes and reservoirs surveyed by the United Nations Environment Programme, 54% of Asia, 53% of Europe, 28% of Africa, 48% of North America, and 41% of South America have eutrophication problems (UNEP, 1994). Prevention and recovery of water quality is often difficult due to sources of nutrients from non-point areas and lack of sewage systems in developing countries.

Furthermore, climate change may increase eutrophication in many water bodies due to complex interactions with increased amounts of rainfall and snowfall, changes in water temperature, alterations of mixed layer depth, and changes in species composition to favor cyanobacteria (Dokulil et al., 2009). Eutrophication remains a challenge to manage because of the complex interactions of factors that can drive its occurrence in lakes, and different lakes may have different sensitivities to different factors.

Objectives of this Study

The objectives of the research on Upper Saint Croix Lake is to better understand the role of seasonality and associated driving factors (physical, chemical, and biological) to the development of the algal community, especially cyanobacteria through the season; to examine the influence of year to year variability on the seasonal dynamics of the phytoplankton community; and to use the results of the study to recommend possible early warning signs of cyanobacteria blooms and evaluate possible remedial actions.

Chapter 2: Cyanobacteria Blooms in Upper Saint Croix Lake

Introduction:

Upper Saint Croix Lake:

Upper Saint Croix Lake is a mesotrophic to eutrophic lake located in the Upper Saint Croix – Eau Claire River Watershed in Douglas and Bayfield Counties, Wisconsin (Figure 2). The lake is a 3,350,800m^2 (828-acre) natural drainage lake, with a maximum depth of 6.7m, at the headwaters of the St. Croix River (Figure 3) (Table 1). The littoral zone includes areas of the lake to a depth of 5m and comprises 84.5% of the lake (Turyk and Macholl, 2010). The average residence time of the lake is relatively short at 0.45 years (Turyk and Macholl, 2010). However, during low flow periods in later summer, when downstream vegetation growing in the riverbed may impound the outflowing river; the retention time in the lake increases. The lake receives water from groundwater (22%), seven tributaries (45%), surface runoff (18%), and direct precipitation (15%) (Table 2). The Upper Saint Croix Lake basin is of glacial origin, and was a spillway for glacial outwash about 11,000 years ago during the retreat of glaciers at the end of the last ice age (Manners et al. 2001). Local geography of the area consists of a thick covering of glacial drift in excess of 200 feet in some areas with underlying sandstone, basalt, conglomerate, shale, and metamorphic rocks (Manners et al., 2001).

Approximately 67% of the 15.29 km (9.5 miles) shoreline of Upper Saint Croix LakeSaint Croix is developed for seasonal and annual residential occupancy (DNR, 1997). The village of Solon Springs is located mostly on the western shore of the lake with a population of 576. The lake is used recreationally by residents and visitors and plays an important role in the economic life of the community. However, very little of the

developed shoreline has significant vegetative buffers that would provide habitat and reduce/filter runoff to the lake. During snowmelt or a rain event, water moves across the surface of the landscape towards lower elevations and the lake. The capacity of the landscape to hold water and filter particulates that are carried by runoff plays a role in determining the water quality, habitat, and amount of erosion into the water. The lack of vegetative buffers in the urban setting may lead to nonpoint phosphorus and nitrogen pollution into the lake, reducing water quality (Carpenter et al. 1998).

The average annual phosphorus budget for Upper Saint Croix Lake indicates that groundwater (645 kilograms per year) and internal release (829 kilograms pounds per year) are the major sources of phosphorus entering the lake (Table 3). The amount of phosphorus leaving the lake (506 kilograms per year) is much less than the amount of phosphorus entering the lake (1552 kilograms per year) (Turyk and Macholl, 2010). Of all the inflows, groundwater phosphorus concentrations are highest and range from 20 to 762µg/L with an average value of 107µg/L (Turyk and Macholl, 2008). The phosphorus retention time factor ($TP_{in} – TP_{out} / TP_{in}$) for Upper Saint Croix Lake is 0.67. The retention of phosphorus is lake specific, and lakes with a high flushing rate tend to have lower relative phosphorus retention than lakes with a slower flushing rate (Søndergaard et al., 2001). Despite Upper Saint Croix Lake's short residence time, the phosphorus retention time factor indicates that phosphorus is being retained within the lake, allowing internal phosphorus loading to have a major impact on the amount of phosphorus in the lake.

Recently, Upper Saint Croix Lake is showing signs of moderate nutrient enrichment and a shift towards dominance by cyanobacteria algae (Turyk and Macholl,

2010). Water quality monitoring and presence of significant cyanobacteria algae blooms suggest that Upper Saint Croix Lake is often eutrophic especially during late summer. The source of this eutrophication is thought to have originated in soil erosion in the watershed beginning around 1940 (Garrison, 2004). The result of the soil erosion is an elevated sedimentation rate that further increased the concentration of nutrients. Increased internal loading and continuing high groundwater phosphorus inputs have led to an imbalance in the amount of phosphorus in Upper Saint Croix Lake.

Cyanobacteria:

The eutrophication of temperate lakes often leads to changes in phytoplankton community structure and diversity with dominance by large, colony forming cyanobacteria (formerly referred to as blue-green algae), which can have detrimental effects on aquatic food webs (Paerl, 1988). Bloom formation of cyanobacteria generally has unfavorable effects on the domestic, industrial, and recreational uses of water bodies due to unpleasant taste and odor produced by surface scum. Certain species of cyanobacteria also produce toxins that can be potentially toxic to fish, livestock, and humans (Carmichael, 1986).

Cyanobacterial blooms are often composed of both toxic and nontoxic strains (Vezie et al., 2002). Environmental influences and nutrient concentrations can also effect the microcystin (cyanotoxin produced by cyanobacteria) production in different cyanobacteria strains (Sivonen, 1990, Vezie et al., 2002). Environmental conditions also influence microcystin concentrations directly by influencing cellular microcystin

production and indirectly by influencing cyanobacteria species composition (Orr and Jones, 1998).

Although increased input of nutrients is a main cause of the selective pressures that favor cyanobacteria, there are many conditions that promote cyanobacteria dominance. Cyanobacteria have higher temperature optima compared to other algal groups (McQueen and Lean, 1987), low light-energy requirements (Niklisch and Kohl, 1989) for many species, lower TN:TP ratios (Smith, 1983), the ability of some species to fix molecular nitrogen (Blomquist et al., 1994), migration from the sediments to the water column to gain a competitive advantage by storage of internal phosphorus reserves (Pettersson et al., 1993), the presence of gas-vesicles that allow movement to optimal light and nutrient conditions (Reynolds, 1987), and minimized mortality through grazing by zooplankton due to lower nutritional value, toxin production (Lindholm et al., 1989), and large colonies interfering with the filtering systems of zooplankton (Lampert, 1987). Rarely will a single factor be responsible for the bloom formation of cyanobacteria, but a combination of several factors, including hydrodynamic and meteorological effects, may influence their dominance (Spencer and King, 1989). Therefore, the occurrence of cyanobacteria blooms can be viewed as a problematic event that is dependent on numerous complex interactions within a system, but high nutrient concentrations, especially phosphorus, is a critical feature in their dominance in most lakes (Downing et al., 2001).

Objectives:

The objective of this chapter was to examine temporal and spatial variability in the phytoplankton community in Upper Saint Croix Lake. Temporal variability included seasonal and inter-annual changes in the composition of the phytoplankton community, with emphasis on cyanobacteria. Spatial variability included depth and area variations in algal abundance within the lake.

Methods:

Sampling on Upper Saint Croix Lake began in July, 2008, and ended in September, 2010. In 2008 and 2009, sampling was performed once a month throughout the summer, and in 2010, one sampling was completed in March through ice cover and regular sampling started just a few weeks after ice melt (April) at a frequency of approximately every two weeks. Four sites were originally chosen on Upper Saint Croix Lake to assess the spatial and depth development of the phytoplankton community and structure (Figure 3). Two deep-site locations, located farthest off-shore, were chosen because they corresponded to the same two sites that the Wisconsin DNR has used to examine the lake in previous studies (identified as DHN and DHS). Two shallow, near-shore sites were also chosen (identified as HDN and LWS). DHN and HDN are located on the northern end of the lake and DHS and LWS are located on the middle/southern end of the lake. Starting at the beginning of the 2009 sampling season, a fifth site was chosen (ISL). This fifth site is the furthest south site and was added to expand the spatial coverage of the study.

All measurements at Upper Saint Croix Lake were made between approximately 9:30am-1:00pm, with all samples processed or preserved the same day as collection. At

each site, in situ spectofluorometric (Fluoroprobe®) profiles (2009 and 2010 only) were taken by lowering the instrument slowly (<0.1m/s) through the water column to the sediments (slower deployment increases depth resolution of the instruments). Phytoplankton samples were collected at the surface using a 10µm net to determine species composition of larger taxa. Phytoplankton samples without concentration were also collected at the surface and deeper depths using a 4L van-Dorn. Water samples were typically collected at the surface (0-1m), mid-water column (2-3m), and near sediment (4-5m) for chemical analysis of nutrients. Unfiltered phytoplankton samples were preserved with Lugol's iodine, and samples that were collected for chemical analysis were preserved by freezing. Lugol preserved phytoplankton samples in 2008 were analyzed for cyanobacteria by Tom Mowbray by light microscopy, and one Lugol preserved sample was evaluated for total phytoplankton community species composition and biomass by phytoplankton taxonomist Hedy Kling (Algal Taxonomy and Ecology Incorporated, Winnipeg, Canada). Cyanobacteria counts performed by Tom Mowbray are given as units per milliliter, with a unit defined as a single filament or colony.

Chlorophyll *a* pigment concentrations were determined from material collected on 0.7µm glass-fiber filters that were extracted with 90% acetone and read on a fluorometer. Chlorophyll *a* fluoresces in the red wavelengths when excited by blue wavelengths of light, which is detected by a photomultiplier and compared to standards prepared from the blue-green algae *Anacystis,* which is free of chlorophyll b. The standards were calibrated on the spectrophometer to determine exact values using equations from Parsons and Strickland (1963).

Total algal biomass as chlorophyll and estimates of the contribution of the different algal groups were measured with a Fluoroprobe® (bbe Moldanke). The Fluoroprobe® is a highly sensitive fluorometric instrument for the in situ analysis of chlorophyll and assigning algal class determination based on characteristic pigments of different classes. The fluorescence of algae (emission at approximately 700nm wavelength) due to excitation by visible light depends on the presence of chlorophyll *a* and other pigments. The emission LEDs at 450nm, 525nm, 570nm, 590nm, 610nm cause fluorescence of chlorophyll *a*, based on the energy transfer from the different pigments, and result in an excitation spectrum characteristic for green algae, cyanobacteria, diatoms/dinoflagellates ("brown" algal groups), and cryptophytes taxonomic classes. This enables the analysis of the occurrence and distribution of algae on site with precision over depth and among lake stations without the necessity of the laboratory analysis. Possible interferences due to yellow substances are minimized by an integrated colored dissolved organic material (CDOM) correction factor. Results, however, are determined from factory calibrated broad-ranged algae cultures, not from lake-specific algae cultures. Linear regression of Fluoroprobe® estimates of total chlorophyll compared to extracted chlorophyll *a* was almost 1:1 ($y=0.965x+0.122$, $R^2=0.632$, $p(x)<0.001$, and $n=76$).

The Phyto-PAM® was used to estimate algal biomass and the photosynthetic characteristics of the different algal groups. The Phyto-PAM®, like the Fluoroprobe®, allows determination of active chlorophyll fluorescence in natural surface waters and differentiates between differently pigmented groups of algae (green algae, diatoms (browns), and cyanobacteria) using emission LEDs at 470nm, 520nm, 645nm, and 665nm. These wavelengths are fewer and different from those used by the Fluoroprobe®,

so less resolution is possible in defining algal groups and differences between the instruments on group assignment may be expected. The Phyto-PAM®, moreover, cannot provide profiles and can only operate in bench top mode with collected water samples. The Phyto-PAM® can, however, assess the photosynthetic performance and light-adaptation state of the various types of phytoplankton. The Phyto-PAM® was used to derive chlorophyll *a* concentrations when no extracted chlorophyll *a* data was available, by taking the linear regression of the Phyto-PAM® fluorescence units compared to extracted chlorophyll *a* samples ($y=0.102x+1.818$ and $R^2=0.510$ and $n=80$).

Both the Fluoroprobe® and Phyto-PAM® were used to assess the phytoplankton community structures and relative biomass. Instrument overlap occurring in 2009, but only the Phyto-PAM® was employed in 2008 and only the Fluoroprobe® was employed in 2010, with the exception of one sampling date in July with the Phyto-PAM® in 2010.

In 2008, Microcystin-LR concentrations were analyzed with EnviroLogix QualiTube kits for microcystin. The QualiTube Tube kit for microcystin is a competitive Enzyme-Linked ImmunoSorbent Assay (ELISA). The immunoassay for microcystin is based on the polyclonal and monoclonal antibodies against Microcystin-LR (Brooks and Codd, 1988). Microcystin toxin in the water samples collected throughout the summer of 2008 competed with enzyme (horseradish peroxidase)-labeled microcystin for a limited number of antibody binding sites on the inside surface of the test tubes. After a simple wash step, the outcome of the competition is visualized with a color development step with sample concentration inversely proportional to color development. The color development of the standards (0 ppb, 0.5 ppb, and 3.0 ppb microcystin) were read on the spectrophotometer at 450nm. Samples concentrations were determined from the linear

regression of the standards. This qualitative microcystin test has a limit of detection of 0.3 ppb.

Data was organized in an Excel spreadsheet (Microsoft Office 2007) and analyzed using the Systat statistical package (version 10). In cases describing correlations, a Pearson correlation was performed. Other statistical values were determined using a one-way ANOVA Tukey pairwise comparison. Error bars in the figures represent standard error.

Results:

Total chlorophyll *a* summer concentrations varied between the three sampled years (Figure 4). 2008 had significantly higher total chlorophyll during July-September (17.5 ± 13.5µg/L) than 2009 during the summer months (7.5 ± 1.3µg/L). There was no significant difference between 2010 and the other years during the summer months in total chlorophyll *a* (13.9 ± 5.9µg/L). During the month of July, there was no significant difference between 2008 (5.6 ± 1.1µg/L), 2009 (6.3 ± 0.7µg/L), and 2010 (7.1 ± 3.4µg/L). During the month of August, 2008 (20.5 ± 11.8µg/L) and 2010 (15.9 ± 6.1µg/L) had significantly higher chlorophyll than 2009 (8.4 ± 0.5µg/L). During the month of September, 2008 (17.6 ± 1.6µg/L) had significantly higher chlorophyll than 2009 (7.8 ± 1.5µg/L) and 2010 (9.6 ± 1.3µg/L). Furthermore, there was no significant difference between any of the stations within all three years, but chlorophyll *a* was significantly higher in the surface water than at 4m depth in all years.

The composition of the summer phytoplankton community varied slightly between the three summers. According to the Phyto-PAM® (Figure 5), all three years

began with similar low relative fluorescence values in the three phytoplankton groups (12.2 ± 5.8 fluorescent units for cyanobacteria, 14.5 ± 10.9 fluorescent units for green algae, and 16.8 ± 16.1 fluorescent units for diatoms). In 2008, green algae (74.4 ± 56.8 fluorescent units in August and 84.4 ± 18.9 fluorescent units in September) and cyanobacteria (101.1 ± 88.7 fluorescent units in August and 65.5 ± 8.4 fluorescent units in September) dominated, with low concentrations of diatoms present (7.4 ± 12.0 fluorescent units peak in August). In 2009, green algae were below detectable limits in August and September, cyanobacteria remained low (11.7 ± 1.6 fluorescent units in August and 22.5 ± 1.9 fluorescent units in September), and diatoms dominated (40.7 ± 7.4 fluorescent units in August and 36.7 ± 18.7 fluorescent units in September) (Figure 5). The Fluoroprobe®, however, detected green algae (3.9 ± 1.3µg/L in August and 2.7 ± 0.6µg/L in September) as the most abundant phytoplankton group in 2009, with diatoms (0.9 ± 0.5µg/L in August and 0.1 ± 0.1µg/L in September) as the lowest (Figure 6). In 2010, the Fluoroprobe® determined that green algae (4.5 ± 1.7µg/L in July, 6.1 ± 1.3µg/L in August, and 5.9 ±0.8µg/L in September) dominated, cyanobacteria concentrations were low in July (1.8 ± 1.7µg/L) and September (1.1 ± 1.1µg/L), but peaked in August (4.7 ± 3.2µg/L), and diatoms stayed low all summer (1.6 ± 1.8µg/L peak in July and decline to 0.6 ± 0.6µg/L in September) (Figure 6).

2008 had a significantly greater amount of cyanobacteria than 2009 or 2010 in August and September. Cyanobacteria blooms in 2008 did not develop until early August and lasted until late August/early September. 2008 and 2010 had the same trend of cyanobacteria concentrations increasing throughout July and August with a decline in September (Figure 7). However, a small increase in cyanobacteria did develop in late

September of 2009 when concentrations increased for the first time that summer (Figure 6).

The Lugol preserved algae sample collected from DHN 0.5m on July 23, 2008, which was identified by Hedy Kling (Table 4), indicated that 71.1% (biomass) of the algae present were cyanobacteria (Table 4) with over half being potentially toxic *Microcystis* (over $15x10^6$ cells/L). The high concentrations of cyanobacteria were in accordance with Phyto-PAM® measurements. Microscopy of cyanobacteria by Tom Mowbray throughout the summer of 2008 indicated that *Anabaena*, *Aphanizomenon*, *Coelosphaerium*, and *Microcystis* were the most common cyanobacteria, with *Anabaena* and *Microcystis* dominating (Figure 8).

The high concentrations of *Microcystis* led to higher levels of Microcystin-LR measured in August and September 2008 (Figure 8). On August 25, 2008, Microcystin-LR concentrations were at 1.8 ppb, which rose to 2.7 ppb by September 14, but then quickly declined to 1.2 ppb by September 25. All measured microcystin values in August and September of 2008 exceeded World Health Organization (WHO) drinking water standards (>1 ppb microcystin).

Discussion:

Chlorophyll *a*, which is related to algal biomass, naturally varies seasonally and from year-to-year in Upper Saint Croix Lake. The three sampling years exhibited similar trends with chlorophyll increasing throughout the summer with a peak in August before declining in September, with the exception of 2009 (no decline in September). However, the years varied considerably in maximal chlorophyll and cyanobacteria concentrations

achieved. 2008 and 2010 both had high August chlorophyll peaks, but only 2008 had high enough cyanobacteria concentrations to form a bloom (visible cyanobacteria biomass on the surface of the lake). 2009 had the lowest chlorophyll concentrations throughout the summer, with even lower cyanobacteria concentrations, but a small peak in cyanobacteria occurred in September. The small cyanobacteria rise in September, 2009, the time when cyanobacteria are beginning their decline in 2008 and 2010, is likely due to changes in conditions that favor cyanobacteria; similar conditions to 2008 (see Chapter 3).

In 2009, there is inconsistency between the Phyto-PAM® and Fluoroprobe® classification of the algal community. The Phyto-PAM® indicated that diatoms dominated the phytoplankton community, with low concentrations of cyanobacteria and green algae. However, the Fluoroprobe® indicated that green algae dominated, with low concentrations of cyanobacteria and diatoms. The discrepancy may arise from the fact that the two instruments use different emission wavelengths to detect the algae groups. Chlorophyll c, an accessory pigment found in diatoms, has an absorption maximum of 453nm (Falkowski and Raven, 2007). This absorption maximum is closer to the Fluoroprobe® emission LED wavelength than the Phyto-PAM®. However, the diatoms in Upper Saint Croix Lake have other accessory pigments that may alter the absorption spectra detected by the instruments. It is difficult to determine which instrument is correct because both the Fluoroprobe® and the Phyto-PAM® were factory-calibrated to common types of algal groups with broad ranges of absorption spectras. Based on the factory-set algorithms and absorption spectras, the phytoplankton samples in 2009 provided different results. Both instruments would need to be calibrated with Upper Saint Croix Lake

phytoplankton cultures to provide more accurate results due to differences in factory settings and actual lake present algae pigments. Most importantly, for the present study, the two instruments do agree that cyanobacteria were not abundant in 2009.

The results from the EnviroLogix QualiTube kits for microcystin indicate the cyanobacteria present in Upper Saint Croix Lake are producing microcystin (primarily from species of *Microcystis* and *Anabaena*). The presence of microcystin-producing *Microcystis* and *Anabaena* indicate that the cyanobacteria bloom in 2008 was high in toxicity. During the 2008 cyanobacteria bloom, which was dominated by *Microcystis* and *Anabaena*, microcystin concentrations were high. These results, however, may be skewed due to several different factors. The QualiTube kit for microcystin does not distinguish between the microcystin variants, but detects their presence to varying degrees. Microcystin-LR is the variant that is reported due to it being the most common variant and having the highest affinity to the immunoassay antibodies. Because not all microcystin variants are equally responsive and some nontoxic microcystin have a higher affinity to the immunoassay antibodies, an over- or under-estimate of total microcystin may occur. Furthermore, the microcystin ELISA is a quantitative field test that gives rapid results, but with lower sensitivity. To help define the actual toxicity risk, it is recommended in the future to perform ELISA in conjunction with Protein Phosphatase Inhibition Assay (PPIA), which measure direct effects of microcystin toxicity. However, for rapid and relatively inexpensive results, the ELISA for microcystin gives a reliable estimate for microcystin concentrations that correlated to the *Microcystis* and *Anabaena* concentrations in Upper Saint Croix Lake.

Conclusions:

The three sampling years presented different concentrations and community structures of the phytoplankton groups. 2008 and 2010 had a similar amount of total chlorophyll *a* during the summer, but 2008 had significantly more cyanobacteria. 2009 had lower concentrations of chlorophyll than the other two years although there was some increase in cyanobacteria in September when in other years cyanobacteria were in decline. Of the cyanobacteria that dominated the summer of 2008, *Anabaena* and *Microcystis* were the most abundant. The bloom of cyanobacteria began in mid-August and lasted, to a lesser degree, up to the last sampling date at the end of September. The high concentration of cyanobacteria led to visible scum formation, dense accumulation on the surface, and elevated toxicity. The occurrence of the cyanobacteria bloom in 2008, but not the other years, will allow the comparison of lake conditions between the years to identify factors that promoted the cyanobacteria bloom. To determine the factors that caused the dominance of cyanobacteria in Upper Saint Croix Lake during 2008, physical (water column stability, oxygen saturation, light availability, and meteorological conditions), chemical (nutrients), and biological (photosynthetic performance and light-adaptation state) controls were examined in Chapter 3.

Chapter 3: What Factors Control Cyanobacteria in Upper Saint Croix Lake

Objectives:

The objectives of this chapter were to determine the physical, chemical and biological factors influencing the composition and abundance of cyanobacteria in Upper Saint Croix Lake. The temporal and spatial variability of the physical, chemical and biological factors were also assessed to determine seasonal and inter-annual variations, along with depth and regional variations.

Introduction:

Cyanobacteria exhibit an increase in both biomass and relative contribution to total phytoplankton biomass as temperate lakes become more eutrophic (Pick and Lean, 1987). Although it is clear that the increased input of nutrients is the prime cause of selective pressure on phytoplankton, other factors in the system determine if cyanobacteria will dominate (Smith et al., 1987). Cyanobacterial blooms have been attributed to a variety of physical, chemical, and biological factors (Reynolds and Walsby, 1975; Paerl 1988).

The occurrence of high concentrations of cyanobacteria is closely linked with the eutrophication of lakes (Dokulil and Teubner, 2000). Associated with the dominance of cyanobacteria are negative effects, such as reduced transparency, decreased biodiversity, elevated primary production, oxygen depletion, which may result in fish kills, and toxin production (Reynolds, 1991). High concentrations of toxic cyanobacteria can also pose a potential health hazard with skin irritation and liver damage through chronic uptake of contaminated drinking water (Chorus, 1993).

Cyanobacteria are common to many eutrophic lakes, dispersed throughout the epilimnion. However, when high concentrations of cyanobacteria accumulate on the lake surface, dense blooms form. One of the factors that leads to the blooms of cyanobacteria is their ability to regulate their buoyancy, which allows them to move vertically through the water column to obtain optimal nutrient and light conditions (Oliver, 1994). Environmental conditions that regulate buoyancy in cyanobacterial blooms include temperature, light, nutrients, turbulence, and weather conditions (i.e. high light availability, low wind speed, and low precipitation) (Sorrano, 1997). Ultimately, for surface blooms to occur, turbulence caused by the wind must be low enough for upward migrating cyanobacteria to overcome the downward force of the wind-cased turbulence (Reynolds and Walsby, 1975). Thus, cyanobacteria surface blooms usually only form when there is low turbulence and cyanobacteria abundance is high.

Nutrients play a crucial role in the state of lakes due to the fact that open water primary production in summer is generally limited by nutrient availability. Increased nutrient inputs can lead to increased lake productivity, with effects on the upper trophic levels (Jeppesen et al., 1998). There is a large range of nutrient concentrations and variability between lakes and seasonally. Phosphorus originates primarily from soil minerals and will accumulate in various forms in lake sediments. Nitrogen, however, originates from the atmosphere and is closely tied to organic matter throughout its biogeochemical fractions. Nitrogen also tends to accumulate in lake sediments (Schindler et al., 1971). Before cultural eutrophication, most lakes were phosphorus limited. However, in part due to eutrophication, particularly the enrichment of phosphorus in lakes, a shift from clearwater state with submerged littoral macrophytes as often the most

significant primary producer to a turbid state with phytoplankton, especially cyanobacteria, have become more common, especially in shallow lakes (Jeppesen et al., 2005).

The increase in nutrient enrichment leads to an increase in phytoplankton biomass (Dillion and Rigler, 1974). However, an increase in a single nutrient may not be sufficient for cyanobacteria blooms to occur unless other nutrients are available in excess to the requirements of cyanobacteria. Cyanobateria need favorable gradients of nutrients and physical conditions to thrive. Nutrient gradients are expressed between the lake biota and its chemical-physical environment (Quiros, 2002). The occurrence of bloom cyanobacteria is often related to low TN:TP ratios (Smith and Bennett, 1999), as well as high total phosphorus (Downing et al., 2001).

Pulse amplitude modulated (PAM) fluorometry has become a common, non-invasive and rapid technique used to measure the variability of chlorophyll fluorescence and photosynthetic performance in phytoplankton (Baker, 2008) (Oxborough et al., 2000). The fluorescence measured with the PAM is emitted from the chlorophylls of Photosystem II reflecting the efficiency of photochemical energy conversion of Photosystem II reaction centers (Krause and Weis, 1991). Stressed phytoplankton cells can include stresses by nutrient limitation, high light intensity, low temperature, and high salinity concentrations for marine algae (Li et al., 2008). Effective quantum yield, which gives the efficiency of the fluorescence process, is used to measure the physiological state of phytoplankton, specifically nutrient limitation in dark adapted samples. Non-limited phytoplankton have effective quantum yield values from 0.6 to 0.7 with lower values in nutrient stressed cultures (Kromkamp and Peene, 1999). However, in natural

environmental settings, quantum yields can show wide variations among algal species with ranges from 0.4 to 0.8 for different phytoplankton classes even when nutrients are not limiting (Buechel and Wilhelm, 1993).

Methods*:*

Secchi depth and photosynthetically active radiation (PAR) readings were taken at each station between the hours of 10:00-12:00 every sampling day. Incident light at the lake surface was measured, as well as the attenuation of incident light down through the water column, was measured every 0.25m with a Li-Cor LI-192 underwater quantum sensor during the 2009 and 2010 sampling years. The LI-192 underwater quantum sensor has an absolute calibration of ±5% in air traceable to NBS, sensitivity of 4µA per 1000µmol s^{-1} m^{-2} in water, and a response time of 10 µs.

Water temperature and dissolved oxygen was measured with a Fisher Scientific accumet AP74 portable temperature/DO probe every 0.5m down to 4m in 2008. The AP74 DO probe has a relative accuracy of ±1.5% and a resolution of 0.01 mg/L with a two point calibration setting. The AP74 temperature probe has a relative accuracy of ±0.3°C and resolution of 0.1°C with automatic temperature compensation. In 2009 and 2010, water temperature and dissolved oxygen was measured with a SAIV SD204 CTD and binned into 0.5m intervals. The oxygen sensor in the SD204 CTD (SAIV205) has an accuracy of ±0.2mg/L and a resolution of 0.01mg/L. The temperature sensor in the SD204 CTD has an accuracy of ±0.01°C, resolution of 0.001°C, and response time of 0.2 seconds.

Relative thermal resistance to mixing (RTRM) is an index that quantifies the intensity of density differences due to the temperatures of adjacent water strata. The higher the RTRM, the greater the density difference, thus, the more difficult it is for wind mixing of adjoining layers to occur. RTRM was calculated as the density difference between surface and bottom divided by the density difference between water at 4° and 5°C (Birge, 1910). Generally, a strongly stratified lake will exhibit an RTRM >80. Due to the small size of Upper Saint Croix Lake, only one deep station (DHS) was characterized for RTRM.

Precipitation amounts were recorded by resident volunteer Jim Heim every day at 8:00am with a rain gauge placed on the shore of Upper Saint Croix Lake. At the same time every day, relative lake level was recorded with a meterstick that was placed in the lake after ice melt. The meterstick did not remain in the lake between the seasonal years, so the actual lake level could not be determined, only comparative changes in lake level from when the meterstick was positioned in the lake. Changes within a season of deployment were used to estimate changes in net water balance over the season.

At the same time physical measurements were made, chemical samples were collected for laboratory analysis of nutrients. Samples were collected by van-Dorn all three years from the surface water (0-1m) and mid-water column (2-3m), and additionally near-sediment (4-5m) in 2010. Samples were stored frozen and analyzed within 3 months of collection. Samples were analyzed for total phosphorus, total nitrogen, and soluble reactive silica for all three sampling years, and particulate phosphorus, soluble reactive phosphorus, total dissolved phosphorus, nitrate and nitrite, ammonia, and total dissolved nitrogen were analyzed for 2009 and 2010. Samples that were analyzed for dissolved

inorganic and organic nutrients were filtered through a 0.20µm polycarbonate filter prior to freezing. Particulate phosphorus was collected on a 0.45µm glass-fiber filter.

At the Large Lakes Observatory in the aquatic biology laboratory, soluble reactive phosphorus was determined using ammonium molybdate (Stainton et al., 1977) and total phosphorus, total dissolved phosphorus and particulate phosphorus were determined with the same method, but with an added procedure of digesting the samples in the presence of potassium persulfate (Ameel et al., 1993). Total nitrogen and total dissolved nitrogen were digested with the same method as total phosphorus and total phosphorus (oxidized using potassium persulfate) and then passed through a column containing granulated copper-cadmium to reduce nitrate to nitrite, diazotized with sulfanilamide and coupled with N-(1-napthyl)-ethylenediamine dihydrochloride (Zimmerman et al., 1991). Nitrate + nitrite was determined with the same method as total nitrogen and total dissolved nitrogen, but without the persulfate oxidization digestion. Ammonia was determined by fluorometric method in the presence of a single working reagent consisting of orthopthaldialdehyde, sodium sulfite, and sodium borate (Holmes et al., 1999). Silica was determined by reaction with molybdate and reduction with stannous chloride (Parsons et al., 1984).

Water collected by van-Dorn from the three water depths was filtered through 0.7µm GF/F for analysis of chlorophyll *a*. Filters were stored frozen and in the dark until particulate matter collected on the filters was extracted in acetone. After extraction, fluorescence was measured twice by fluorometer, before and after acidification to correct for phaeopigments (Parsons and Strickland, 1963).

The Phyto-PAM® (pulse-amplitude-modulation) was used on the same day as samples were collected in the lab to assess effective quantum yield, which is efficiency of the conversion of light energy to chemical energy process. Effective quantum yield is defined as the ratio of the number of photons emitted to the number of photons absorbed. Samples were dark-adapted for at least 20 minutes prior to measurement and were corrected for non-photosynthetic fluorescence.

Winter Samples:

During March of 2010, data and water samples were collected from DHN and DHS from holes drilled through the ice (approximately 0.75 m thick) from just under the ice, mid water-column, and near sediments. Ice typically covers Upper Saint Croix Lake from December through April. During this time, groundwater inputs become more dominant over surface water inputs.

In winter, ice cover on the lake prevents oxygenation while degradation of organic matter by biological activity depletes oxygen concentrations at the sediment-water interface, which can cause anoxia or near-anoxia in the hypolimnion. Similar to summer stratification, low oxygen concentrations can release phosphorus that is bound to sediment. Spring mixing then distributes the released phosphorus throughout the water column (Welch and Cooke, 1995). This internal release of phosphorus can delay or even prevent the recovery of lakes from cultural eutrophication if this internal release is a significant contribution to the phosphorus budget.

Results*:*

Water Transparency

The lake was more transparent in the summer of 2009 than in the summer of 2008 or 2010 (Figure 9) (Figure 10). Mean Secchi depth transparency in 2008 was 1.7 ± 0.1m in July, 1.2 ± 0.4m in August, and 1.5 ± 0.0m in September. Mean Secchi depth transparency in 2009 was 3.9 ± 0.6m in June, 3.0 ± 0.0m in July, 3.1 ± 1.1m in August, and 2.8 ± 0.2m in September. Mean Secchi depth transparency in 2010 was 2.6 ± 0.6m in June, 1.5 ± 0.2m in July, 1.2 ± 0.2m in August, and 1.3 ± 0.3m in September. The mean Secchi depth transparency during the summer months of 2009 (3.2 ± 0.7m) was nearly double that of 2008 (1.4 ± 0.3m) and 2010 (1.8 ± 0.7m). No significant difference was detected between the different sampling stations and Secchi depth. Mean light extinction in 2009 at Station 4 (DHN) was 0.6 in June, 0.5 in July, 0.7 in August, and 1.4 in September. Mean light extinction in 2010 at Station 4 (DHN) was 0.7 in June, 1.0 ± 0.3 in July, 1.7 ± 0.5 in August, and 1.5 in September. The mean light extinction during the summer months of 2009 (0.8 ± 0.4) was greater than that of 2010 (1.1 ± 0.5).

Temperature

The lake was significantly cooler in June and July of 2009 (19.4 ± 1.1°C and 20.4 ± 0.3°C respectively) than 2008 (23.6 ± 0.9°C in July) or 2010 (21.5 ± 1.1°C in June and 24.9 ± 0.5°C in July) (Figure 11). 2010 was significantly cooler in September (14.4 ± 0.2°C) than 2008 (17.4 ± 0.3°C) or 2009 (16.6 ± 0.2°C). However, there was no significant difference in water temperature across the three sampling years in August (22.5 ± 1.7°C in 2008, 22.9 ± 0.4°C in 2009, and 23.4 ± 1.2°C in 2010). No significant

difference was detected between surface water and bottom water or between stations for any sampling date.

Dissolved Oxygen

There was no significant difference in dissolved oxygen (DO) in the lake across the three years (Figure 12). Whole-lake DO concentrations ranged from 8.23 ± 0.63mg/L (2008), 8.51 ± 0.65mg/L (2009), to 7.87 ± 1.12mg/L (2010). There was also no significant difference between surface-water and near-sediment DO concentration in 2008 or 2009. However, during the summer months of 2010, surface-water was 8.75 ± 0.52mg/L (0-1m) and near-sediment water (>4m) was 6.71 ± 1.21mg/L.

Water Column Stability

The summer mean relative thermal resistance to mixing (RTRM) at DHS did not vary significantly between the three years and remained very low, with the highest RTRM occurring in June of 2009 (1.792 ± 0.691), which was nearly triple that of 2010 (0.24 ± 0.31) (Figure 13). RTRM ranged from 0.24 ± 0.73 in 2008, 0.53 ± 0.83 in 2009 and 0.53 ± 0.52 in 2010.

Precipitation

The three sampling years were different in the amount of rainfall that the lake received. Average precipitation that enters Upper Saint Croix Lake from June 1st to September 30th is 40.7 cm. 2009 was the driest year (22.9 cm of precipitation), 2010 was the wettest year (60.5 cm of precipitation) and 2008 received near normal precipitation

(46.0 cm) (Figure 14). 2010 also had the greatest number of large rain events (>2 cm) with 12, 2009 had the fewest large rain events with 6, and 2008 had 9 large rain events.

Lake level initially declined in 2008, but then returned to initial lake level by June before remaining fairly stable throughout the summer months with a slight peak in September (0.17m). Lake level in 2009 slowly increased throughout the summer months before peaking in September (0.39m). Lake level in 2010 raised continuously until it peaked on August 20th (1.14m) and then rapidly declined (0.79m on September 1st) (Figure 15).

Phosphorus

Mean total phosphorus concentrations were consistently higher in 2008 (50.4 ± 12.8µg/L) during the summer months than those in 2010 (21.5 ± 6.6µg/L). There was no significant difference in 2009 (33.9 ± 9.5µg/L) during the summer months compared with either 2008 or 2010. However, the mean monthly averages varied between each year (Figure 16). In 2008, total phosphorus concentrations remained at a high concentration during the summer months with a slight rise in August (43.7 ± 11.5µg/L in July, 54.9 ± 12.1µg/L in August, and 47.6 ± 12.8µg/L in September). In 2009, total phosphorus concentration began high (38.4 ± 5.7µg/L in June and 40.3 ± 10.4µg/L), but decreased in August (22.3 ± 1.8µg/L) before rising again in September (36.1 ± 6.2µg/L). 2010 total phosphorus concentrations started the summer months with the lowest concentration and slowly increased throughout the summer (15.2 ± 2.6µg/L in June, 19.7 ± 3.1µg/L in July, 25.9 ± 3.4µg/L in August, and 31.4 ± 3.8µg/L in September). Comparing total phosphorus concentrations in just the month of August, 2008 (54.9 ± 12.1µg/L) was

significantly higher than 2009 (22.3 ± 1.8µg/L) and 2010 (25.9 ± 3.4µg/L). All three years ended with similar total phosphorus concentrations by the end of September (47.6 ± 12.8µg/L in 2008, 36.1 ± 6.2µg/L in 2009, and 31.4 ± 3.8µg/L in 2010). Total phosphorus was low during the winter sample collected in March, 2010 (14.6 ± 2.8µg/L). No significant difference was detected between any stations or between surface-water or near-sediment water.

Mean particulate phosphorus concentrations was similar in both 2009 (9.0 ± 1.0µg/L) and 2010 (8.4 ± 2.6µg/L). Both years also had similar trends with the lowest concentrations during the summer months occurring in June (7.8 ± 0.5µg/L in 2009 and 7.0 ± 1.7µg/L in 2010) and increasing to the highest concentration in September (10.1 ± 0.5µg/L in 2009 and 12.6 ± 1.0µg/L in 2010) (Figure 17). Particulate phosphorus was lowest during the winter sample of March, 2010 (2.8 ± 0.6µg/L). No significant difference was detected between any stations or between surface-water or near-sediment water.

Mean soluble reactive phosphorus concentrations remained low during the summer months of 2009 (3.2 ± 0.8µg/L) and 2010 (4.5 ± 1.6µg/L) (Figure 18). However, the monthly mean averages from 2009 were lower than 2010 in July (2.8 ± 0.2µg/L in 2009 and 4.9 ± 1.0µg/L in 2010), August (3.0 ± 0.7µg/L in 2009 and 4.0 ± 0.7µg/L in 2010), and September (2.9 ± 0.1µg/L in 2009 and 6.8 ± 1.3µg/L in 2010). Soluble reactive phosphorus was high during the winter sample of March, 2010 (6.4 ± 1.9µg/L). No significant difference was detected between any stations or between surface-water or near-sediment water.

Mean total dissolved phosphorus concentrations were much higher and variable in 2009 (25.4 ± 11.2µg/L) than 2010 (8.2 ± 3.2µg/L). Total dissolved phosphorus remained low in 2010 and varied little during the summer months ranging from lowest concentration in June (7.3 ± 3.5µg/L) to the highest concentration in September (10.7 ± 3.1µg/L). Total dissolved phosphorus varied greatly in 2009 with high concentration in June (34.6 ± 8.0µg/L) and July (29.8 ± 19.6µg/L) to lower concentration in September (15.5 ± 2.8µg/L) (Figure 19). No significant difference was detected between stations or depth, except in 2010, with total dissolved phosphorus concentrations being greater at 2m than surface-water.

Nitrogen

Monthly mean total nitrogen increased in all three years during the summer months, but only in 2009 did total nitrogen concentration decrease in September (Figure 20). Total nitrogen was lowest in 2008 (616.6 ± 276.3µg/L) and 2010 (577.7 ± 165.2µg/L) during the summer months and highest in 2009 (800.5 ± 220.4µg/L). No significant difference was detected between any stations or between surface-water or near-sediment water.

Nitrate + nitrite concentrations were below detectable levels (0.1µg/L) during all summer sampling months of 2009 and 2010, except for the month of September in 2010 (57.6 ± 13.9µg/L) (Figure 21). High nitrate + nitrite levels were also detected in March 2010 (120.7 ± 48.9µg/L). No significant difference was detected between any stations or between surface-water or near-sediment water.

Mean averages of ammonia were similar in 2009 (12.5 ± 7.5µg/L) and 2010 (11.6 ± 10.9µg/L). 2009 and 2010 had slight increases in ammonia concentrations during the summer months with concentrations lowest in June (6.4 ± 1.6µg/L in 2009 and 9.0 ± 7.9µg/L in 2010) and rising to the highest concentration in September (16.0 ± 6.3µg/L in 2009 and 17.7 ± 5.3µg/L in 2010) (Figure 22). No significant difference was detected between any station in either 2009 or 2010 and there was no difference between concentrations and depth in 2009. However, in 2010, ammonia concentrations were significantly higher at near-sediment water than either mid-water column or surface-water.

Mean total dissolved nitrogen was higher during the summer months of 2010 (504.7 ± 103.1µg/L) than 2009 (346.8 ± 44.8µg/L). Total dissolved nitrogen had a similar trend in both 2009 and 2010 with concentrations lowest in June (293.6 ± 11.4µg/L in 2009 and 394.4 ± 37.5µg/L in 2010) and rising to the highest concentrations in September (389.7 ± 11.4µg/L in 2009 and 624.5 ± 21.1µg/L in 2010) (Figure 23). Total dissolved nitrogen was high during March, 2010 (450.1 ± 46.5µg/L), but dramatically decreased in April (262.1 ± 12.7µg/L). No significant difference was detected between any stations or between surface-water or near-sediment water.

Silica

Mean soluble reactive silica (SiO2) concentrations were highest in 2009 (5970 ± 476µg/L) and lowest in 2008 (4430 ± 641µg/L) during the summer months. All three sampling years had similar trends of soluble reactive silica rising throughout the summer months to a peak in September (Figure 24). Soluble reactive silica concentrations

remained high across all years with lowest concentrations occurring in June or July (4080 ± 718µg/L in 2008, 5080 ± 91µg/L in 2009, and 4680 ± 114µg/L in 2010) and highest concentrations occurring in September (5100 ± 158µg/L in 2008, 6330 ± 104µg/L in 2009, and 5810 ± 112µg/L in 2010). Soluble reactive silica was high in March, 2010 (5990 ± 278µg/L), but dramatically decreased in April (4900 ± 50.9µg/L). No significant difference was detected between any stations or between surface-water or near-sediment water.

Nutrient Ratios

Total nitrogen to total phosphorus molar ratios (TN:TP) were lowest in 2008 with a summer mean of 25.9 ± 11.9, and TN:TP were highest in 2010 with a summer mean of 71.7 ± 13.1. TN:TP increased each month of 2008, but remained relatively low (16.4 ± 6.8 in July, 24.9 ± 10.1 in August, and 39.0 ± 8.8 in September). TN:TP in 2009 increased from June (39.4 ± 22.9) to August (103.1 ± 8.0) before dropping dramatically in September (31.4 ± 7.3). In 2010, TN:TP started high in March (69.3 ± 12.7), dropped in April (56.2 ± 7.0), peaked in June (80.6 ± 13.1), and then declined slightly every month through September (55.9 ± 6.5) (Figure 25). No significant difference was detected between any stations or between surface-water or near-sediment water.

Chlorophyll a

Mean chlorophyll *a* concentrations were highest in 2008 (16.0 ± 11.7µg/L) and lowest in 2009 (6.9 ± 1.7µg/L) during the summer sampling months. Chlorophyll *a* concentrations increased throughout 2008 with the lowest concentration in July (5.6 ±

1.1µg/L) and peaked in August (20.5 ± 13.8µg/L). 2009 and 2010 had the same trend as 2008 with increasing chlorophyll *a* concentrations with lowest concentration in June (4.3 ± 0.6µg/L in 2009 and 5.8 ± 2.3µg/L in 2010), peaks in August (8.4 ± 1.5µg/L in 2009 and 15.9 ± 6.1µg/L in 2010), and a decline in September (7.8 ± 1.5µg/L in 2009 and 9.6 ± 1.3µg/L in 2010) (Figure 4). No significant spatial variability was detected between any stations or depth in 2008 or 2009, but chlorophyll *a* was significantly higher in the surface-water than near-sediment water in 2010.

Effective Quantum Yield

2008 and 2009 had similar trends in effective quantum yield (Fv/Fm) in that the yields were lowest in July (0.45 ± 0.08 in 2008 and 0.44 ± 0.07 in 2009), reached a near peak in August (0.53 ± 0.07 in 2008 and 0.57 ± 0.02 in 2009) and remained relatively stable through September (0.56 ± 0.07 in 2008 and 0.55 ± 0.01 in 2009). 2010, however, peaked in June (0.051 ± 0.06) and decreased in July (0.45 ± 0.09) and remained low in August (0.46 ± 0.03) (Figure 26). No significant difference was detected between any stations or depth in any sampling year.

Discussion*:*

Cyanobacteria are able to move vertically through the water column by regulating their buoyancy through intra-cellular vacuoles (Walsby, 1969). This mechanism gives cyanobacteria the advantage of positioning themselves at optimal light and nutrient availability depths within a stable water column, especially in small lakes where wind stress is low (Reynolds et al., 1987). The cyanobacterial blooms in 2008 were associated

with decreased light availability and transparency (low Secchi depths). Light availability and transparency in 2009 was double that of 2008 and 2010, and had the lowest concentration of cyanobacteria. Low light levels, however, may not contribute to the development of cyanobacteria blooms, but may be a result of the bloom or higher total phytoplankton biomass (Shapiro, 1997). This was observed in 2010, when water transparency was similar to the 2008, but no cyanobacteria bloom developed. The similar water transparency in 2008 and 2010 responded to total chlorophyll, indicating that low light alone does not result in a cyanobacteria bloom. Furthermore, light availability and penetration in Upper Saint Croix Lake is naturally low due to the high amounts of humic substances that give the lake a brown color.

Cyanobacteria dominance generally occurs at water temperatures >20°C (McQueen and Lean, 1987). Water temperature in August exceeded 20°C in all three years, but temperatures in 2008 and 2010 exceeded 21°C more often than 2009. The water temperature also exceeded 20°C sooner in both 2008 and 2010 (June) than 2009 (late July). However, 2010, which had no cyanobacteria bloom, was 1°C warmer in both July and August than 2008. This suggests temperature is likely not the primary cause for phytoplankton succession, but rather a single enabling factor (or an associated factor that reflects the meteorological year differences) in combination of other factors (Jacoby et al., 2000). Warmer water temperatures often indicate water column stability due to density differences between surface water and deeper water. However, no stable water conditions were observed during any year, likely due to the very shallow nature of Upper Saint Croix Lake (84.5% of the lake is <3m). Cyanobacterial blooms developed in 2008 despite unstable water column conditions (RTRM).

Low dissolved oxygen can be an indicator of high cyanobacteria concentrations with decomposition depleting oxygen. Throughout the water column, dissolved oxygen remained relatively high (>7mg/L) across all three years, even during the cyanobacteria bloom in 2008. Dissolved oxygen concentrations may have been elevated during the cyanobacteria bloom due to photosynthetic reactions. The oxygen levels near the sediment could not be measured in 2008 due to the DO probe only being able to reach a maximum depth of 4m (anoxic conditions may have been present at the deep sites near the sediments). The shallow nature of the lake likely allows for mixing between the surface-water and near-sediment water allowing high dissolved oxygen concentrations. However, near anoxic conditions (<1mg/L) did occur in August of 2010 near the sediments even though cyanobacteria concentrations remained low. The brief period of anoxic conditions could be the result of the decomposition of rich organic material that composes the lake sediment, warm temperatures, and low mixing on that date. Anoxia did not occur during March of 2010, but ice cover and biological degradation might cause anoxia during some winters.

Precipitation likely affected nutrient influxes, water column stability, and lake level. 2008 had near average precipitation, which led to stable lake levels. 2009 had nearly half of average precipitation, but the lake level continued to rise slowly throughout the summer, probably due to dense aquatic vegetation near the outflow. 2010 had 20cm over the average amount of precipitation, which caused lake levels to raise over 1m and caused flood conditions around the lake. Upper Saint Croix Lake is a drainage lake with a single shallow outlet channel that fills with aquatic vegetation (mostly wild rice) (Turyk and Macholl, 2010) during the summer months. The aquatic vegetation assists in

maintaining stable lake levels as was observed in 2008 and 2009. However, the large amounts of precipitation in 2010 caused the lake level to rise above the aquatic vegetation. When the excessive precipitation stopped, a rapid decline in lake level was observed, which greatly increased the flushing rate and decreased the water retention time for the summer of 2010. The increased flushing rate may explain why 2010 did not experience a cyanobacteria bloom like 2008, despite having similar water temperatures, light availability and transparency.

Phosphorus limits algal biomass in 80% of Wisconsin's lakes (Shaw et. al, 2002); as is the case with Upper Saint Croix Lake based on elevated total nitrogen concentrations, high nitrogen to phosphorus molar ratios, and phosphorus to chlorophyll *a* correlation (Figure 27). Enrichment with phosphorus is usually a precursor to cyanobacterial bloom formation (Paerl, 1988). In general, the potential for cyanobacteria blooms rises rapidly as total phosphorus increases from 30 to 100µg/L (Downing et al., 2001). Higher total phosphorus (and low TN:TP) concentration in Upper Saint Croix Lake during the summer of 2008 may have favored the cyanobacterial bloom that year. Furthermore, cyanobacteria levels stayed minimal in 2010 when total phosphorus low (below 30µg/L for most of the summer).

Although phosphorus is often the limiting nutrient for algal biomass, nitrogen can also play an important role in algal growth when low TN:TP ratios occur (Jin and Hongjuan, 2010). This was demonstrated in 2009, when during the early summer, phosphorus concentrations were as high as 2008, but nutrient ratios did not favor cyanobacteria. Bloom-forming cyanobacteria tend to dominate lakes when the TN:TP molar ratio was less than 29 due to nitrogen limitation (Smith, 1983). Furthermore, low

TN:TP ratios promote the growth of nitrogen-fixing cyanobacteria (Peter and Kaji, 2006), such as *Anabaena*, which dominated Upper Saint Croix Lake in August, 2008. TN:TP remained high in 2010 and varied greatly in 2009, but TN:TP ratios only decreased below 29 during the summer of 2008; the only year to have a cyanobacteria bloom.

The variable silica concentrations between the years likely did not influence the phytoplankton community. Silica is necessary for diatom growth, but the lowest observed concentrations of silica in Upper Saint Croix Lake were still very high (>4000µg/L). The high concentrations of silica are likely due to the shallow nature of the lake, with wind-driven mixing silica-rich sediment. Diatom abundance usually remained relatively low in Upper Saint Croix Lake, probably due to green algae and cyanobacteria outcompeting the diatoms for nitrogen (likely nitrate and ammonia).

2008 was a unique year, not only for the cyanobacteria blooms, but also for the stable lake water levels. In contrast, 2009 and 2010 had rising water levels through the summer, implying that water inputs (most likely surface runoff) were exceeding outflow. The net result is increased flushing and dilution of groundwater inputs, which are rich in phosphorus, in 2009 and 2010, compared to 2008.

While winter samples were only collected once, nutrient concentrations did not vary much from the spring samples, with the exception of soluble reactive phosphorus, nitrate + nitrite, total dissolved nitrogen, and soluble reactive silica. The high concentration of soluble reactive silica is likely a carryover from the high concentrations observed during the fall of 2009, with the rapid decrease in April due to the spring diatom bloom. Higher soluble reactive phosphorus values during the winter could be a result of internal phosphorus release, however, since oxygen values were not near anoxia,

phosphorus release from the sediment may not be the source of the phosphorus in winter of 2010. The source of the increase in phosphorus during the winter may be a result of groundwater inputs, which are naturally high in soluble reactive phosphorus and have a greater influence than surface water inputs during the winter. The source of the nitrate + nitrite may also be from the groundwater. Inorganic nitrogen is very soluble and does not bind to soils, which allows them to readily leach into groundwater. The sandy soil in the Upper Saint Croix Lake allows inorganic nitrogen to leach into the groundwater with ease. The high concentration of total dissolved nitrogen is likely due to the elevated nitrate + nitrite concentrations observed in March 2010.

Total chlorophyll *a* indicates that the phytoplankton biomass was highest in 2008 and lowest in 2009 during the summer months. However, all three years had similar chlorophyll concentrations in July (6.4µg/L ± 2.5). The similar chlorophyll concentrations during the early summer, despite the different physical and chemical conditions suggest that the phytoplankton assemblages were not strongly limited by any factor until mid to late summer. Seasonal succession is the temporal change in the relative abundance and dominance of species comprising the natural assemblages (Reynolds, 1988). Through July, fast growing and reproducing phytoplankton that exploit resources (nutrients and light), likely dominated Upper Saint Croix Lake. After July, changes in phytoplankton groups and abundance occurred due to environmental fluctuations apparently related to nutrient conditions, with selective advantages moving to cyanobacteria in 2008. In 2010, chlorophyll concentrations increased to levels almost as high as 2008, but cyanobacteria dominance did not occur due to environmental

influences, most likely low nitrogen availability. Total chlorophyll *a* was understandably low during the winter, as light availability is greatly limited due to snow and ice cover.

The effective quantum yield measurements ranged from 0.44 to 0.57 with a mean of 0.50 across the three sampling years. The factors that cause cell stress included temperature, light, and nutrient limitation. The stress factor that had the greatest influence on the effective quantum yields likely varied throughout the summer and between the years. Stress from high light, which can depress effective quantum yield, could have the greatest impact in the spring and early summer (May to June), during the clear water phase. The clear water phase is the period when the seasonal phytoplankton minimum occurs due to intense zooplankton grazing. However, colored water of Upper Saint Croix Lake, caused by the high concentrations of humic substances, prevents high light intensity from penetrating deep into the water column, reducing the affect of light stress on the phytoplankton, even during the clear water phase. Water temperature likely did not contribute to stressed conditions as the temperature remained within favored conditions for most phytoplankton species. Nutrient stress is speculated in July and August of 2010, when both effective quantum yield and nutrient concentrations were low. However, nutrient stress did not occur during the cyanobacteria bloom of 2008 as effective quantum yields remained high. While the effective quantum yield was often below 0.60, it does not necessarily indicate that the phytoplankton experienced stressed conditions during the three summers. Cyanobacteria typically have a lower quantum yield than green algae, even when the cells are not stressed (Buechel and Wilhelm, 1993). Therefore, phytoplankton in Upper Saint Croix Lake have favorable conditions to maintain high photosynthetic efficiency (high nutrient concentrations and optimal light and

temperature) to grow, and nutrients, especially phosphorus, could be reduced to decrease the probability of cyanobacteria dominance.

Conclusions:

The hydrology around Upper Saint Croix Lake has been altered by humans with the filling of wetlands, the addition of impervious surfaces, and the formation of stream impoundments. These alterations can result in increased runoff entering the lake during precipitation or snowmelt causing a greater influx of nutrients and warmer water, which may lead to increased algal growth. Furthermore, much of the landscape surrounding the lake has had native vegetation (grasses, scrubs, and trees) removed, which naturally filter and uptake nutrients before entering the lake. Even though the hydrology around the lake has been changed by human influences, the influence is beginning to decline. Due to improvements through better wastewater treatment, removal of septic tanks within the watershed, prevention of phosphorus fertilizers, and the addition of a rain garden that filters runoff from downtown, fewer nutrients are likely entering the lake. However because of past periods of high nutrient loading to the groundwater and lake and the relatively poor flushing, it may take decades before nutrients that have entered the lake and sediments are naturally sequestered or flushed out.

Although the alteration of the hydrology around Upper Saint Croix Lake has likely influenced the amount of nutrients that enter the lake, groundwater appears to influence the phosphorus concentrations to a much greater degree. Soluble reactive phosphorus found in groundwater discharge to Upper Saint Croix Lake has been documented to range from 20 to 762µg/L with an average of 107µg/L. These

concentrations are very high for groundwater and it is still unknown whether the sources of the phosphorus in the groundwater near the lake are natural or anthropogenic. With 22% of the water entering Upper Saint Croix Lake originating from these groundwater sources, most of the phosphorus that enters the lake comes from groundwater (645 kilograms per year) (Table 3).

Cyanobacteria blooms only occurred during the summer of 2008 (with the exception of the small observable surface bloom in September of 2009) during the three observation years. The factors that appear to have the greatest influence on controlling the cyanobacteria concentrations in Upper Saint Croix Lake are phosphorus, nitrogen, water temperature, and rainfall. Upper Saint Croix Lake is phosphorus limited (Figure 27), so when phosphorus concentrations increased to 30µg/L and higher in 2008, cyanobacteria blooms occurred. Cyanobacteria blooms, however, did not appear in 2009, even when phosphorus concentrations were above 30µg/L in June and July when high nitrogen concentrations prevented low TN:TP ratios that would favor cyanobacteria. Furthermore, the water temperature was below 20°C (optimal temperature for cyanobacteria is >20°C) in June and July of 2009. Rainfall in 2010 was likely the largest reason that cyanobacteria did not dominate in Upper Saint Croix Lake. Rainfall can influence the amount of nutrients that enter and leave the lake through runoff, dilution, and flushing rate. In 2010, the amount of precipitation was much higher than average, leading to an increase of runoff entering the lake. The large amount of runoff increased the lake level and diluted the amount of phosphorus that enters the lake through the groundwater. Furthermore, the high lake level increased the flushing rate, which can also dilute nutrients, quickly removed the nutrients from the lake, and prevented phosphorus

concentrations from rising above 30µg/L all summer. However, to ascertain whether rainfall or tributary runoff is diluting phosphorus, the concentration of phosphorus in rain and runoff must be determined.

Chapter 4: Summary and Recommendations

Summary:

The main role for determining the factors that control for cyanobacteria dominance in Upper Saint Croix Lake is to allow the watershed managers and residents to possibly avoid the conditions under which cyanobacteria become dominant. Cyanobacteria will always be present in Upper Saint Croix Lake, but certain conditions are needed for toxic blooms. During the summer, if phosphorus concentrations are above 30µg/L, nitrogen concentrations are below 750µg/L, TN:TP ratios are below 40, and water temperature is above 20°C, the likelihood that cyanobacteria will dominant should be expected based on the results of this study.

Nutrient loading throughout the years has lead to abundant nutrients being accrued in the lake sediments. Internal nutrient loading, in addition to unusually high groundwater nutrient inputs, has created more favorable conditions for cyanobacteria in Upper Saint Croix Lake. The high phosphorus, however, is susceptible to inflow and flushing from the system with precipitation. Precipitation also adds relatively high nitrogen concentrations. Thus, low flushing during the summer sets up bloom potential in Upper Saint Croix Lake as internal phosphorus loading and groundwater inputs increase nutrient concentrations.

Recommendations:

Nutrient loading into Upper Saint Croix Lake can be controlled to a certain extent by the residents that surround the lake. Much progress has been made in recent years to remove all the septic tanks around the lake and to improve the treatment of wastewater.

Furthermore, the addition of stormwater diversion and a rain garden that filters runoff from downtown before entering the lake will prevent excessive addition of nutrients. The recent ban on fertilizers containing phosphorus will also assist in the prevention of additional phosphorus entering the lake. However, much of the native vegetation around the lake was removed when homes were built. If more natural buffer zones with native vegetation were built along the lake shore, there would be less nutrient inputs due to runoff filtration and prevention of bank erosion. Furthermore, more vegetative buffers would provide additional shoreline habitat for the native fish that are valued in the lake. However, the amount of nutrients removed may be negligible with results not being observed for many years.

Biomanipulation:

Biomanipulation is a possible method of lake restoration (Shapiro and Wright, 1984). Biomanipulation is typically used in lakes that are small, shallow, and closed systems. Biomanipulation could work in Upper Saint Croix Lake as the method tends to work well in shallow lakes, since organisms are not spatially separated by depth. Biomanipulation involves removing a large percentage of planktivorous and benthivorous fish, either through chemicals (such as Rotenone), nets, or stocking with piscivores. The expected results would be that the abundance of phytoplankton would decrease due to an increased zooplankton abundance and mean size. This in turn would decrease chlorophyll *a* concentrations and increase Secchi transparency. Nutrient concentrations may also decrease because of changes in nutrient fluxes due to the manipulation of the food chain (Carpenter et al., 1992).

However, there are many disadvantages for using biomanipulation. Rotenone may produce undesirable effects, such as killing unintended lake biota. Using nets to remove planktivorous and benthivorous fish is labor intensive, costly, and often inefficient. The success of piscivore stocking is also limited. When used as the sole technique, stocking does not appear to provide long term effectiveness (Shapiro, 1990). One problem is the inability to stock the number of fish required to control abundant planktivore populations (Cooke, 1986). While biomanipulation may produce positive results for several years, the results are often not long-term. Furthermore, many zooplankton species may not graze on cyanobacteria (Bernardi and Giussani, 1990).

Specifically in Upper Saint Croix Lake, biomanipulation is an option in reducing future cyanobacteria blooms, but its success is questionable. Upper Saint Croix is a small lake with the piscivore walleye present. An increase in stocking walleye fingerlings, which have not been stocked in the lake since 1995 (WI DNR, 2012), and a possible increase in minimum size limit of walleye from 38cm to 45cm would likely increase the walleye biomass while decreasing the presently abundant planktivore yellow perch, which in turn, may increase the size and abundance of the present zooplankton. The reduced planktivory through alteration of the trophic levels could reduce algal density and increase water clarity (Lathrop et al., 2002). However, due to the lake's small size and limited habitat for walleye, the Upper Saint Croix Lake may not be able to support increased walleye biomass. Furthermore, without reductions in the amount of phosphorus loading in the lake, piscivore stocking alone may not be sufficient in reducing future cyanobacteria blooms (McQueen et al., 1986). The lake is also susceptible to turbidity because of resuspension of the nutrient-rich sediment by wind. During the summer

months, high nutrient concentrations are often common, which can increase the amount of cyanobacteria before the biomanipulation technique has altered the food chain and reduced nutrient fluxes.

Aluminum Sulfate:

Because phosphorus is the limiting nutrient in Upper Saint Croix Lake, reduction in water column phosphorus may lead to increased transparency, decreased phytoplankton abundance and a shift in species abundance away from cyanobacteria (Edmonson and Lehman, 1981). Sediment phosphorus release is an important source of phosphorus in Upper Saint Croix Lake. Aluminum sulfate (as known as alum) can be a potentially long-term method of controlling sediment phosphorus release. Alum is added to the water column to form aluminum phosphate and a colloidal hydroxide floc, which binds to phosphorus. The floc then settles and consolidates with the sediments where it continues to sorb and retain phosphorus (Welch and Schrieve, 1994). However, to control phytoplankton, repeated treatments are necessary. Furthermore, bioaccumulation of aluminum may occur at toxic levels in fish due to the repeated applications (Cooke, 1986). Although aluminum sulfate would effectively reduce the amount of phosphorus in Upper Saint Croix Lake and likely decrease the probability of cyanobacteria dominance, the expensive cost of the aluminum and potentially loss of fish due to aluminum toxicity would outweigh the advantages.

Addition of Nitrogen

Theoretically, the addition of nitrogen to Upper Saint Croix Lake should increase the N:P ratio to favor more suitable algal communities. However, without a reduction of phosphorus to alter the N:P ratio, cyanobacteria could still dominate. If nitrogen is added to Upper Saint Croix Lake, cyanobacteria, which can utilize diverse forms of nitrogen (both inorganic and organic) (Paerl, 1988), may have a competitive advantage over other phytoplankton. Organic nitrogen and ammonium enrichment has also been documented to favor cyanobacteria (Pinckney et al., 1997) and toxicity of bloom species (Paerl and Millie, 1996). Furthermore, with the relatively short residence time of Upper Saint Croix Lake, nitrogen additions would likely not provide a long-term benefit, if any benefit to cyanobacteria control occurred.

Sediment Removal:

Nutrients are released in large quantities from sediment in Upper Saint Croix Lake (Hoverson and McGinley, 2007). Heavy equipment or specialized hydraulic dredges can remove accumulated sediments to increase depth and remove nutrient-rich sediment. By removing nutrient-rich sediment, water quality may improve due to reduced internal loading of phosphorous (Lehrke, 1997). However, sediment removal is labor intensive, expensive, and the success is limited in shallow lakes. Sediment removal leads to resuspension of nutrients through agitation of the sediment during dredging activities. Resuspension can also move phosphorus into the euphotic zone, leading to algal blooms. Sediment removal would not be a practical method in decreasing the probability of cyanobacteria dominance in Upper Saint Croix Lake as even if internal phosphorus

loading is decreased, high phosphorus groundwater inputs would still add excessive phosphorus to the lake.

Dilution:

Dilution involves the addition of low-nutrient water into the lake as a means of diluting and flushing the nutrients from the lake. This method creates a situation similar to the high rainfall levels that were observed in 2010. The increased flushing can remove surface phytoplankton and replace the high-nutrient lake water with lower nutrient water. However, for dilution to succeed, large volumes of low-nutrient water are required. Furthermore, the dilution method does not remove sources of nutrients from the sediments. Dilution has successfully been employed for lake restoration at Moses Lake and Green Lake, both in the state of Washington (Welch, 1981). Dilution would likely be a successful method of lake restoration and reduce the probability of cyanobacteria dominance in Upper Saint Croix Lake. However, dilution is not possible for Upper Saint Croix Lake because there are no rivers or large lakes with low-nutrient water close enough to divert into the lake. Even though Lake Superior is only 30 miles from Upper Saint Croix Lake, the cost of diverting water (either by river or pipeline) would be too expensive and impractical.

Final Remarks*:*

Unfortunately, there is no immediate solution to the problem of occasional cyanobacteria blooms in Upper Saint Croix Lake. Anthropogenic nutrients entering the lake have been reduced to a minimal level, but nutrients that enter the lake through

groundwater cannot be altered (unless the phosphorus sources in the groundwater are anthropogenic). Because nutrient concentrations and weather conditions cannot be controlled, the factors that control for cyanobacteria dominance will vary each year. Conditions that favor cyanobacteria dominance (high phosphorus, low TN:TP, and warm water temperature) like in 2008 may not occur for several more years (low water temperature in 2009 and high flushing rates in 2010).

Since there is little that can be done to address the conditions that favor cyanobacteria in Upper Saint Croix Lake, the best option is to try and predict when cyanobacteria dominance will occur before a bloom occurs. An inexpensive instrument that could be installed in Upper Saint Croix Lake is a piezometer. A piezometer is used to measure static liquid pressure by measuring the pressure of water at a certain point. The piezometer would allow accurate and precise information to be gathered on when the lake level is changing. This information would update the residents of Solon Springs whether groundwater or surface water is influencing the water in Upper Saint Croix Lake. Even without a piezometer, a simple warning system for cyanobacteria blooms is measuring lake level changes, especially in June and July. If lake level remains stable or decreases in June and July, the lake may not experience enough flushing and dilution of groundwater inputs, which could result in higher phosphorus concentrations in the late summer, leading to cyanobacteria blooms if other conditions are met. Additionally, increased water temperature measurements could be made during the summer to determine when the water rises above 20°C, the critical point before cyanobacteria dominance can occur.

Figures and Tables

Table 1: Upper Saint Croix Lake Morphometry (Turyk and Macholl, 2010)

Characteristic	Value
Surface Area	3,350,797m^2
Mean Depth	~3m (varies)
Max Depth	~6.7m (varies)
Littoral Zone (<5m) Area	84.5%
Residence Time	0.45 year (varies)
Shoreline Length	15.29 km
Developed Shoreline Length	10.30 km

Table 2: Hydrologic Budget of Upper Saint Croix Lake (Turyk and Macholl, 2010)

Source	Percentage
Tributaries	45%
Groundwater	22%
Direct Surface Runoff	18%
Direct Precipitation	15%

Table 3: Annual Phosphorus Budget of Upper Saint Croix Lake (Turyk and Macholl, 2010)

Source	P Quantity Kilograms/Year
Internal Release	829
Groundwater and Streams	645
Atmospheric Deposition	78
Total Phosphorus Entering Lake	1552
Phosphorus Leaving Lake in Outflow	506

Table 4: Algal taxonomic identification and counts by Hedy Kling from site DHN (0.5m) on July 23, 2008

Length	Width	Count	Group	Volume	Cells or units/L	mg/m^3	Taxon name
11.2	11.2	320	CYAN	735.6	320000	235.4	Anabaena crassa
12.6	11.2	8	HETE	0	8000	0	Anabaena heterocysts
24	2	1	CHLO	37.7	29838	1.1	Ankyra lanceloata (Kors) Fott
1.4	1	320	CYAN	1.1	9548160	10.5	Aphanothece bachmanii
1.2	1.2	1650	CYAN	0.9	49232700	44.5	Aphanocapsa delicatissima
1	1	2000	CYAN	0.5	59676000	31.2	Aphanocapsa holsatica (Lemm) Cronb & Kom.
28	8.4	26	DIAT	1551.7	26000	40.3	Aulacoseira ambigua
44	8.4	81	DIAT	2438.4	81000	197.5	Aulacoseira granulata
24	6.4	17	DIAT	772.1	17000	13.1	Aulacoseira granulata

44	40	2	PERI	36861.4	2000	73.7	Ceratium furcoides
4.2	2.8	1	HAPT	11.5	29838	0.3	Chrysochromulina parva
84	3.2	1	CHLO	225.2	1000	0.2	Closterium acutum
3	3	256	CYAN	14.1	256000	3.6	Coelospaerium kuetzingiana
12	2	1	CHLO	18.8	29838	0.6	Elakatothrix genevensis
84	4.2	9	DIAT	387.9	9000	3.5	Fragilaria capucina
11.2	4.2	61	DIAT	51.7	61000	3.2	Fragilaria crotonensis
38	20	2	PERI	5305.8	2000	10.6	Gymnodinium uberimuum
11.2	9.6	2	CHRY	540.5	59676	32.3	Large chrysophytes (Ochromonads)
4.8	4.2	50	CYAN	44.3	1491900	66.1	Microcystis aeruginosa
5	4.2	9750	CYAN	46.2	9750000	450.3	Microcystis aeruginosa
6.4	5.6	4352	CYAN	105.1	4352000	457.3	Microcystis wesenbergi
5	4.2	6	CHRY	46.2	179028	8.3	Ochromonas sp
12.6	8.4	4	CHLO	310.3	4000	1.2	Oocystis lacustris Chod.
100	84	1	ROTI	246301	1000	246.3	Polyarthra
6.4	6.4	32	CHLO	137.3	32000	4.4	Pseudosphaerocystis lacustris
66	40	1	ROTI	36861.4	1000	36.9	Rotifer
40	20	1	DIAT	12566.4	1000	12.6	Stephanodiscus niagarae
2.8	2	4	CHRY	5.9	119352	0.7	Tiny chrysophyte flagellates
1.2	1	2720	CYAN	0.6	81159360	51	micro bluegreens
1.4	1.2	480	CHLO	1.1	14322240	15.1	micro greens
5.6	5.6	1500	CHLO	92	1500000	137.9	Urotricha farcta (Scuticociliate)
5.6	2.8	4	CYAN	23	119352	2.7	Woronichinia naegeliana (Unger) Elenkin
6.4	3.2	512	CYAN	34.3	512000	17.6	Woronichinia naegeliana (Unger) Elenkin

	mg/m^3	**%**	**Cells or units/L**	**%**
Cyanophyte	1370.4	71.1	216417472	92.9
Chlorophyte	160.6	8.3	15918916	6.8
Euglenophyte	0	0	0	0
Chrysophyte	41.2	2.1	358056	0.2
Haptophyte	0.3	0	29838	0
Bacillariophyte	270.2	14	195000	0.1
Cryptophyte	0	0	0	0
Dinophyte	84.3	4.4	4000	0
Xanthophyte	0	0	0	0
	========	==========	==========	==========
TOTAL	1927	100	232923282	100

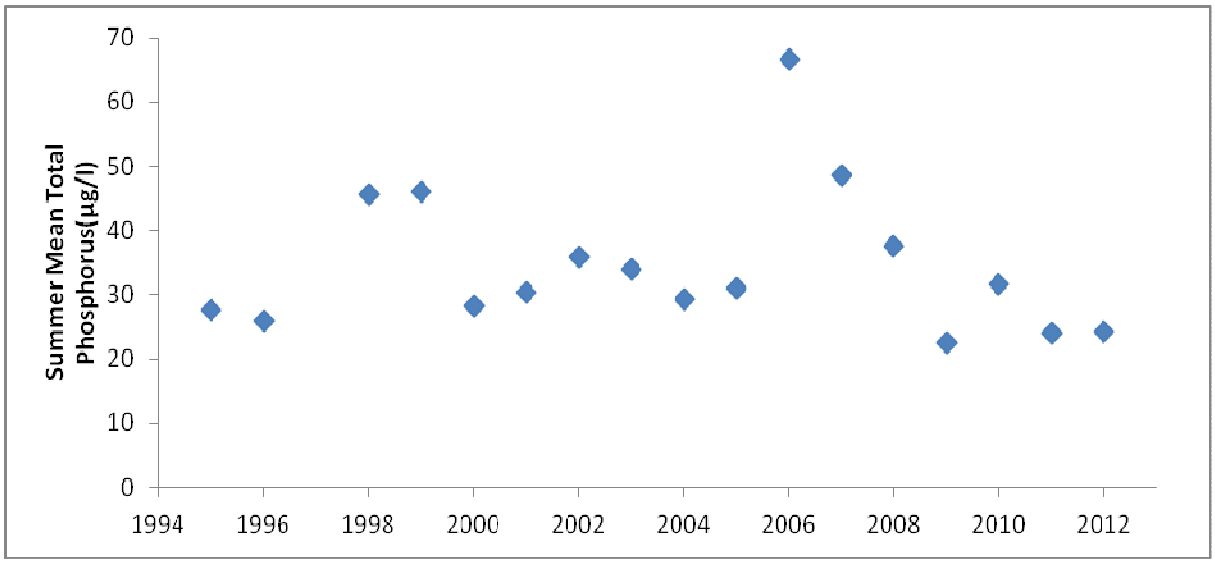

Figure 1: Annual changes in mean summer total phosphorus concentrations at DHS (WI DNR, 2012). Mean total phosphorus concentration values from June through September. Data collected and analyzed by WI DNR.

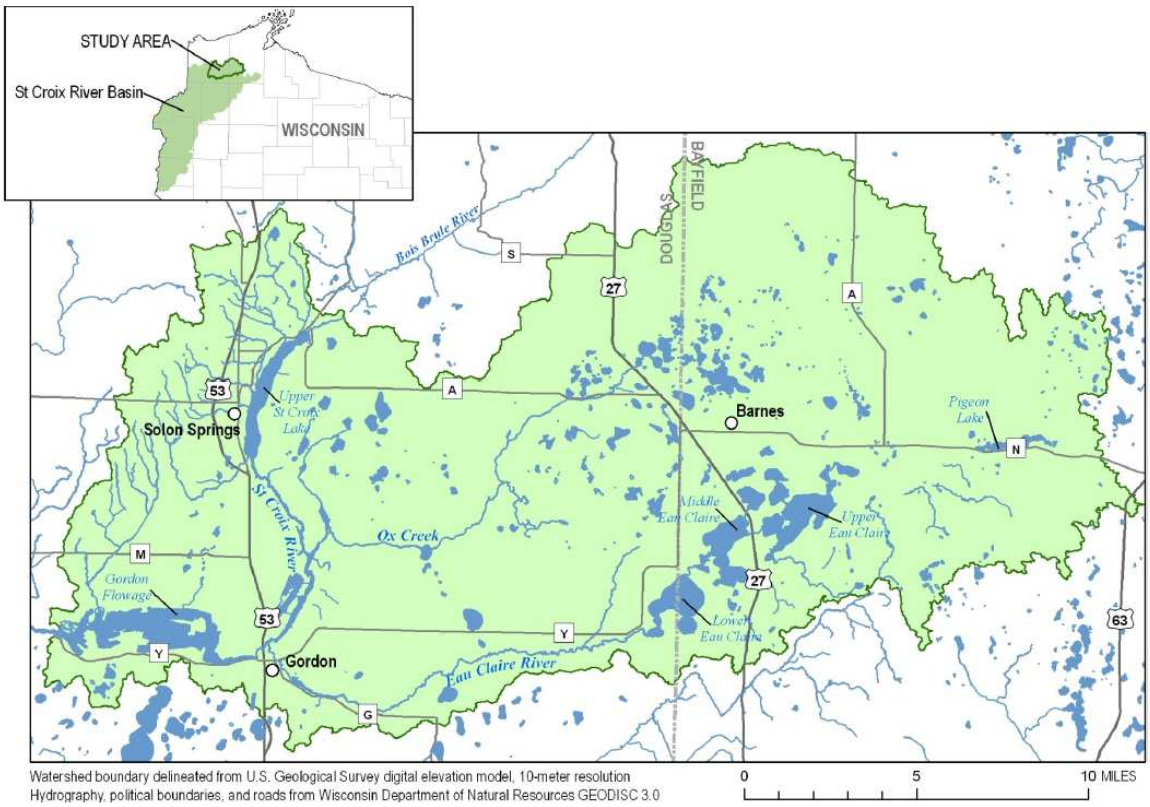

Figure 2: Upper St. Croix – Eau Claire River Watershed in Douglas and Bayfield Counties, Wisconsin (UW-Stevens Point Center for Watershed Science and Education et al., 2011).

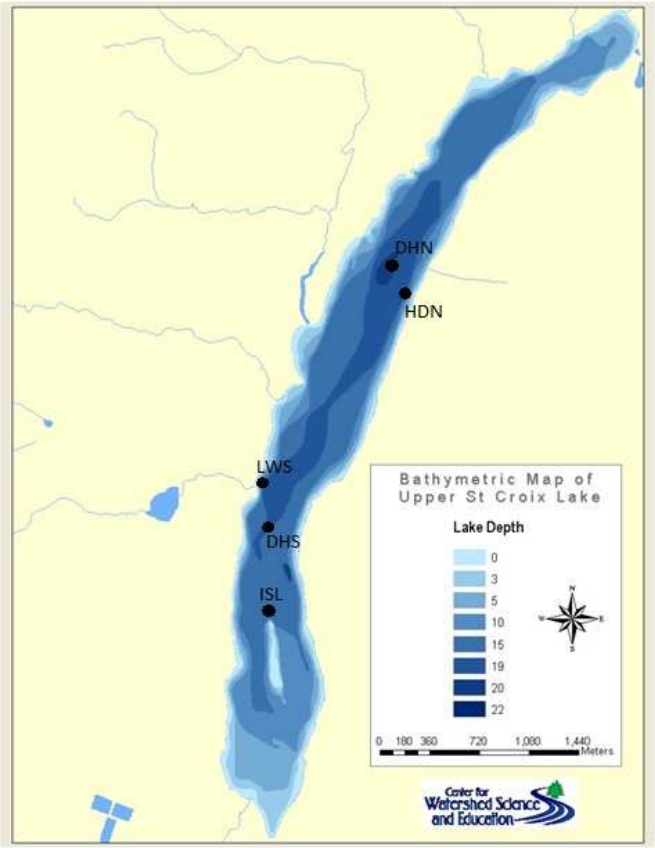

Figure 3: Bathymetric map of Upper Saint Croix Lake with sampling sites (Turyk and Macholl, 2010).

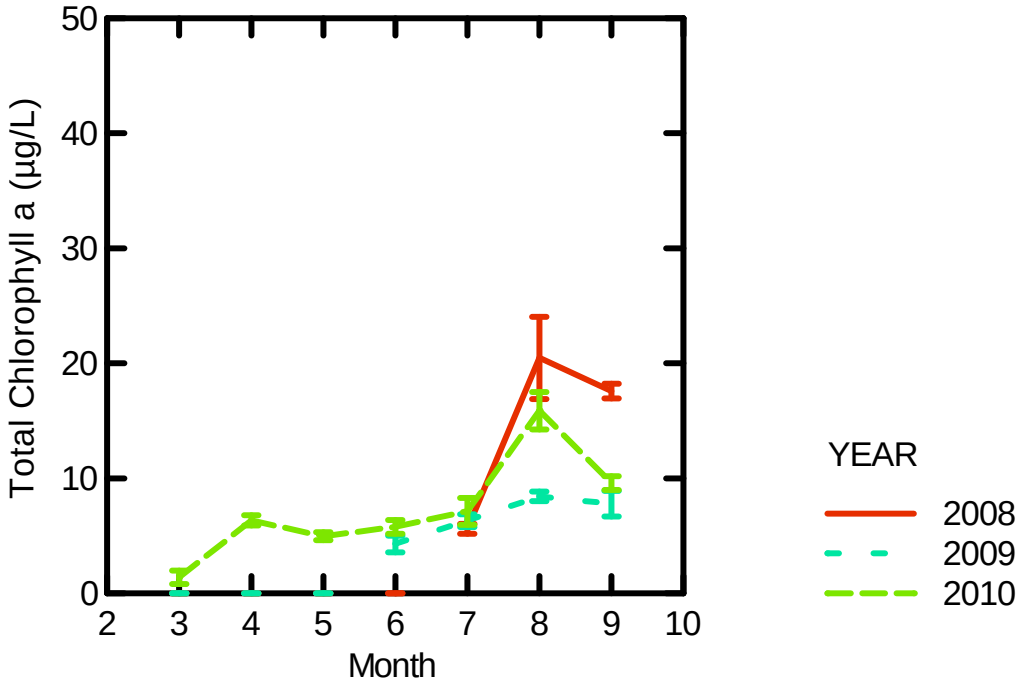

Figure 4: Seasonal changes in extracted chlorophyll *a* during sampling years. Monthly means taken from the average of all stations and depths. Error bars given in SE. No significant difference between stations. There was no significant difference between chlorophyll *a* concentration and depth in 2008 or 2009, but chlorophyll a was significantly higher in the surface water than 4m in 2010.

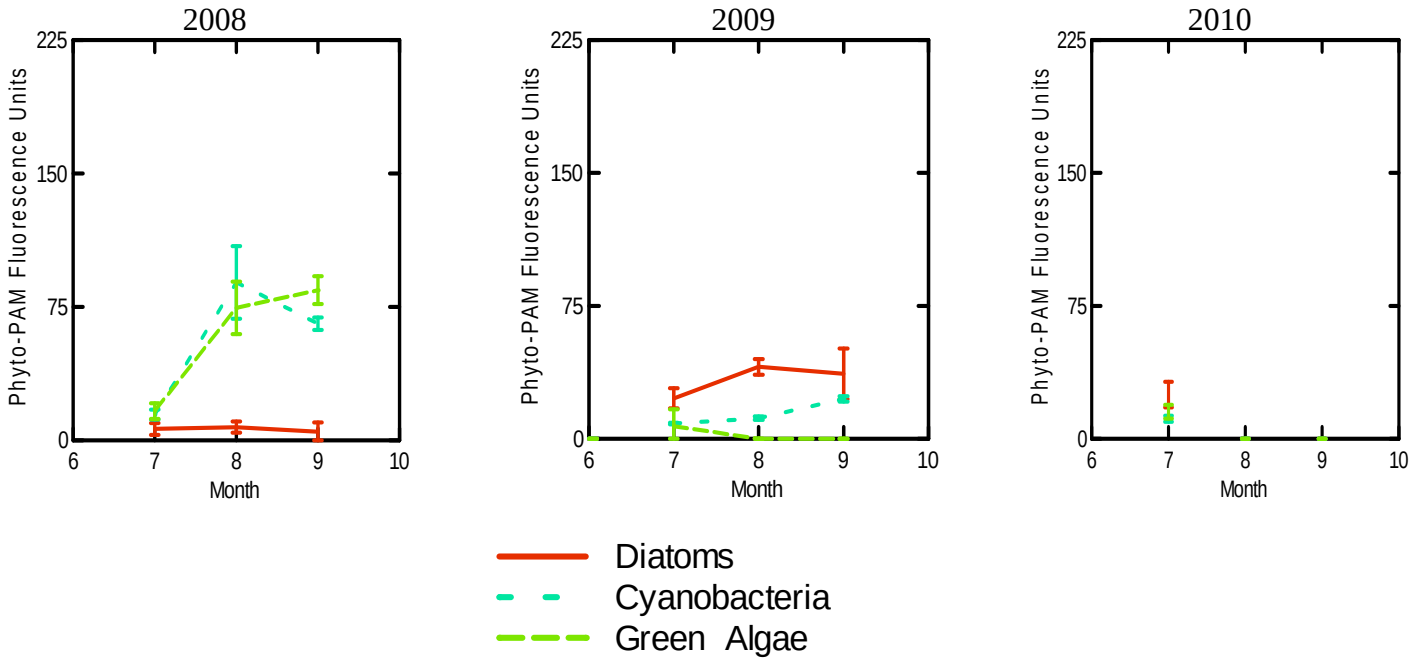

Figure 5: Seasonal changes in algal relative abundance (unitless) measured with the Phyto-PAM® during the summer months. Value for each month is the average of all stations and depths sampled on that date. Error bars given in SE. Data only available for July in 2010.

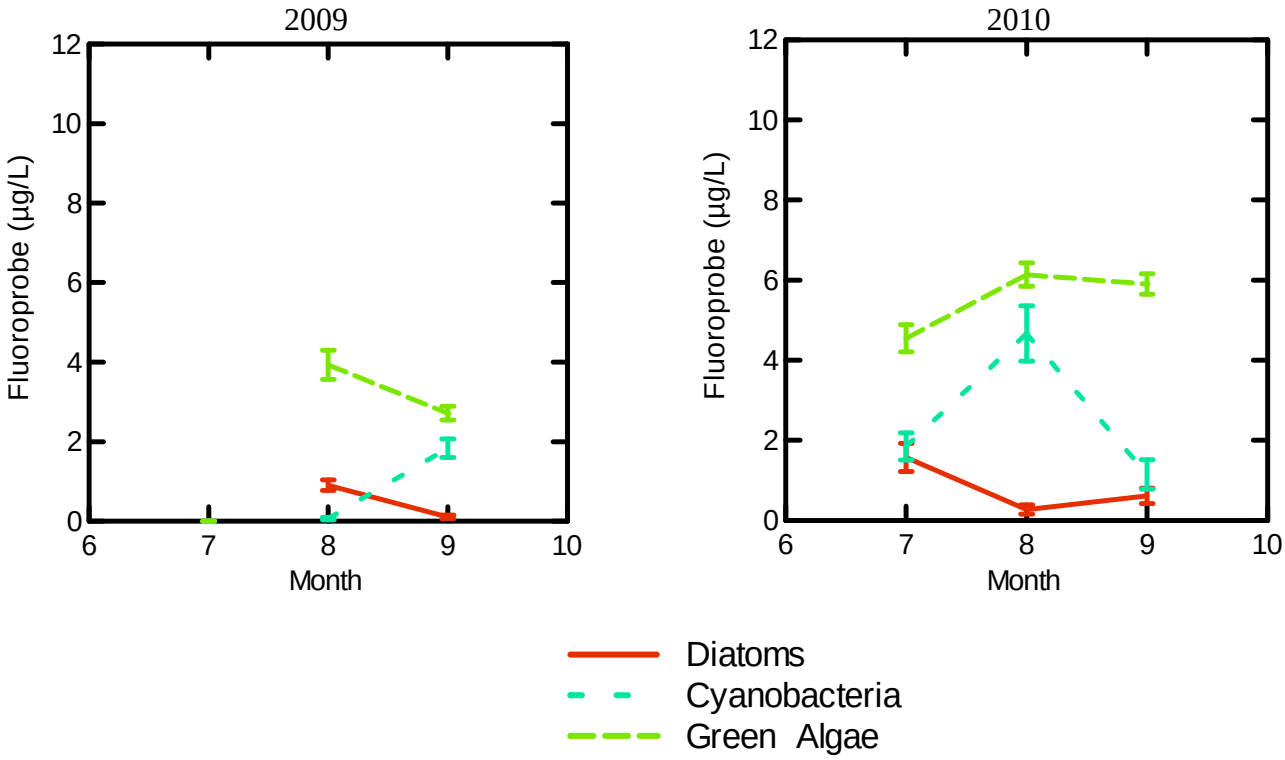

Figure 6: Changes in algae concentrations measured with the Fluoroprobe® during the summer months. Value for each month is the average of all stations and depths sampled on that date. Error bars given in SE. No data available from 2008.

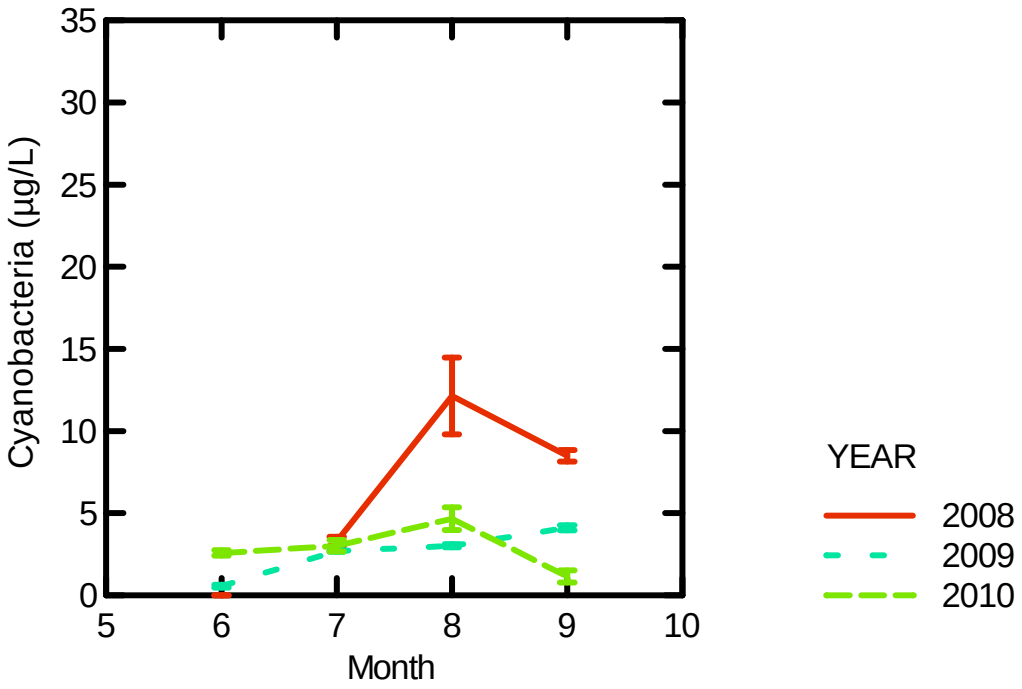

Figure 7: Seasonal changes in cyanobacteria chlorophyll concentrations during sampling years. Value for each month is the average of all stations and depths sampled on that date. 2008-2010 (July) data is derived from Phyto-PAM® linear regression and 2010 (August and September) is derived from Fluoroprobe® linear regression. Error bars given in SE.

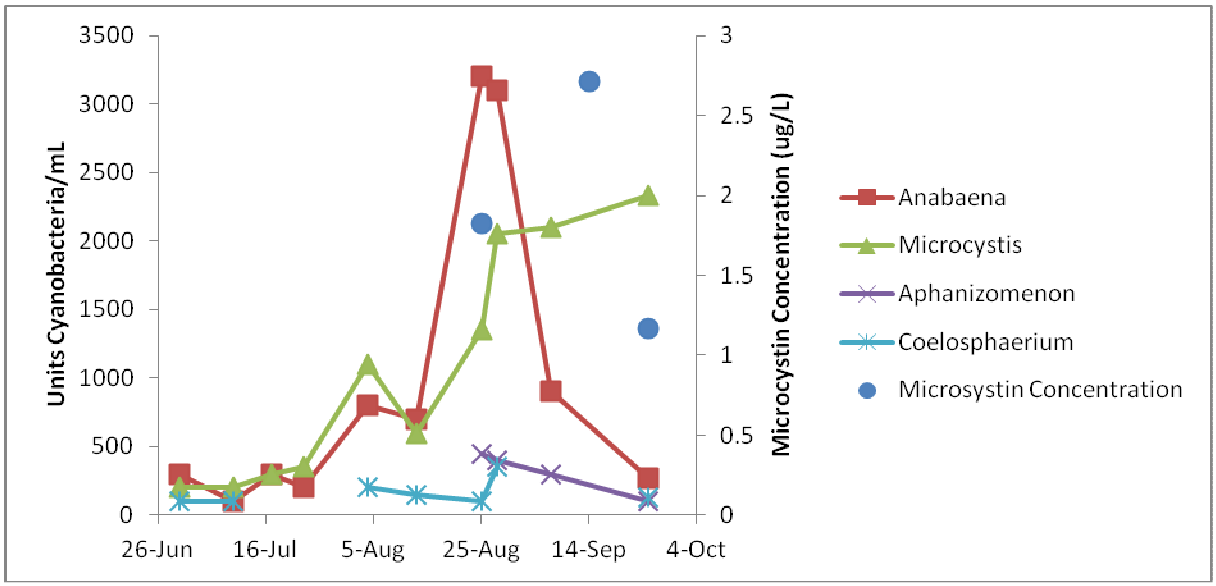

Figure 8: Cyanobacteria and Microcystin-LR concentrations during the cyanobacteria bloom year of 2008. The total number of visually counted units is equal to total number filaments and colonies per milliliter. >1.0 µg/L (ppb) indicates the maximum level for microcystin concentration in drinking water as recommended by the World Health Organization.

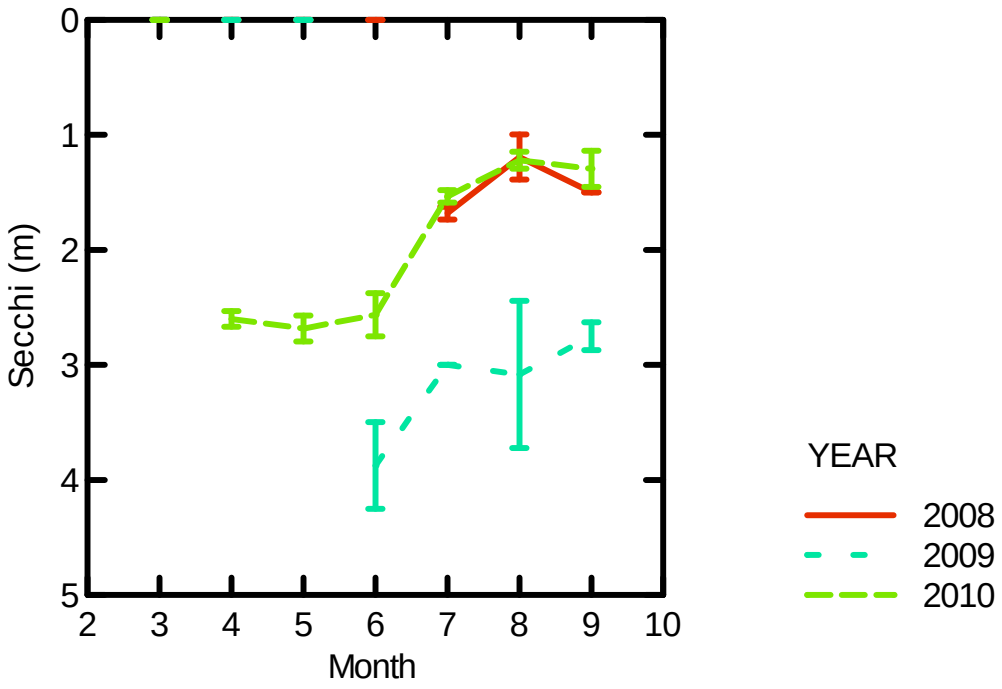

Figure 9: Seasonal changes in Secchi depth across sampling years. Monthly means taken from the average of all stations. Error bars given in SE. Secchi depth in 2009 is significantly deeper than 2008 and 2009.

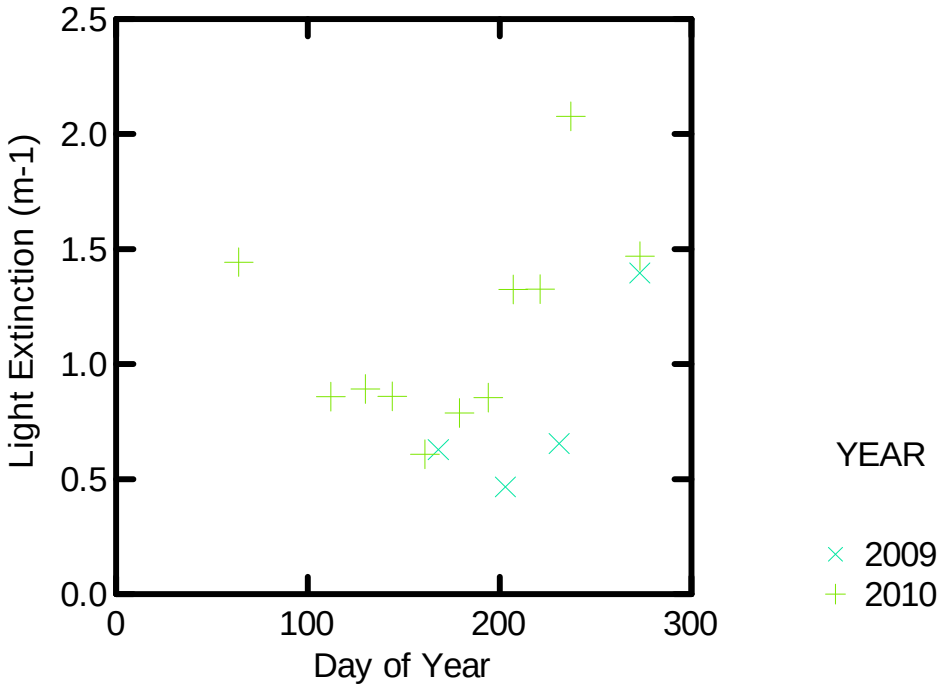

Figure 10: Seasonal changes in light extinction at Station 4 (DHN) across sampling years derived from Li-Cor light meter. Error bars given in SE. January 1st is day 0 and December 31st is day 365. No data from 2008.

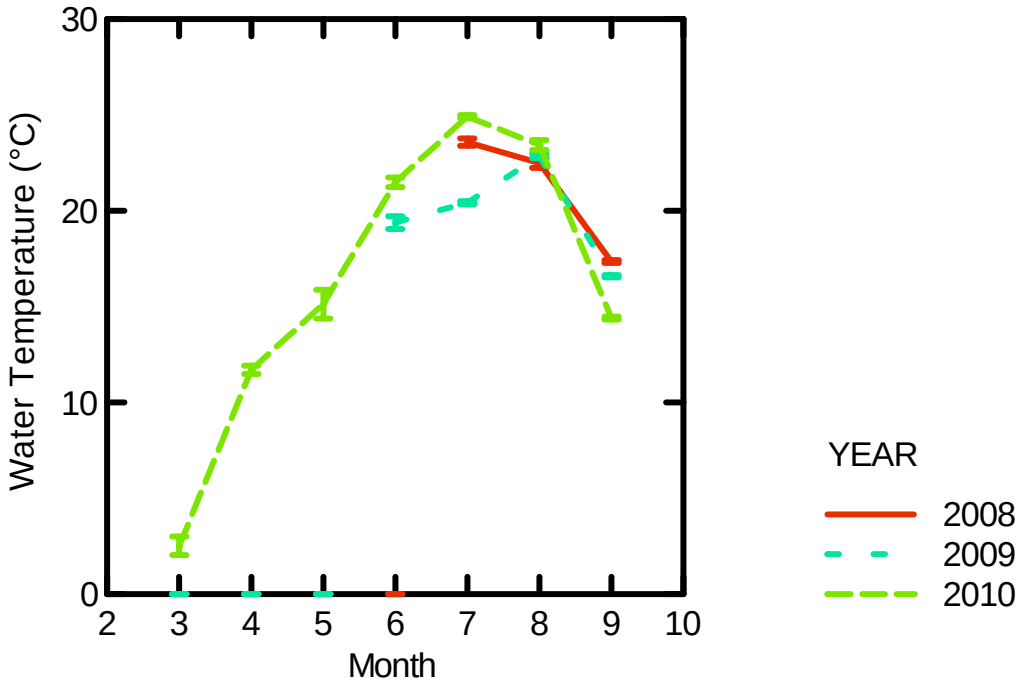

Figure 11: Seasonal changes in surface water temperature across sampling years. Monthly means taken from the average of all stations. Error bars given in SE. There is no significant difference in water temperature throughout the water column or between stations.

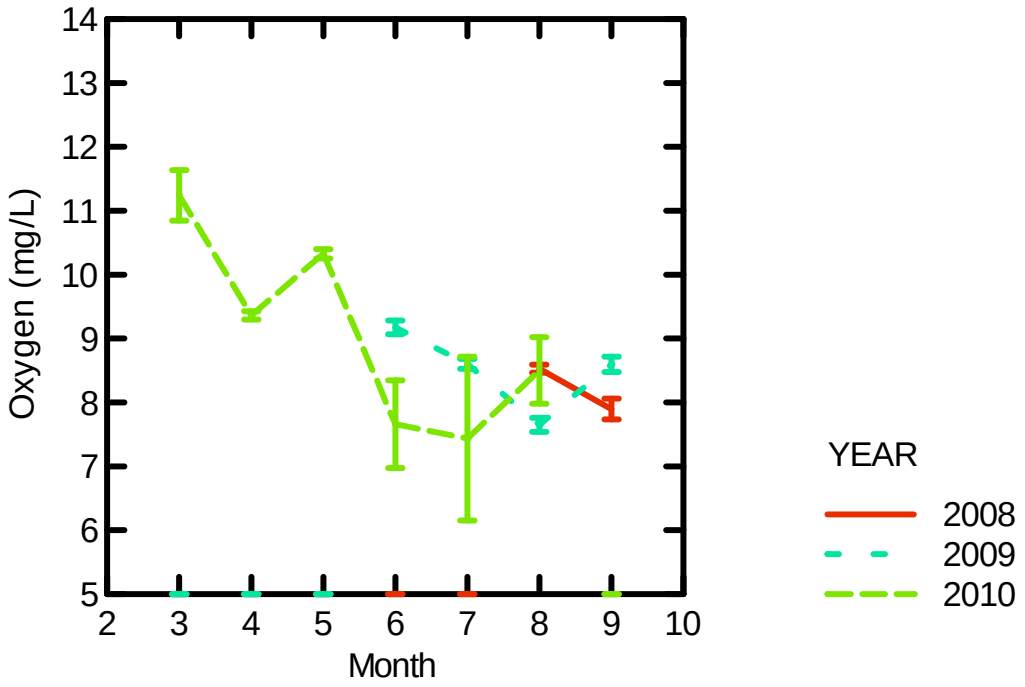

Figure 12: Seasonal changes in oxygen concentrations across sampling years. Monthly means taken from the average of all stations. Error bars given in SE. There was no significant difference in oxygen concentration between any years or with depth in 2008 and 2009. Surface oxygen concentration was significantly higher than 4m and deeper in 2010.

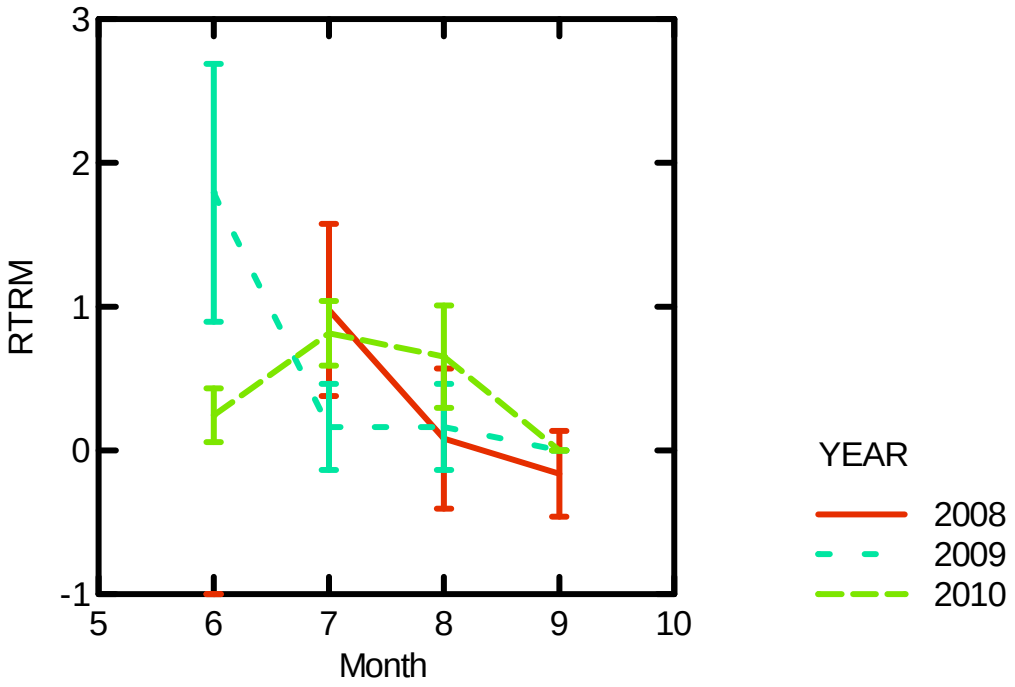

Figure 13: Seasonal changes in relative thermal resistance to mixing across sampling years at DHS. Monthly means taken from the average of all stations. Error bars given in SE. There was no significant difference in RTRM between any years, except in June 2009.

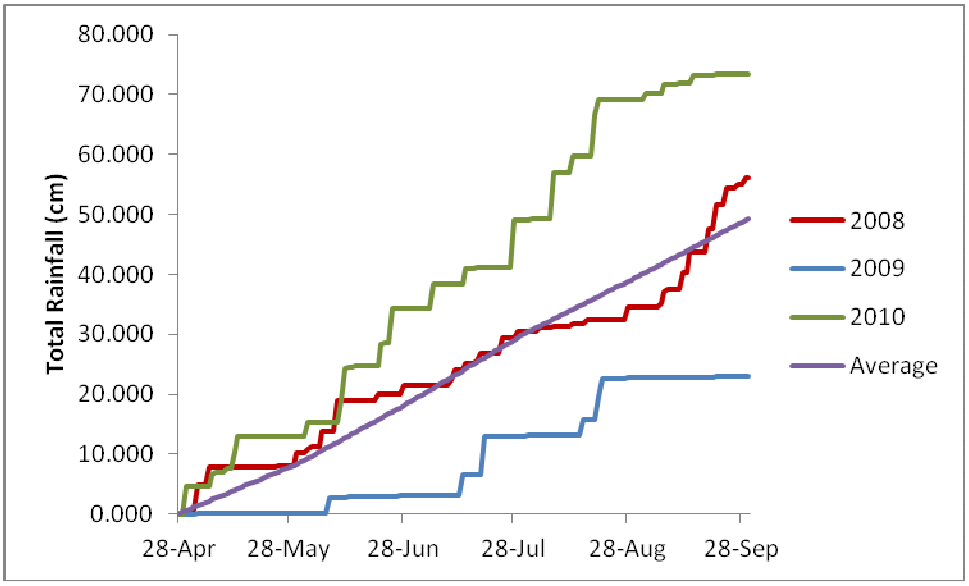

Figure 14: Rainfall totals displaying every rain event during the sampling years. The average amount of rainfall is the total amount of precipitation that normally falls from May through September divided evenly by the total number of days during that period. Rainfall data collected daily from rain gauge placed by shoreline near station HDN.

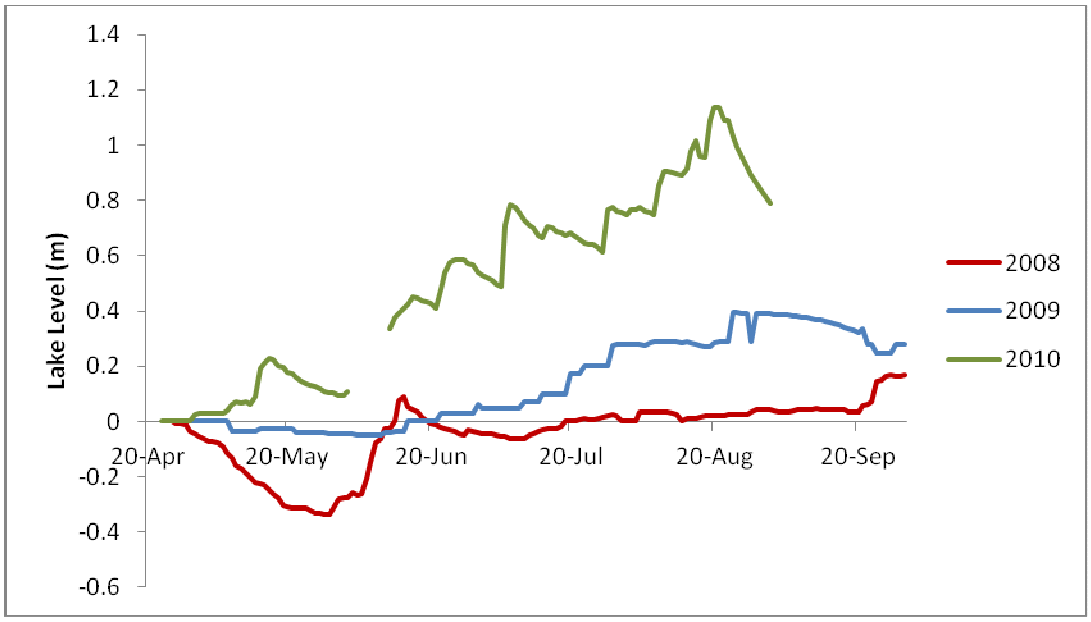

Figure 15: Changes in relative lake level for each sampling year. When lake level is stable, inflow is equal to outflow. When lake level is increasing, inflow > outflow. When lake level is decreasing, inflow < outflow. Relative lake level determined daily by changes in water depth near station HDN shoreline.

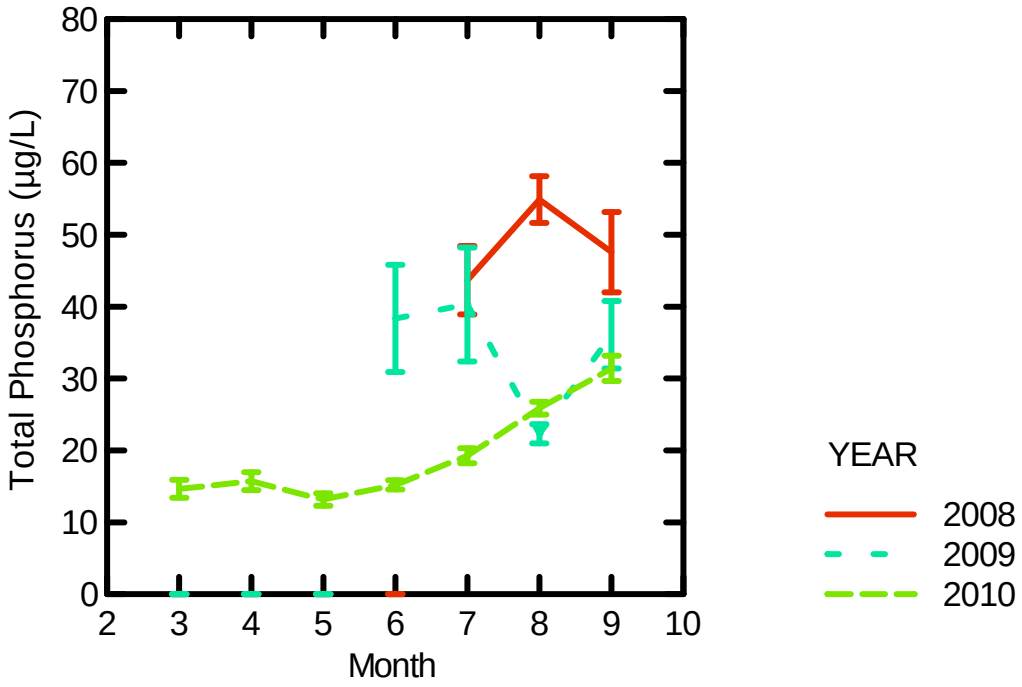

Figure 16: Seasonal changes in total phosphorus across sampling years. Monthly means taken from the average of all stations and depths. Error bars given in SE. No significant difference between station and depth during any sampling year. August is the critical period for phosphorus availability for cyanobacteria.

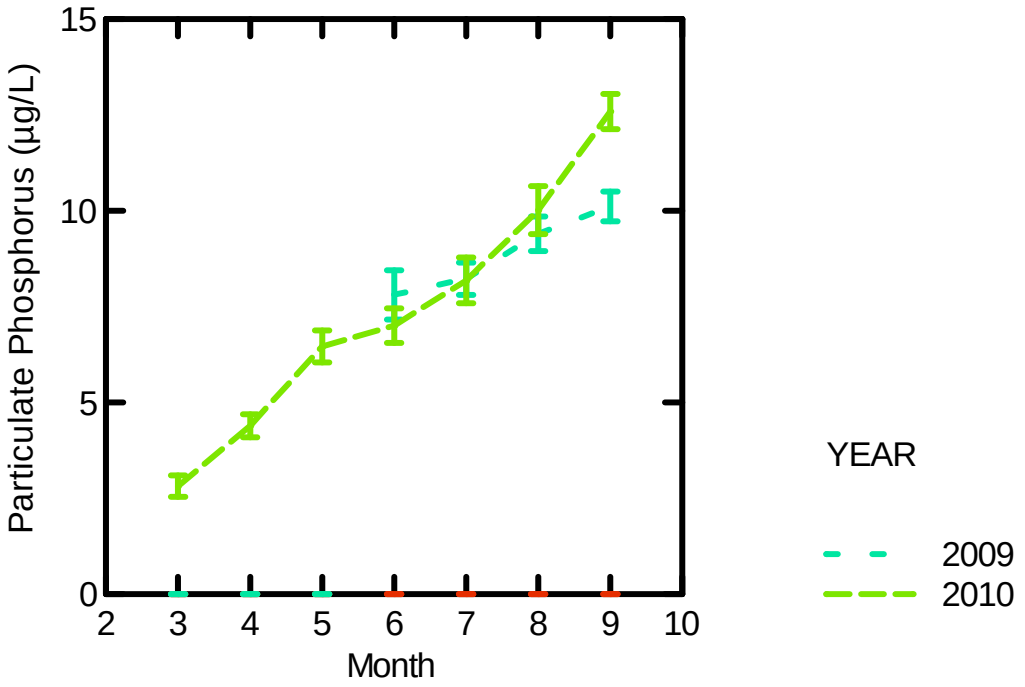

Figure 17: Seasonal changes in particulate phosphorus across sampling years. Monthly means taken from the average of all stations and depths. Error bars given in SE. Average across all depths and stations. No significant difference between station and depth during any sampling year. No data from 2008.

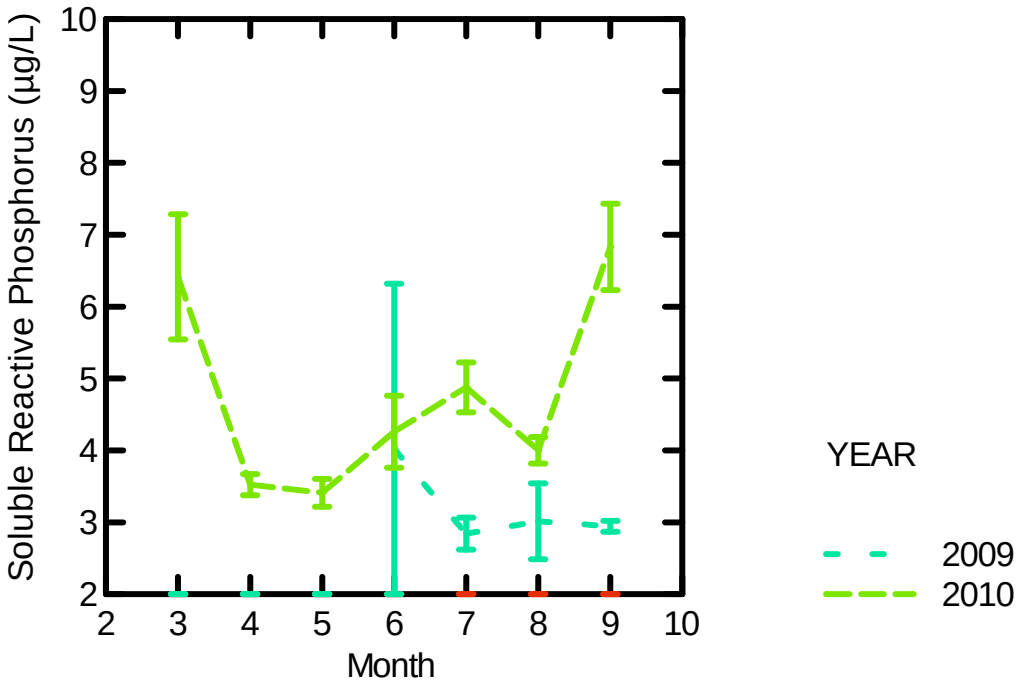

Figure 18: Seasonal changes in soluble reactive phosphorus across sampling years. Monthly means taken from the average of all stations and depths. Error bars given in SE. Average across all depths and stations. No significant difference between station and depth during any sampling year. No data from 2008.

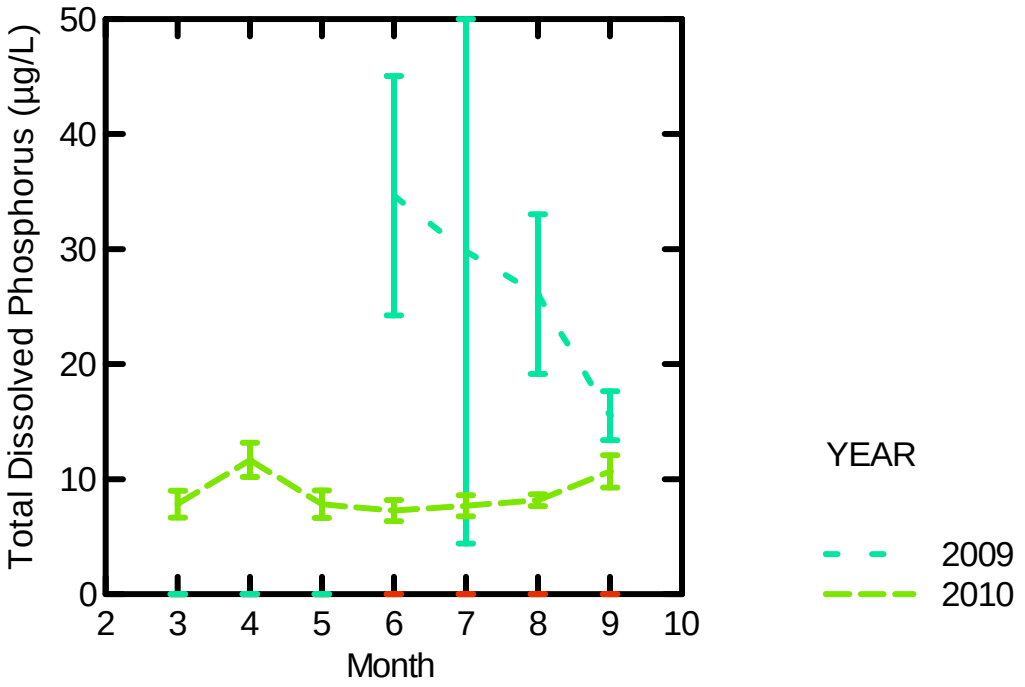

Figure 19: Seasonal changes in total dissolved phosphorus across sampling years. Monthly means taken from the average of all stations and depths. Error bars given in SE. Average across all depths and stations. No significant difference in station during any sampling year. In 2010, TDP was significantly higher at 2m than surface water. No data from 2008.

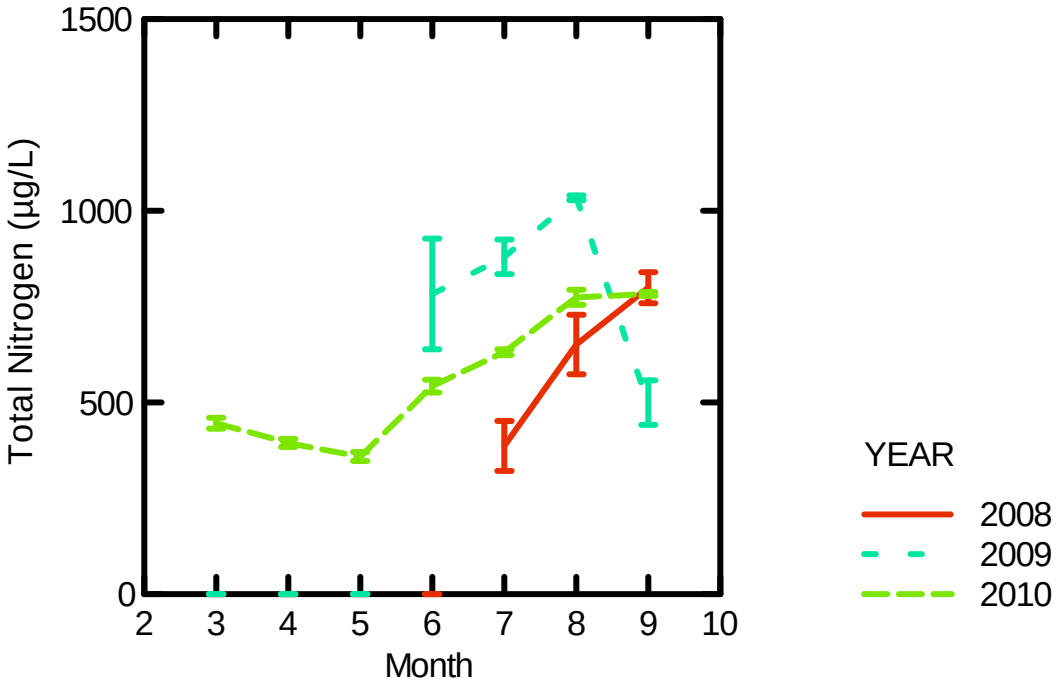

Figure 20: Seasonal changes in total nitrogen across sampling years. Monthly means taken from the average of all stations and depths. Error bars given in SE. Average across all depths and stations. No significant difference between station and depth during any sampling year.

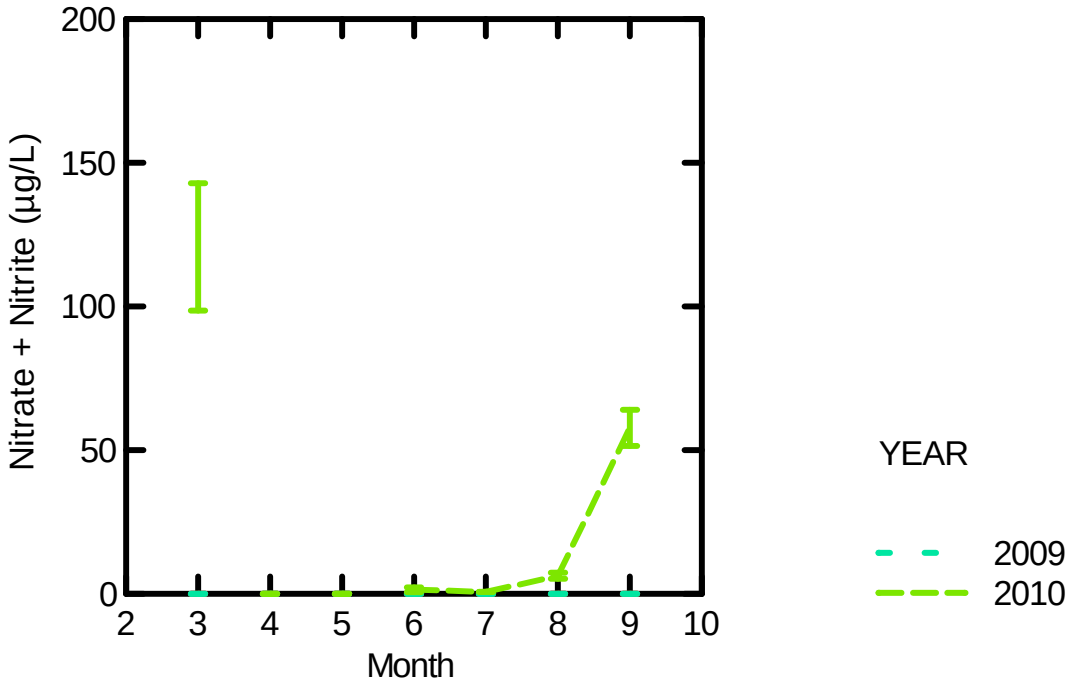

Figure 21: Seasonal changes in nitrate + nitrite across sampling years. Monthly means taken from the average of all stations and depths. Error bars given in SE. Average across all depths and stations. No significant difference between station and depth during any sampling year. All nitrate + nitrite concentrations from 2009 were below detection limit (< 0.1µg/L). No data from 2008.

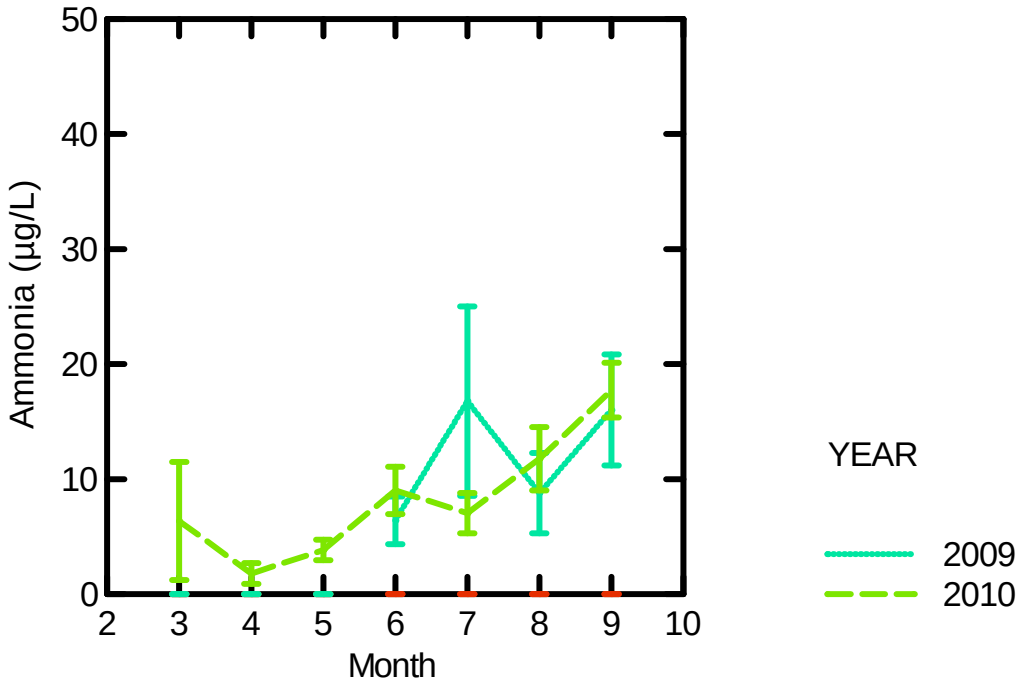

Figure 22: Seasonal changes in ammonia across sampling years. Monthly means taken from the average of all stations and depths. Error bars given in SE. Average across all depths and stations. No significant difference between stations during any sampling year. Ammonia was significantly higher at 4m than 2m or surface water in 2010. No data from 2008.

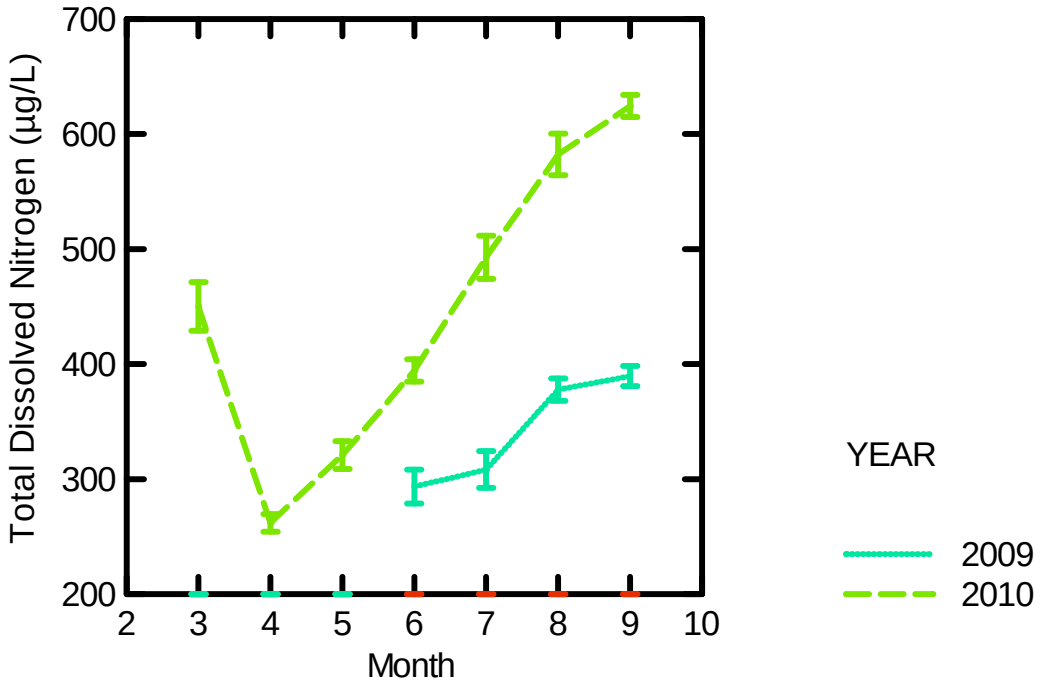

Figure 23: Seasonal changes in total dissolved nitrogen across sampling years. Monthly means taken from the average of all stations and depths. Error bars given in SE. Average across all depths and stations. No significant difference between station and depth during any sampling year. No data from 2008.

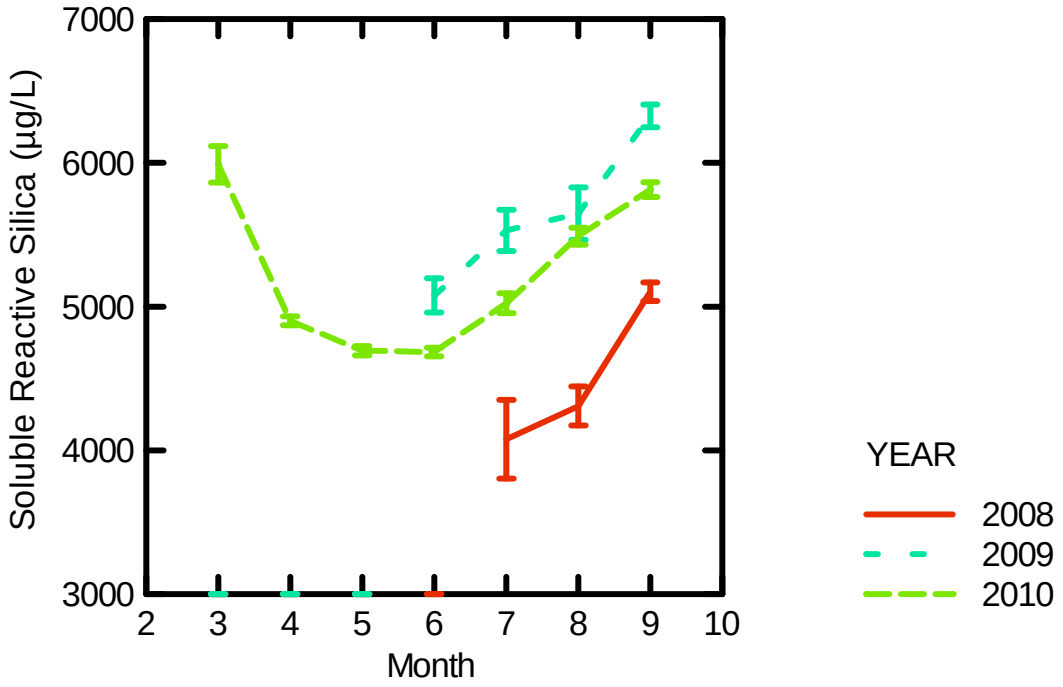

Figure 24: Seasonal changes in soluble reactive silica (SiO2) across sampling years. Monthly means taken from the average of all stations and depths. Error bars given in SE. Average across all depths and stations. No significant difference between station and depth during any sampling year.

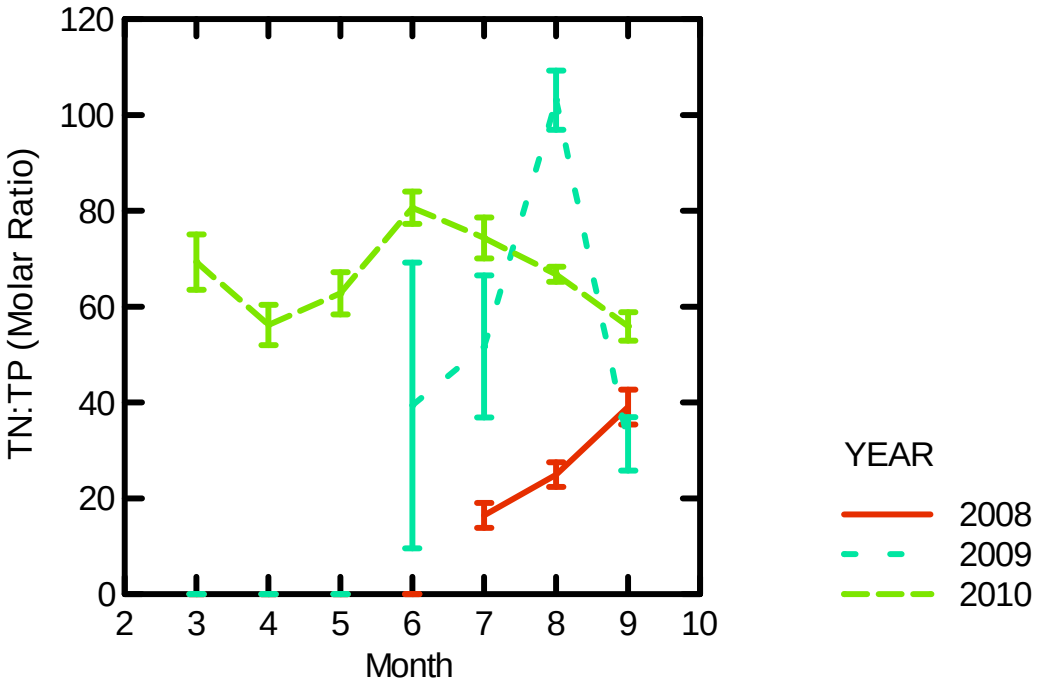

Figure 25: Seasonal changes in the molar ratio of total nitrogen to total phosphorus across sampling years. Monthly means taken from the average of all stations and depths. Error bars given in SE. Average across all depths and stations. No significant difference between station and depth during any sampling year.

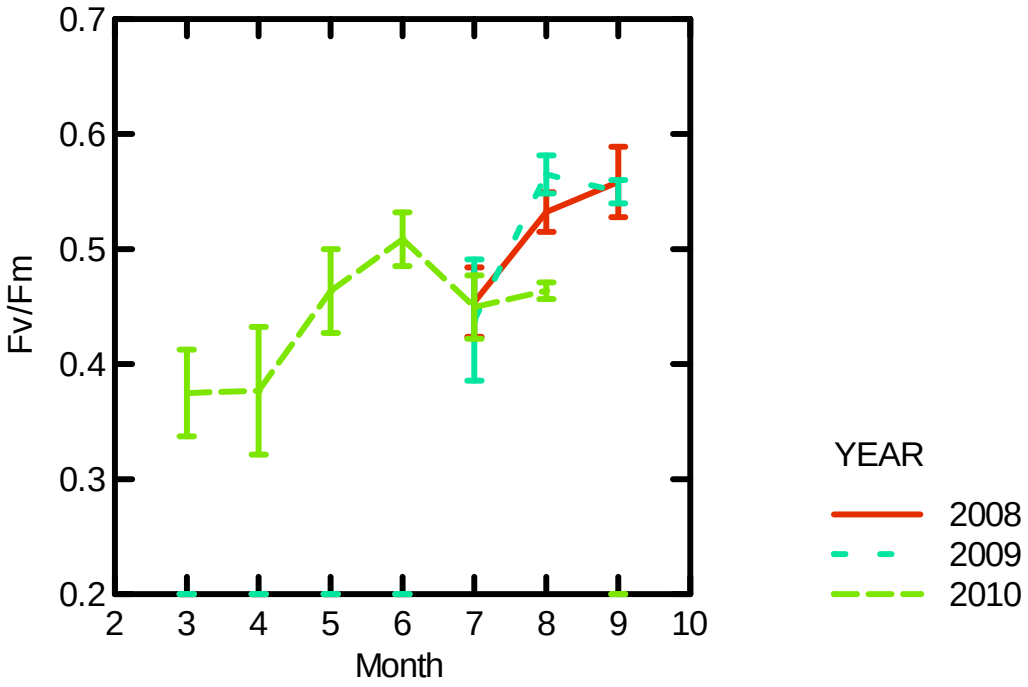

Figure 26: Seasonal changes in effective quantum yield (Fv/Fm) values across sampling years. Monthly means taken from the average of all stations and depths. Error bars given in SE. No significant difference between depths and stations.

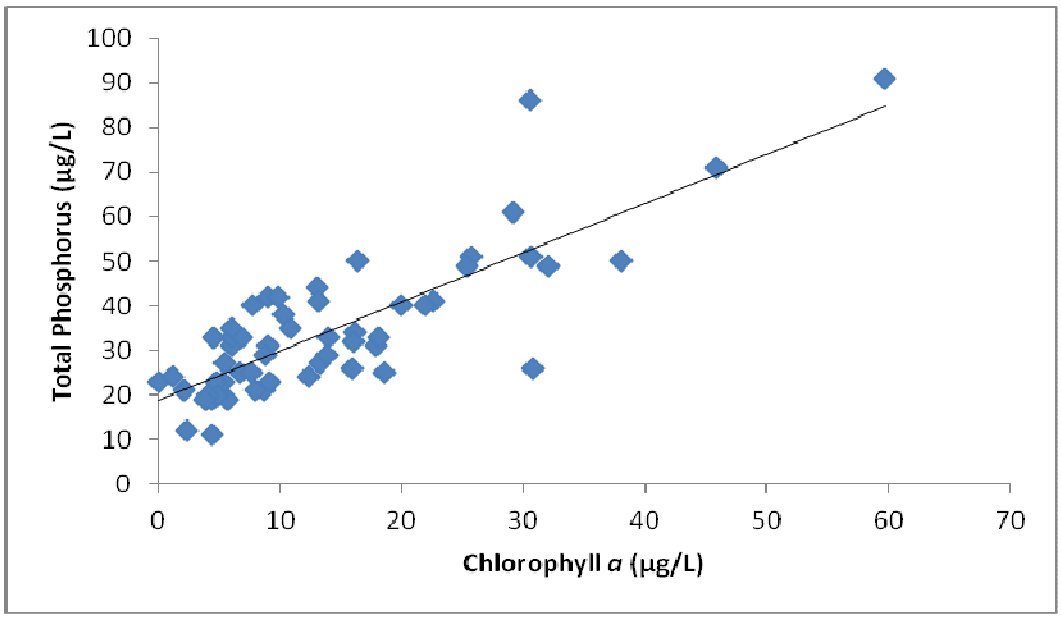

Figure 27: Correlation between total phosphorus and chlorophyl *a* in Upper Saint Croix Lake (WI DNR, 2012). Y=1.107x + 18.714, R^2=0.6861, N=58. Data collected and analyzed by WI DNR for DHS during the summer from 1995 through 2012.

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