Diatoms and chrysophyte cysts as indicators of past and current water quality conditions: a case study from White Lake, Eastern Ontario, Canada

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Abstract:

Paleolimnology can infer past conditions of water systems. Diatom analyses provide effective paleolimnological data that can be used to determine past and current water quality in freshwater bodies. Shifts in diatom and chrysophyte cyst assemblages provide indications of environmental changes, and can reveal anthropogenic impacts on water systems. These analysis techniques were applied to 16cm of a 20cm core sample obtained from White Lake in Eastern Ontario, Canada. The results infer changes that have occurred in the last approximately 150 years.

White Lake is a large, shallow lake with unique characteristics and a sensitive ecosystem. A shift encountered in the chrysophyte to diatom ratio and an increase in *Fragilaria pinnata* within the last five centimetres of the core sample is indicative of recent water quality changes within the lake. The variation in species abundance and loss of biodiversity encountered in the sediment sample over time is likely due to increasing anthropogenic nutrient additions to the system. These impacts are visible in the diatom-inferred decline of water quality revealed in this study.

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Introduction:

Freshwater Ecosystems

Freshwater ecosystems are among the most sensitive environments to anthropogenic disturbance on Earth. A vast array of external and internal factors contributes to shifts in water quality in freshwater ecosystems. Climate change, nutrient inputs and invasive macrophyte and animal species all contribute to complex ecological relationships in freshwater systems (FOCA, 2011). Anthropogenic activities have contributed to system transitions in Canada through time, particularly in the post-industrial era. Eastern Ontario lakes and rivers have experienced an increase of these environmental stressors, creating disturbances which result in both short and long-term environmental changes (Watchorn et al., 2007).

Under regular circumstances, the presence of algae plays an integral role in lake ecosystems, and can lead to positive gains for biodiversity (Cloern, 2010). The concern lies with anthropogenic additions of nutrients, primarily phosphorus (P) (often measured as total phosphorus; TP), to the system, which can greatly increase algal growth at an unsustainable rate. The current measurement for trophic conditions in Eastern Ontario lakes are as follows:

- Oligotrophic, TP 0-10ug/L
- Mesotrophic, TP 10-20 ug/L
- Eutrophic, TP above 20 ug/L (Environment Canada, 2013)

Eastern Ontario is currently forecasted to undergo higher rainfall volumes due to climate change within the next 50 years, leading to further addition of pollutants through runoff (McBean, 2012). This increased stormwater runoff, along with land-use development, sewage, septic tank failures, aquaculture discharge, fertilizers and cleaning products, eroded shorelines and agricultural practices all contribute to increasing TP concentrations in lakes and rivers throughout Ontario.

Paleolimnological Techniques

A variety of limnological techniques can be used to infer water quality conditions within water systems. Commonly secchi disk readings, pH and TP measurements are acquired to assess the current quality status within a waterbody. Secchi disk readings can identify water clarity at the time of administration. Chemical compositions can also infer quality variations through time. Currently, measuring TP within freshwater bodies is a definitive method in determining nutrient concentrations and thus trophic level that define water quality objectives. Though other nutrients can contribute significantly to macrophyte and algal growth (e.g. nitrogen) (Anderson et al., 2002), phosphorus is a key limiting nutrient in freshwater systems which is readily used for algal and macrophyte growth as it becomes available (Elser et al., 2007). That is, the more TP within a system, the more primary production will occur in the particular waterbody. As algal growth increases, so too does the decomposition process of plant matter. This decomposition results in the uptake of dissolved oxygen, and can lead to anoxic conditions within the system (Ansari et al., 2014). The procession of these conditions can lead to fish kills, and ultimately a shift in water quality and a reduction in biodiversity.

Phosphorus is naturally present in these systems, however anthropogenic inputs can increase the concentration from a variety of point and non-point sources (Canadian Council of Ministers of the Environment, 2004). Though TP and Secchi disk readings permit effective and relatively concise indications of water quality, often times these readings have not been administered historically in time scales necessary for long-term records (Fuller et al., 2004, Dixit et al., 1999).

Paleolimnology approaches permit research on past and present quality conditions in waterbodies. Paleolimnological data can infer a variety of historical occurrences through multiple means of examination. The implications of stressors like climate variations, invasive species and nutrient inputs on freshwater systems can be measured while having little to no previously collected water quality data (i.e. TP counts, secchi disk readings etc.) (Last and Smol, 2001). The collection of sediment cores permits past conditions to be interpreted by analyzing physical, chemical and biological indicators preserved within the sediment record (Alberta Environment, 2007). Textural and particle size analysis of sediment can infer conditions like runoff sources and rates, water turbidity, and erosion rates. Collection of mineralogical and magnetic orientation information can also generate interpretations on past water quality states (Last and Smol, 2001).

A number of biological methods can similarly be used for analysis. The identification of pollen and spores in sediment may be used to determine past climate and vegetation patterns, whereas zooplankton remains may aid in identifying past trophic conditions and acidity levels (Alberta Environment, 2007).

Diatom analysis is a well established paleolimnological technique used to reconstruct changes in water quality (Dixit et al., 1992). Diatoms are single celled algae that are present in virtually all water systems. Their sheer abundance and well defined environmental optima make them a dependable data resource across study sites. The preservation ability of their outer silicate shells permits the establishment of microfossils that are identifiable at the species level throughout the fossil record (Watkins, 2004). Diatom species are sensitive to environmental change, and have rapid response time to any shifts occurring within their environment. They appear in both planktonic and benthic habitats, and specific species are only present under particular water quality conditions. This makes diatoms an excellent indicator source, and by determining changes in assemblages through time, researchers can infer a range of historical environmental occurrences (Dixit et al., 1992). Implications in changes at an algal level are also representative of important shifts throughout an ecosystem, as they are primary producers and thus impact the entire food chain (Sudhakar et al., 1993).

Similarly, chrysophyte cysts are also widely distributed in freshwater environments and fossilize readily due to silicate outer shells (Adam and Mahood, 1981). Though cyst microfossils can be more challenging to assign at the species level, generalized genus counts can be used to infer water quality status (Duff et al., 1997). It is believed that chrysophyte cysts may actually be more sensitive to pH alterations within waterbodies than some diatom species, as cysts are planktonic and do not encounter the same sediment buffers that some benthic diatoms do (Cohen, 2003). Chrysophytes have a low tolerance towards eutrophic lake conditions, and can therefore be indicative of trophic states and water quality status as cyst to diatom ratio fluctuations are measured (Cohen, 2003).

Study Area: White Lake

These paleolimnologic techniques of interpreting diatom assemblages were transferred to a core sediment sample collected from White Lake in July 2014 to infer past and current water quality. Residents living by White Lake, Ontario are concerned about the current water quality state of the lake, which experienced a toxic blue-green algal bloom in 2014 (White Lake Preservation Project, 2015). It is a large, shallow waterbody with a unique composition, making it a sensitive environment susceptible to changes via land use and anthropogenic system additions (Discover White Lake, 2015).

White lake is located in Eastern Ontario. It is central to Greater Madawaska (west), Renfrew (northwest), Arnprior (northeast), Ottawa (East), and Perth (southeast) (Discover White Lake, 2015). The waterbody began as several individual basins that were flooded in 1850 by the White Lake dam. Since that time, approximately 400 cottages have been built on the lake, and surrounding cities have grown drastically in population size. This has increased pressures of pollutants and nutrients reaching the lake via runoff. The lake has 11 inlets that contribute to incoming pollutants from all directions, and its watershed ranges 211 square kilometers (White Lake Preservation Project, 2015).

<u>Methods:</u>

Field Sampling

A sediment core was taken from the southern end of White Lake, Latitude 45^{o} 16’ 4”, Longitude -76^{o} 33’ 6” using a gravity corer (Figure 1). Water depth was 18.3 ft., and the area (known as Boggs Island) contains sediment approximately six to nine metres in thickness which is likely underlain by Precambrian base rock (Ferris, 1985). The core sample studied contained approximately 20cm of sediment. Given the length of the core the sediment record likely extends to the last 150 years based on similar studies in the region. The sediment core was sliced and sectioned at 1 cm intervals in the field and placed into individual WhirlPak bags for transport back to the lab. Of the 20cm obtained core, 16cm were analyzed. Using a full core analysis approach allowed depiction of the visual progression of environmental changes through time. This permitted a more detailed timescale than a top-bottom method could achieve (Straub et al., 2006).

Figure 1 – White Lake Study Site

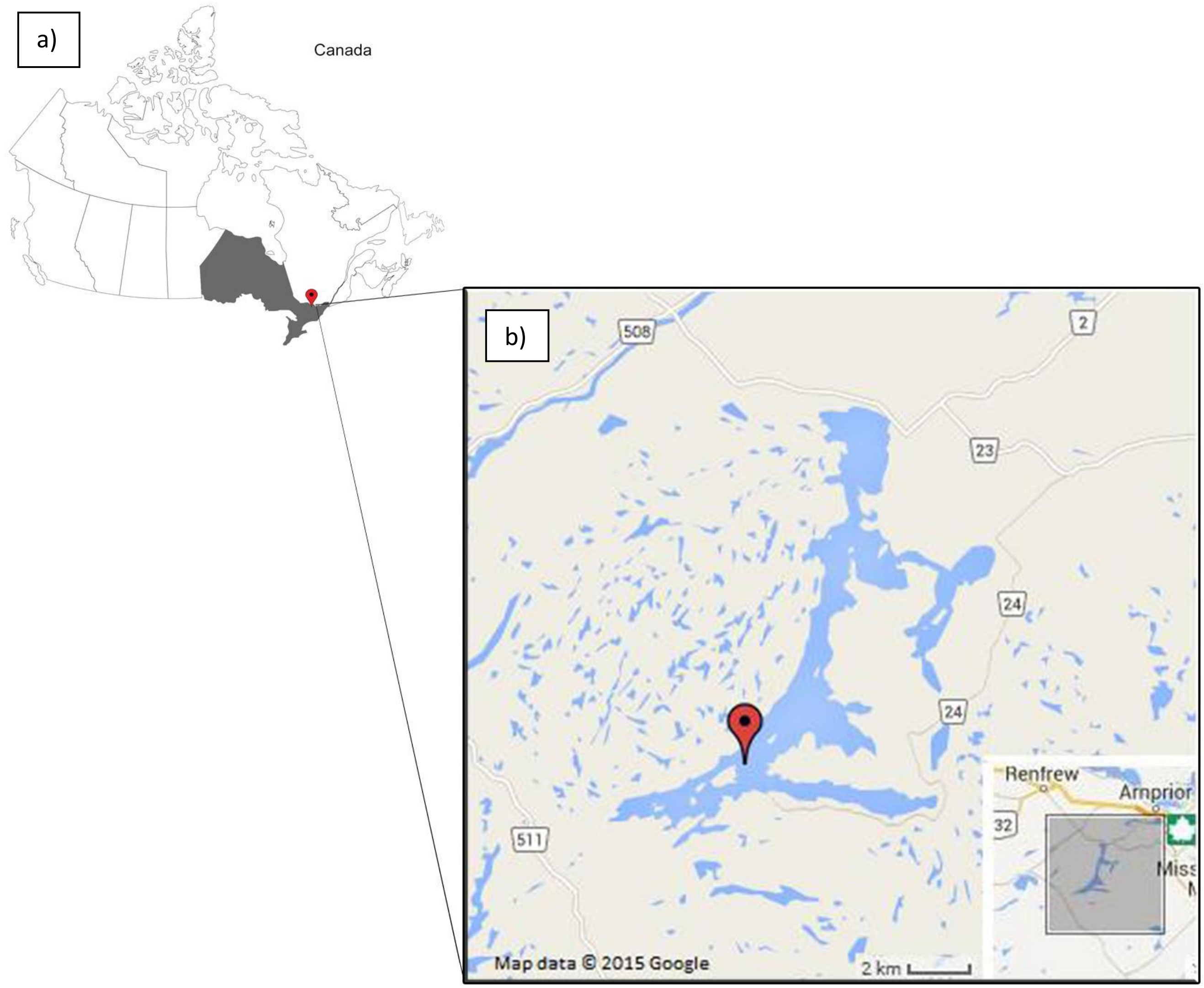


Figure 1: a) Map of Canada with Ontario highlighted and study area marked. b) Map of study area with sample site coordinates marked (Lat. 45° 16" 4', Long. 76° 33" 6'). (Google Maps, 2010)

Organic content

Organic content through time was measured by conducting a loss on ignition (LOI) evaluation. The procedure involved heating samples from each centimetre of the core. Sediment was weighed and heated to 105 degrees Celsius for a 24 hour period to remove water content. This resulted in total percent water calculation. Samples were then weighed again and subjected to heating at 550 degrees Celsius for four hours to burn off organic matter. Final weight difference determined percent organic matter.

Diatom Processing

All sample processing and analysis was done in adherence with the Algal Bioassessment Protocol (ABP) framework prepared by the Ontario Ministry of the Environment (2011). Preparing the diatoms for analysis consisted first of the removal of organic matter. A portion of each divided sample was isolated into test vials which were labelled appropriately by depth. They underwent an H_2O_2 wash for eight hours at 80 degrees Celsius. Once organic material was extracted, each vial received a series of washes over a five day period to thoroughly dilute the H_2O_2 content. The washes occurred once every 24 hours, and involved removing approximately half of the liquid content from each sample with a pipette without disturbing the bottom-settled diatoms (i.e. keeping the sample still). Vials were refilled with distilled water content during each rinsing event and one to two drops of a 10% HCl solution were added upon completion to ensure total emission of organic components. Each sample was sealed on the fifth day until cover slip mounting occurred approximately two weeks later.

Each coverslip was cleaned thoroughly with a 10% ethanol solution to prevent dust particles from drying on slides. Vials were shaken to mix diatoms randomly throughout the solution and samples were drawn promptly afterwards using a pipette. Extracted samples were placed into test tubes and filled to a previously determined point with distilled water. Each diatom slurry went through an additional mixing process using pipettes and the final sample for each cover slip was drawn in one uptake from test tubes. Half of each test tube was extracted and restored with distilled water, and the mixing and uptake process was repeated three more consecutive times until a total of four concentrations per centimetre sample were piped onto cover slips. Samples underwent an air drying process for 48 hours in covered cases. Cover slips were then permanently mounted on slides using naphrax (Acker et al., 2002).

Statistical analysis

Diatom analysis was carried out by identification at the species level. IDing occurred at 1000x magnification to ensure appropriate identifiable characteristics were discernible (Watkins, 2004). One slide concentration was selected from each analysed centimetre sample. Slides chosen for analysis were selected upon relative abundance per field of view; approximately 4-8 diatoms visible at 1000x magnification most frequently were used. Identification of each diatom was recorded at the species level. Diatom taxa were identified following Krammer and Lange-Bertalot visual characteristic models (2004, 2007, 2008a, 2008b).

For each slide, a total of minimum 300 diatom valves were counted. Number of chrysophyte cysts were also recorded up until minimum diatom count was reached. A total of 105 diatom species were identified throughout the core counts. This high volume of diversity is common in diatom assemblages, and is valuable in comparing abundance levels of each species and defining what they infer (Dixit et al., 1999). Species counts were entered into Microsoft Excel, and the relative abundance of each taxa was calculated. Rare species which never reached >2% relative abundance in the record were omitted from statistical analysis. This permitted a more focussed examination of the prominent species assemblages. The new total species count after this omission process was 23.

Data

Data for 23 species was entered in C2 1.7.6 (Juggins 2014). Plotted data depicts percent relative abundance per species per sample (Figure 2). Principal component analysis was created the statistical program R (R Development Core Team 2015) to emphasize variance and trends in the dataset.

Results:

The 23 common diatom taxa data were inputted into C2 to depict species abundance (%) per sample (cm). Results are displayed in Figure 2. Individual graphs are divided by species and display trends in abundance (%) through time. Visual results for a number of species are relatively consistent (Figure 2), however some species' abundance exhibit a marked change through time. The majority of species listed are small, benthic diatoms. *Fragilaria pinnata* in particular experiences a noticeable gain, increasing in abundance within the approximately top 5cm of sediment analyzed. An introduction of a new assemblage of *Fragilaria pinnata* var*. intercedens* occurs within the same sample depth. Within a similar timeframe, disappearance (i.e. *Achnanthese exigua, Navicula minima, Navicula pseudoventralis, Navicula pupula, Navicula recens*) or decrease (i.e. *Aulacoseira ambigua, Aulacoseira islandica)* of some species occurs. A mutilvariate regression tree (MRT) was used to further infer relationships of diatom assemblages through time. These results are also indicative of a distinguishable cluster relationship in the top five centimetres of the core sample (De'ath, 2002).

Principal Component Analysis (PCA) and Chrysophyte Cyst Data

A principle component analysis (Figure 3) was executed to identify key trends in the diatom assemblage species through time (Wang et al. 2009). PC1 axis reflects the major change in the diatom assemblage over time. Results show an increase of *Fragilaria pinnata* in recent years. Chrysophyte Cysts were not included in this component, but are represented in Figure 4, which depicts chrysophyte declination in relation to *Fragilaria pinnata* abundance increase throughout the core sample. Chrysophyte cyst abundance and PC1 axis scores are inversely related in a statistically significant correlation (R=0.001) as relayed in Figure 5.

Water Content

Water content loss results for 20cm sample remained relatively consistent through time. Samples maintain an approximate 95 percent water proportion over an approximately 150 year period that the core sample represents (Figure 6).

Organic Content

Organic content measured by LOI estimation is indicated in Figure 6. Resulting figures indicate a declining trend in organic matter within the top six centimetres. This decline also correlates with chrysophyte cyst abundance decline and PC1 axis results as shown in Figure 5.

Figure 2 – Abundance of 23 prominent diatom species per centimetre in sediment layer. MRT schematic emphasizes changes within the top five centimetres.

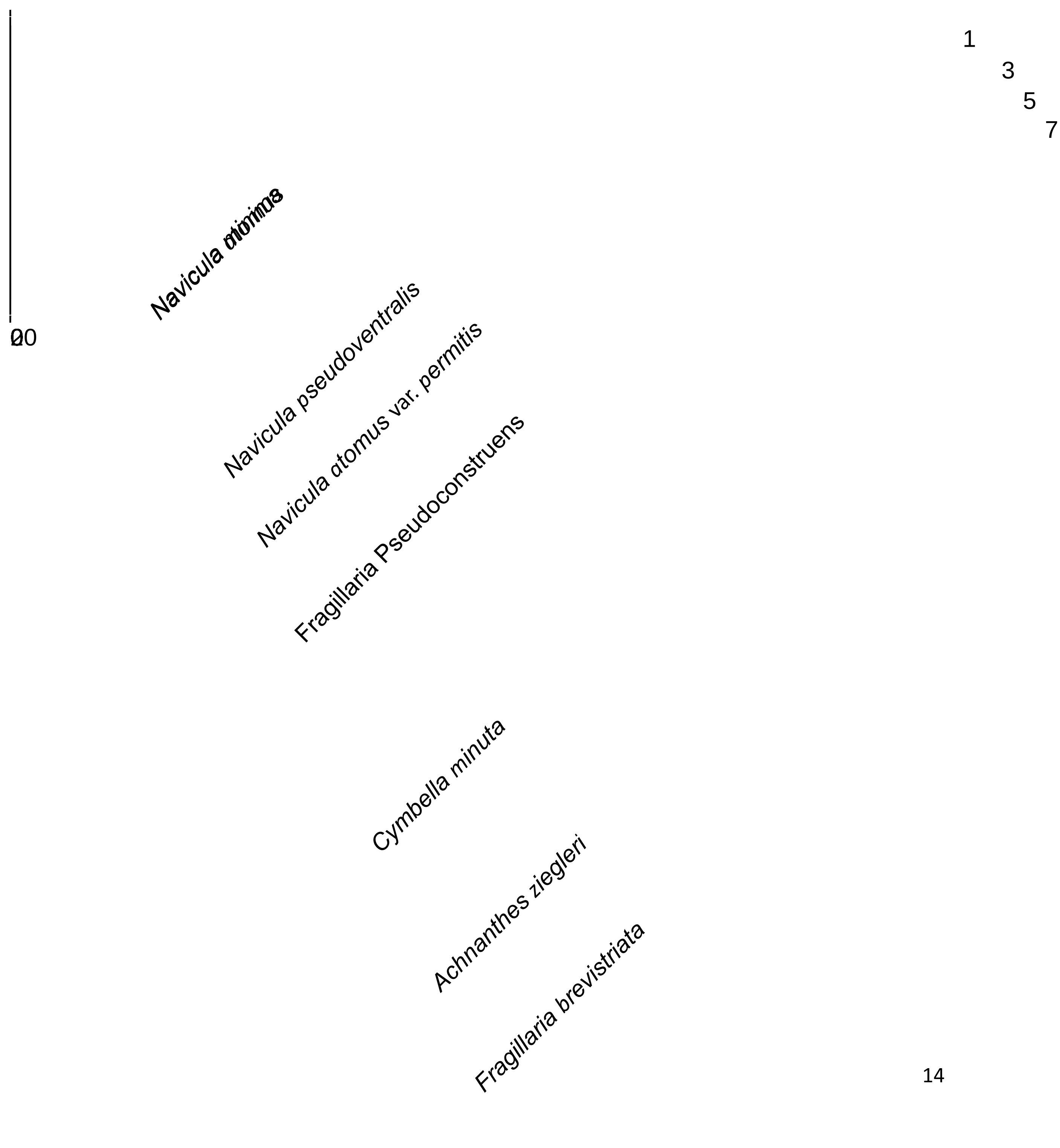


Figure 3 – PCA depicting the diatom assemblage variance through time. PC1 axis reflects the major change in the diatom assemblage over time.

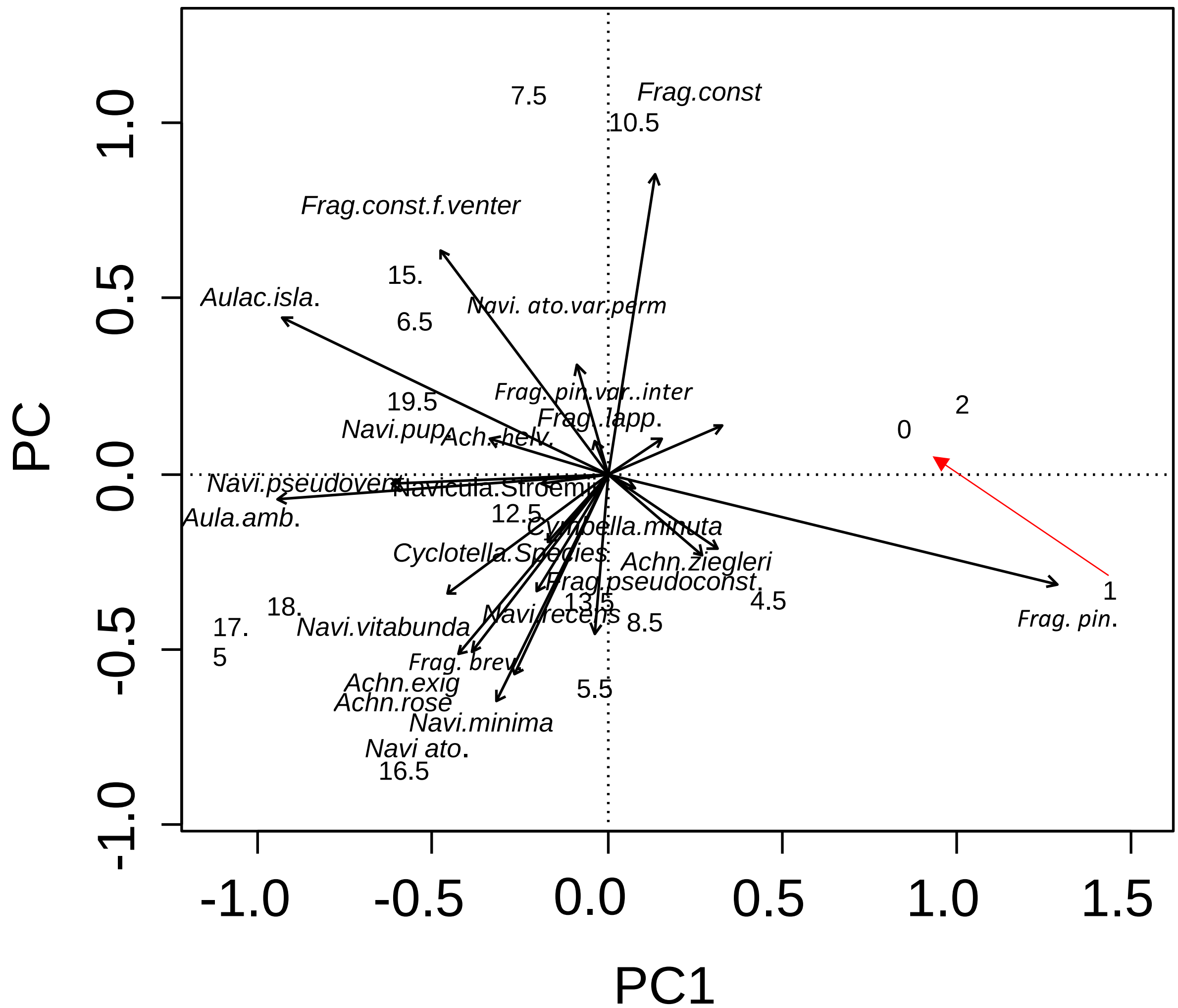


Figure 4 – Inverse relationship of PC1 axis and chrysophyte/diatom ratio indicating the increase in *Fragilaria pinnata* and decrease in cyst populations.

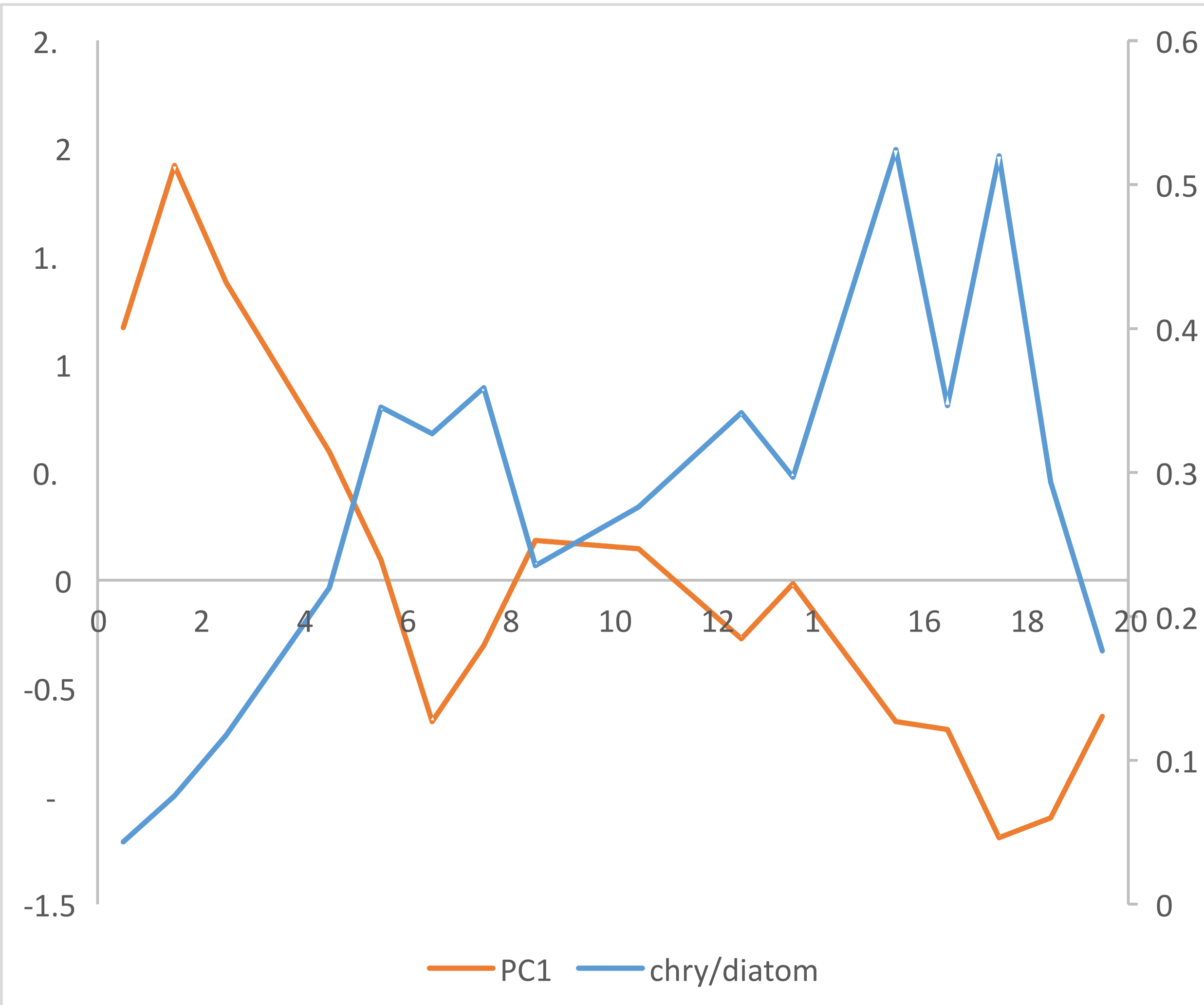


Figure 5 – Visual comparison of percent organic content, chrysophyte/diatom ratio, and PC1 axis. The correlation indicates changes occurring in the top five centimetres of the core sample.

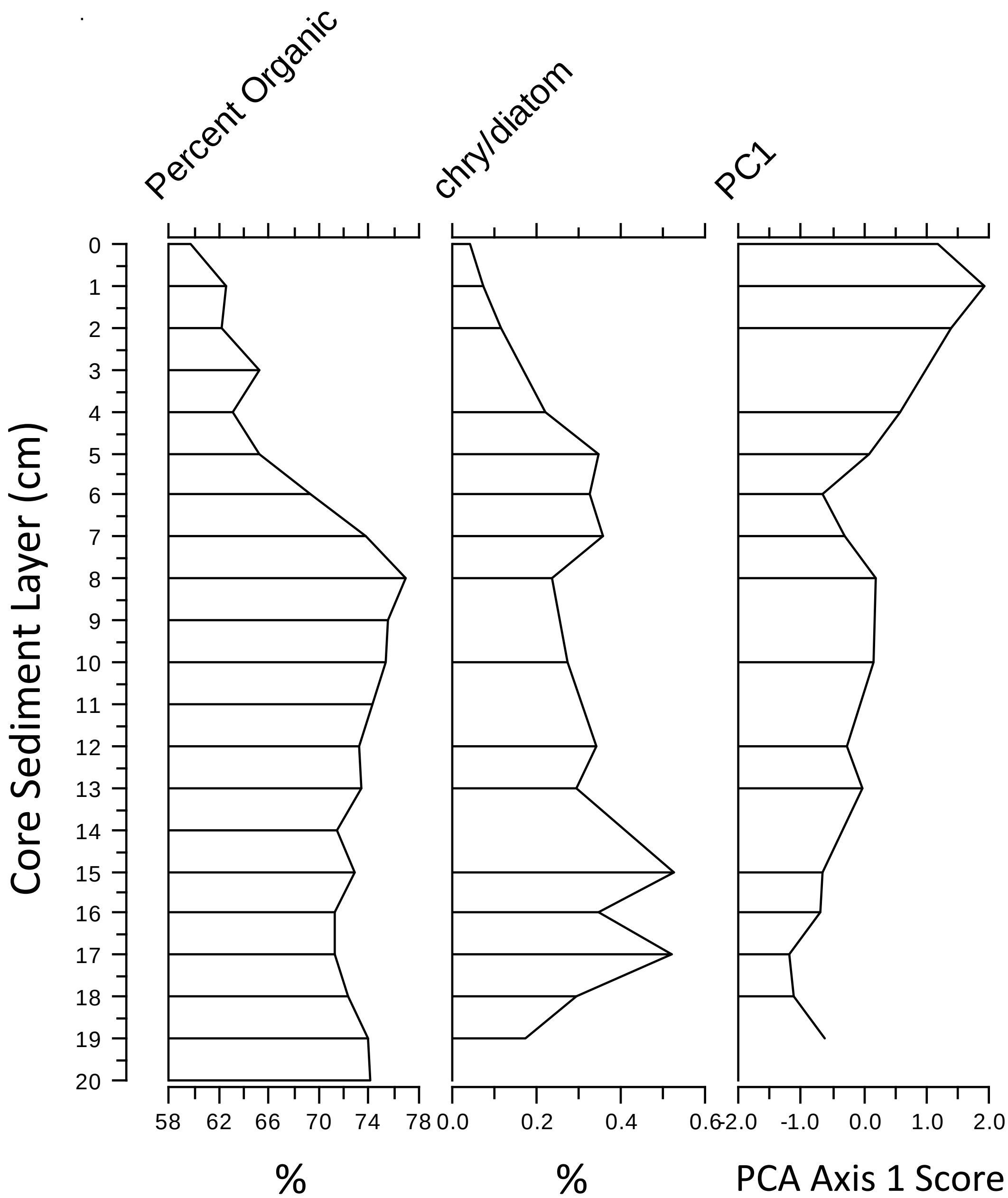
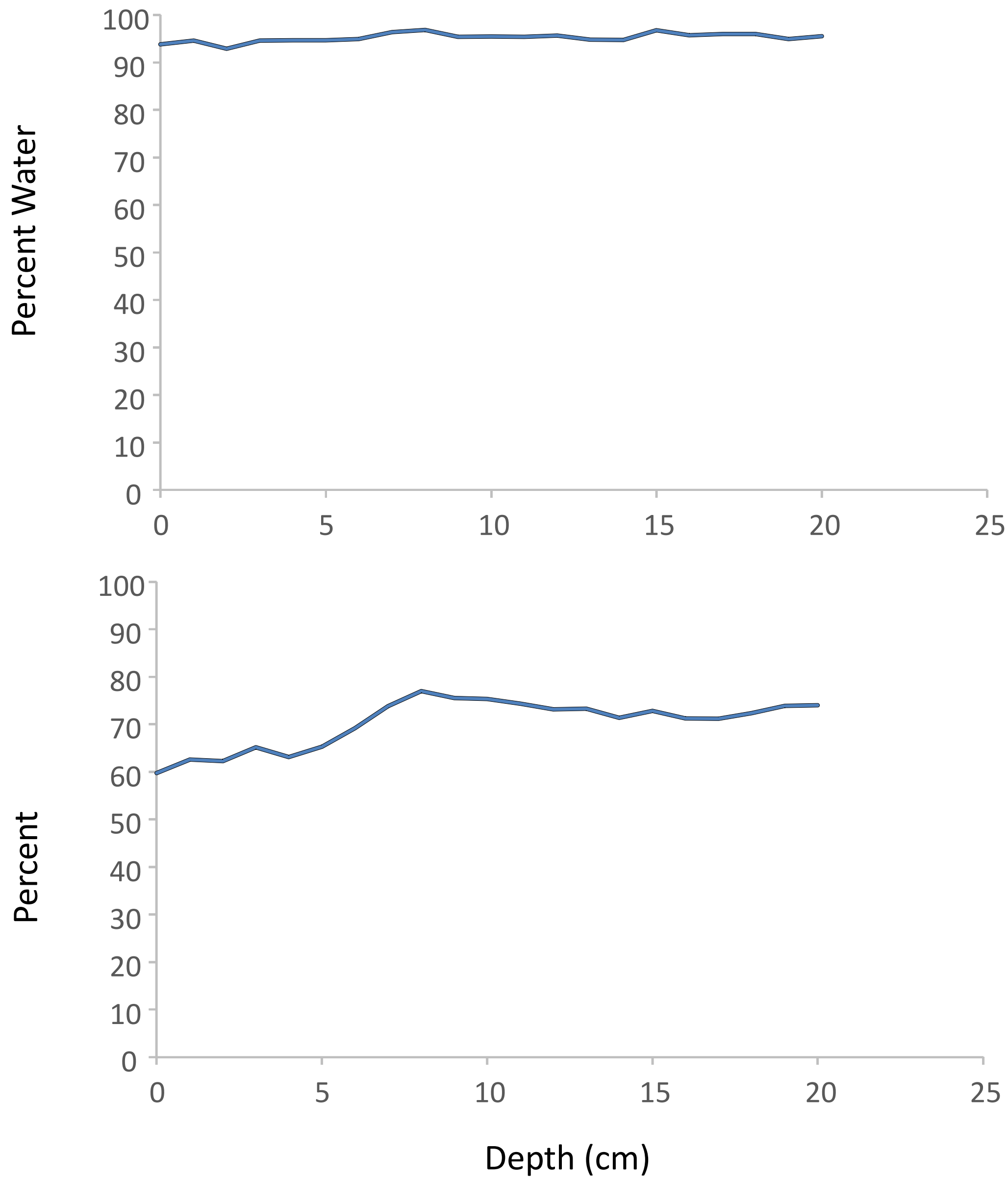


Figure 6 – Percent water and percent organic content per cm throughout the 20cm core.



Discussion

Shallow Lakes

White Lake is a large, shallow lake; its maximum depth is 12.8 meters, and mean depth is 3.1 meters. It is 16 kilometers long with a 98 kilometer perimeter, a 5,608 acre surface area and contains 74.74 million cubic meters water volume. Over 90% of the lake is classified as littoral zone, and is therefore biologically productive (White Lake Preservation Project, 2015). Shallower lakes have distinguishable characteristics, and often administer abundant macrophyte growth due to their high photic zone volume and lack of defined stratified layers (White Lake Preservation Project, 2015). They are also susceptible to sediment mixing through things like wind action and wake-boating activity (White Lake Preservation Project, 2015). All of these characteristics contribute to sensitive ecosystem conditions that are vulnerable to further anthropogenic inputs (Cohen, 2003).

White Lake Characteristics

Lake geology is an important characteristic necessary to interpret lake function. White Lake possesses a unique substrate, with surface geology that is intrinsically linked to lake processes and ecological function (White Lake Conservation Reserve, 2003). The southern section of the lake is composed of calcium rich limestone bedrock, while Precambrian rock constitutes the northern section (see Appendix A for greater detail) (White Lake Preservation Project, 2015). This provides the lake with a relatively dependable buffer for acidity shifts while maintaining calcium rates of approximately 30ppm (White Lake Preservation Project, 2015). Variance of conditions is reflected throughout the lake in TP counts, which changes throughout different regions. Data can be found at White Lake Preservation Project (2015).

The amount of available calcium and warm temperatures within White Lake makes it a potential opportune habitat for zebra mussels. Zebra mussels pose a particular threat to the waterbody, as they prefer relatively low pH levels and optimal temperatures for their adult life cycles is 20-25 degrees Celsius (USGS, 2015). Similarly, spiny water flea have a temperature preference of four to 30 degrees Celsius, and could therefore also be successful in this habitat (USGS, 2015). Most recent data collected by the region's Invasive Species Watch in 2013 indicates that neither of these invasive threats have been located in White Lake, however residents and lake goers should take extra precaution when moving from one waterbody to another to ensure a transfer does not occur (White Lake Preservation Project, 2015).

White Lake has been considered mildly eutrophic since its earliest published studies in 1974 (White Lake Preservation Project, 2015). A State of the Lake Report from 2007 measured mean secchi depth at 4.1 metres and mean euphotic zone TP at 12.8 µg/L (Mississippi Valley, 2007). Chlorophyll *a* quantity in the same report was recorded at 4.6 micrograms/litre, revealing high levels of algal presence (Mississippi Valley, 2007, White Lake Preservation Project, 2015). The lake is currently considered to be in a mesotrophic state, however resulting data from the analyzed core sample indicate a recent decline in water quality.

Diatom-inferred Water Quality Changes

Of 105 species identified in the 20cm core sample, 23 were deemed appropriately abundant. The top five centimetres, however, mark a statistically significant correlation of declining chrysophyte cyst to diatom ratios (R=0.001). As revealed in Figure 5, there is a notable visual correlation between cyst decline, percent organic content per centimeter decrease, and *Fragilaria pinnata* increase. This correlation is also evident in PCA axis 1 score and chrysophyte/diatom ratio (Figure 3).

As Figure 3 displays, significant decline in chrysophyte cysts also occurs. The decrease in water-quality-sensitive cyst species infers the reduction in water quality in White Lake in recent years (Adam and Mahood, 1981). The co-occurrence of these indicators infers diminishing water quality and the potential transition towards eutrophic conditions within the lake (Adam and Mahood, 1981, Dixit et al., 1999).

Small benthic diatoms such as those identified in the genus *Fragilaria* are common in shallow lakes (Wehr and Sheath, 2003). *Fragilaria pinnata* in particular are commonly found in mesotrophic waterbodies like White Lake's current trophic status, however the species is tolerant to environmental shifts and is common and adaptive in systems with declining water quality (Schmidt et al., 2004). *Fragilaria pinnata* can also be indicative of increased nutrient loading, and have a relatively high TP optima range of approximately 14 µg/L (Telford, 2001, Dixit et al., 1999). Appearance of *Fragilaria pinnata* var. *intercedens* also suggests increased nutrient inputs as their TP optima range is high, allowing them to be successful in eutrophic conditions (Dixit et al., 1999).

Similarly, a decrease in the two abundant planktonic diatom types (*Aulacoseira ambigua, Aulacoseira islandica)* prevalent in the counts experienced the same declining trend (see Figure 2). Though *Aulacoseira ambigua* and *Aulacoseira islandica* also maintain relatively high TP optima levels (Dixit et al., 1999, Poister et al., 2011), this may be due to lack of sediment buffer in suspended algae, causing it to be more susceptible to environmental condition changes (e.g. changes in pH) (Cohen, 2003). Increased turbidity caused by increasing sediment input may also inhibit growth for these highly photic-dependant species (Poister et al., 2011). Note that increasing sample size may also aid in determining significance of these changes.

Percent organic matter also decreases in correlation with PCA axis 1 score and chrysophyte/diatom ratio (Figure 5). Shift in organic content suggests increased erosion from greater nutrient-bound sediment input. Erosion via increased sediment is also inferred by escalation of *Fragilaria pinnata* totals, a species which is known to reside in turbid waters due to their adaptive nature (Smol and Stoermer, 2010). They are also tolerant to greater stress from current flows (Wang et al., 2009). Increased sediment input is associated with land clearing increases, anthropogenic system additions and increased flow from input sources (i.e. higher rainfall volumes) (Whitmore and Riedinger, 2002). Increasing input flows via heavier rainfall are expected to accelerate over the next 50 years in Eastern Ontario, making White Lake particularly vulnerable to an increase in these conditions (McBean, 2012).

Recent TP counts measured in 2006 from Pickerel Bay and Three Mile Bay may also be indicative of increased turbidity due to sediment mixing; >30 TP µg/L measurements were determined and are likely due in part to impacts caused by wake-boating. Motors easily stir sediment and are therefore particularly consequential for shallow lakes (White Lake Preservation Project, 2015). Discoveries of anoxic zones within White Lake in 2014 promote further nutrient addition; as oxygen levels decline, increased sediment mixing permits P otherwise trapped in bottom sediment to be released (White Lake Preservation Project, 2015). The flushing rate in the lake is 0.89 times/year, contributing to the sensitive nature of the lake, as it provides a means of greater sediment accumulation over time (White Lake Preservation Project, 2015). Further nutrient sources present in the system can lead to excessive macrophyte and algal growth, which can ultimately lead to increasing anoxic conditions (Anderson et al., 2002). Anthropogenic activity should therefore be monitored to ensure minimal nutrient addition in order to prevent the lake from reaching an accelerated eutrophic state.

Overall fluctuations in diversity also indicate conditional changes. Loss of biodiversity seen in the top five centimetres of the core sample (Figure 2, Figure 3) is indicative of diminishing water quality (GreenFacts, 2015). Ultimately, there is a diatom-inferred decline in water quality throughout this time period. These observed changes in the top five centimetres also suggest nutrient enrichment and increased sediment input in recent years. The progression of these nutrient additions with no preventative actions taken may lead the lake to accelerated eutrophic conditions (Anderson et al., 2002).

Conclusion:

Paleolimnologic diatom analysis was used to infer current and historic water quality changes within White Lake, Ontario. There was a statistically significant correlation between increasing *Fragilaria pinnata* species (PC1 axis) and declining chrysophyte cyst populations within the top five centimetres of the 20cm sediment core studied from White Lake (Figure 5). The decline in percent organic matter also correlates with these figures (Figure 5, 6). These figures infer changes occurring within the lake which are indicative of declining water quality.

The recent declination is likely due to increased land use and anthropogenic pollutant additions, though more information on lake input via runoff is necessary to formulate conclusive reasoning. Diatom inferred changes within the sediment core reveal the sensitive nature of White Lake, and the importance of maintaining its quality to support its diverse and unique ecosystem. Residents may aid in preservation efforts by adhering to wake-boating regulations to diminish sediment mixing, by decreasing nutrient inputs and by maintaining naturalized shorelines that act as critical buffer zones for harmful pollutants (FOCA, 2011).

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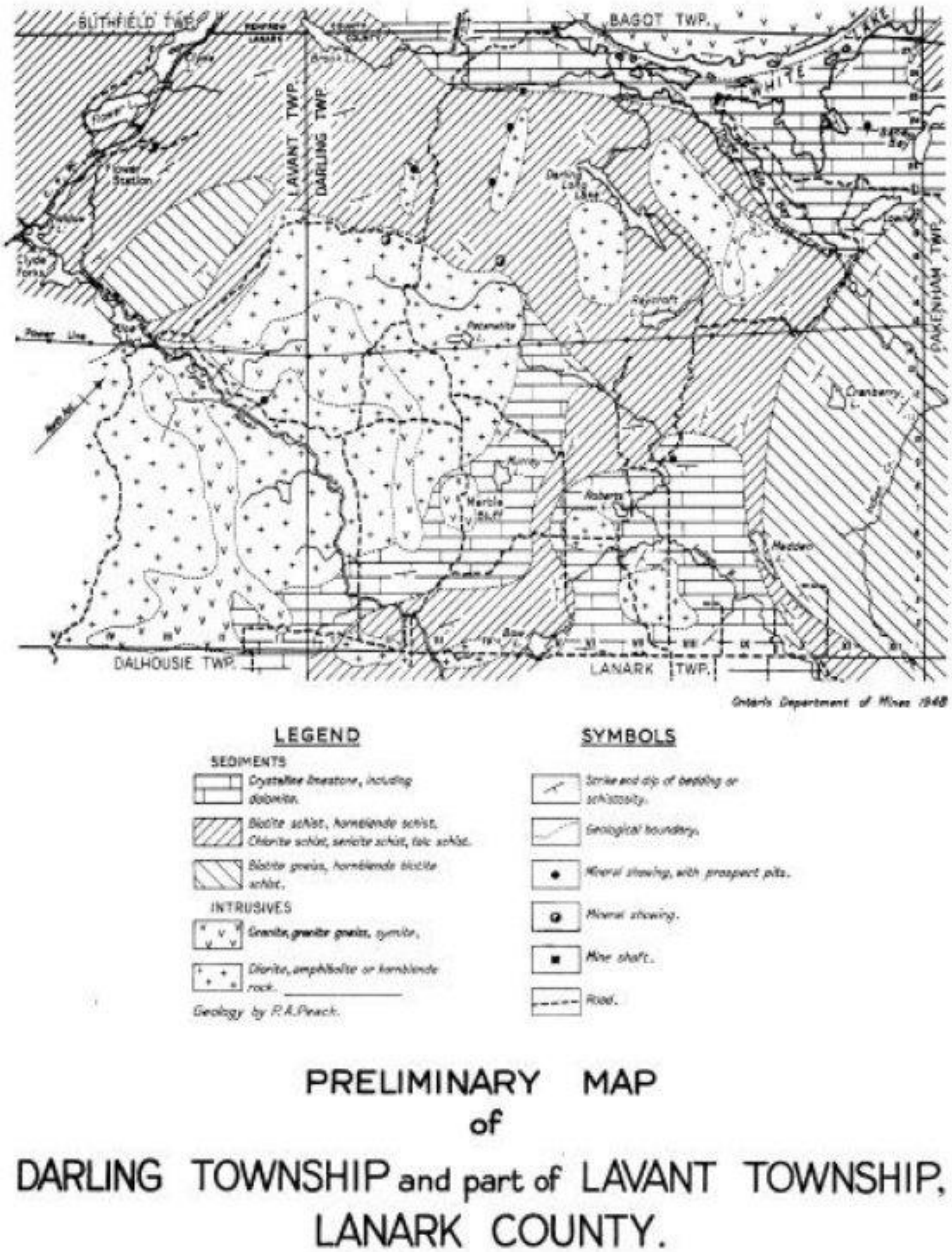
Appendix A



Source: “Geological Setting of White Lake”, White Lake Preservation Project (2015).

Appendix B – Raw data included in hard-copy format.